

CHEMICAL CONSTITUENTS OF THE LEAVES OF *Michelia figo*

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Species belonging to the genus *Michelia* are arboreous plants, growing in temperate zones of oriental India, southern China, Malaysia, and Indonesia. The species most utilized is *Michelia champaca*: its cortex and seeds are used as febrifuge and tonic-aromatic; the roots are employed as emmenagogue, the leaves as astringent, the gemmae in the treatment of hemorrhage, and the flowers and fruits are believed to possess curative properties in enteritis [1]. The less known species, *M. figo*, is used as ornamental plants and to obtain essences [1]. *M. figo* is an evergreen medium shrub, commonly called banana shrub, because of the heavy, sweet fragrant banana scent of its purple flowers. The plant is also known in Indian folk medicine as a remedy against hypertension [2]. To further understand the chemotaxonomy of the *Michelia* species [3–7], *M. figo* was chosen for phytochemical investigation. The chemical constituents of the leaves of this plant have not yet been reported. The compounds derived from the leaves include three alkaloids, (–)-nuciferine (**1**) [8], (–)-anonaine (**2**) [9], and *N*-methylcorydaldine (**3**) [10]; two steroids, β -sitosterone (**4**) [9] and stigmasta-4,22-dien-3-one (**5**) [9]; four benzenoids, *p*-hydroxybenzaldehyde (**6**) [10], *p*-hydroxybenzoic acid (**7**) [11], methylparaben (**8**) [11], and vanillin (**9**) [11]; six chlorophylls, pheophytin a (**10**) [12], pheophorbide a (**11**) [12], pheophytin b (**12**) [13], pheophorbide b (**13**) [13], aristophyll-C (**14**) [14], 13²-hydroxy-(13²-*S*)-pheophytin a (**15**) [15]; and one sesquiterpene lactone, 11,13-dehydrolanuginolide (**16**) [16]. All of these known compounds were obtained for the first time from the leaves of this plant and were identified by direct comparison with authentic samples (TLC, UV, IR, ESI-MS and NMR) and the literature [8–16].

The leaves of *M. figo* (Lour.) Spreng. were collected from Chiayi County, Taiwan, May 2011. Plant material was identified by Prof. Fu-Yuan Lu (Department of Forestry and Natural Resources, College of Agriculture, National Chiayi University). A voucher specimen (*Michelia* 5) was deposited in the School of Medicinal and Health Sciences, Fooyin University, Kaohsiung City, Taiwan. The air-dried leaves of *M. figo* (4.8 kg) were extracted with MeOH (6 L \times 4) at room temperature, and a MeOH extract (121.6 g) was obtained upon concentration under reduced pressure. The MeOH extract, suspended in H₂O (1 L), was partitioned with CH₂Cl₂ (3 L \times 5) to give fractions soluble in CH₂Cl₂ (67.9 g) and H₂O. The CH₂Cl₂-soluble fraction was chromatographed over silica gel (950 g, 70–230 mesh) using *n*-hexane–EtOAc–MeOH mixtures as eluents to give five fractions. Part of fraction 1 (8.24 g) was subjected to silica gel chromatography by eluting with *n*-hexane–EtOAc (60:1) and enriched gradually with EtOAc to furnish five fractions (1-1–1-5). Fraction 1-2 (3.11 g) was further purified on a silica gel column using *n*-hexane–EtOAc mixtures to obtain **4** (12 mg) and **5** (6 mg). Part of fraction 2 (11.78 g) was subjected to silica gel chromatography by eluting with *n*-hexane–EtOAc (60:1) and enriched gradually with EtOAc to furnish five fractions (2-1–2-5). Fraction 2-1 (2.78 g) was further purified on a silica gel column using *n*-hexane–EtOAc mixtures to obtain **10** (3 mg) and **11** (4 mg). Fraction 2-2 (2.17 g) was further purified on a silica gel column using *n*-hexane–EtOAc mixtures to obtain **12** (1 mg) and **13** (2 mg). Fraction 2-3 (2.86 g) was further purified on a silica gel column using *n*-hexane–EtOAc mixtures to obtain **14** (13 mg) and **15** (9 mg). Fraction 2-4 (1.55 g) was further purified on a silica gel column using *n*-hexane–EtOAc mixtures to obtain **16** (18 mg). Part of fraction 3 (16.97 g) was subjected to silica gel chromatography by eluting with *n*-hexane–EtOAc (40:1) and enriched with EtOAc to furnish six further fractions (3-1–3-6). Fraction 3-3 (4.24 g) was further purified on a silica

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gel column using CH_2Cl_2 -MeOH mixtures to obtain **3** (7 mg). Fraction 3-4 (3.24 g) was further purified on a silica gel column using CH_2Cl_2 -MeOH mixtures to obtain **2** (19 mg) and (-)-nuciferine (2 mg). Part of fraction 4 (21.23 g) was subjected to silica gel chromatography by eluting with CH_2Cl_2 -MeOH (40:1) and enriched with MeOH to furnish four fractions (4-1-4-4). Fraction 4-2 (16.72 g) eluted with CH_2Cl_2 -MeOH (40:1) was further separated using silica gel column chromatography and preparative TLC (CH_2Cl_2 -MeOH (50:1)) to give **6** (17 mg), **7** (22 mg), and **8** (13 mg). Part of fraction 5 (16.12 g) was subjected to silica gel chromatography by eluting with CH_2Cl_2 -MeOH (40:1) and enriched with MeOH to furnish four fractions (5-1-5-4). Fraction 5-3 (5.31 g) eluted with CH_2Cl_2 -MeOH (40:1) was further separated using silica gel column chromatography and preparative TLC (CH_2Cl_2 -MeOH (45:1)) to give **9** (14 mg).

(-)-Nuciferine (1). $\text{C}_{19}\text{H}_{21}\text{NO}_2$, brown powder (MeOH), mp 164–166°C. UV (λ_{max} , nm): 230, 274, 312. IR (ν_{max} , cm^{-1}): 1250, 1375, 1425, 1500, 1605. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 2.71–3.15 (4H, m, H-4, 5), 3.65 (3H, s, 1-OCH₃), 3.88 (3H, s, 2-OCH₃), 6.62 (1H, s, H-3), 7.23–7.26 (3H, m, H-8, 9, 10), 8.35 (1H, d, J = 7.6, H-11). ESI-MS m/z 319 $[\text{M} + \text{Na} + \text{H}]^+$ [8].

(-)-Anonaine (2). $\text{C}_{17}\text{H}_{15}\text{NO}_2$, pale yellow powder (MeOH), mp 121–123°C. UV (λ_{max} , nm): 230, 272, 310. IR (ν_{max} , cm^{-1}): 950, 1040. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 2.65 (1H, t, J = 13.4, H-7a), 2.85 (1H, dd, J = 13.4, 5.2, H-7b), 3.11–3.29 (3H, m, H-4a, 4b, 5a), 3.53 (1H, m, H-5b), 3.98 (1H, dd, J = 13.4, 5.2, H-6a), 5.92, 6.06 (each 1H, d, J = 1.6, OCH₂O), 6.55 (1H, s, H-3), 7.21–7.30 (3H, m, H-8, 9, 10), 8.06 (1H, d, J = 7.6, H-11). ESI-MS m/z 289 $[\text{M} + \text{Na} + \text{H}]^+$ [9].

N-Methylcorydaldine (3). $\text{C}_{11}\text{H}_{15}\text{NO}_3$, colorless crystals (CHCl_3), mp 112–113°C. UV (λ_{max} , nm): 200, 219, 263, 303. IR (ν_{max} , cm^{-1}): 1612, 1656. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 2.93 (2H, t, J = 6.8, H-4), 3.12 (3H, s, N-CH₃), 3.56 (2H, t, J = 6.8, H-3), 3.92 (3H, s, 6-OCH₃), 3.93 (3H, s, 7-OCH₃), 6.58 (1H, s, H-5), 7.41 (1H, s, H-8). ESI-MS m/z 245 $[\text{M} + \text{Na} + \text{H}]^+$ [10].

β -Sitostenone (4). $\text{C}_{29}\text{H}_{48}\text{O}$, white needles (CHCl_3), mp 85–86°C. IR (ν_{max} , cm^{-1}): 1375, 1385, 1460, 1620, 1675. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 0.68 (3H, s, H-18), 0.81 (3H, d, J = 6.7, H-26), 0.84 (3H, s, H-27), 0.86 (3H, t, J = 7.1, H-29), 0.92 (3H, d, J = 6.0, H-21), 1.02 (3H, s, H-19), 5.72 (1H, d, J = 1.4, H-3). EI-MS m/z 414 $[\text{M}]^+$ [9].

Stigmasta-4,22-dien-3-one (5). $\text{C}_{29}\text{H}_{46}\text{O}$, white needles (CHCl_3), mp 135–136°C. IR (ν_{max} , cm^{-1}): 1375, 1385, 1460, 1620, 1675. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 0.68 (3H, s, H-18), 0.81 (3H, d, J = 6.7, H-26), 0.84 (3H, s, H-27), 0.86 (3H, t, J = 7.1, H-29), 0.93 (3H, d, J = 6.0, H-21), 1.02 (3H, s, H-19), 5.02 (1H, dd, J = 16.1, 8.3, H-22), 5.12 (1H, dd, J = 16.1, 8.3, H-23), 5.72 (1H, d, J = 1.4, H-3). EI-MS m/z 412 $[\text{M}]^+$ [9].

p-Hydroxybenzaldehyde (6). $\text{C}_7\text{H}_6\text{O}_2$, brown powder (CHCl_3). UV (λ_{max} , nm): 223, 285, 290. IR (ν_{max} , cm^{-1}): 3200, 1660, 1600, 1155, 826. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 6.92 (2H, d, J = 8.4, H-3, 5), 7.80 (2H, d, J = 8.4, H-2, 6), 9.87 (1H, s, CHO). ESI-MS m/z 146 $[\text{M} + \text{Na} + \text{H}]^+$ [10].

p-Hydroxybenzoic Acid (7). $\text{C}_7\text{H}_6\text{O}_3$, brown powder (CHCl_3). UV (λ_{max} , nm): 250, 285, 290. IR (ν_{max} , cm^{-1}): 3500, 1660, 1590, 1165, 845. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 6.85 (2H, d, J = 8.6, H-3, 5), 7.96 (2H, d, J = 8.6, H-2, 6). ESI-MS m/z 162 $[\text{M} + \text{Na} + \text{H}]^+$ [11].

Methylparaben (8). $\text{C}_8\text{H}_8\text{O}_3$, colorless needles (CHCl_3), mp 130–131°C. UV (λ_{max} , nm): 225, 256, 310. IR (ν_{max} , cm^{-1}): 3400, 2950, 1695, 1610. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 3.88 (3H, s, COOCH₃), 5.46 (1H, br.s, OH), 6.87 (2H, d, J = 8.8, H-3, 5), 7.96 (2H, d, J = 8.8, H-2, 6). ESI-MS m/z 176 $[\text{M} + \text{Na} + \text{H}]^+$ [11].

Vanillin (9). $\text{C}_8\text{H}_8\text{O}_3$, yellow powder (CHCl_3). UV (λ_{max} , nm): 220, 280, 310. IR (ν_{max} , cm^{-1}): 3400, 1670, 1595, 1025. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): 3.95 (3H, s, 3-OCH₃), 6.20 (1H, br.s, OH), 7.09 (1H, d, J = 8.0, H-5), 7.31 (1H, d, J = 2.0, H-2), 7.42 (1H, dd, J = 8.0, 2.0, H-6), 9.77 (1H, s, CHO). ESI-MS m/z 176 $[\text{M} + \text{Na} + \text{H}]^+$ [11].

Pheophytin a (10). $\text{C}_{55}\text{H}_{74}\text{N}_4\text{O}_5$, deep green needles (CHCl_3), mp 113–114°C. UV (λ_{max} , nm): 229, 274, 330, 372, 406, 508, 540, 610, 665. IR (ν_{max} , cm^{-1}): 3400, 1740, 1700, 1620. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): -1.62 (1H, br.s, NH, D₂O exchangeable), 0.81, 0.83 (each 3H, d, J = 6.6, H-38, 39), 0.87 (6H, d, J = 6.6, H-36, 37), 1.60 (3H, s, H-40), 1.01–1.10 (21H, m, H-24–35), 1.64 (3H, t, J = 7.6, H-8²), 2.22 (1H, m), 2.32 (1H, m), 2.54 (1H, m), 2.65 (1H, m), 3.22, 3.42 (each 3H, s, H-7¹, 2¹), 3.54 (2H, q, J = 7.6, H-8¹), 3.72, 3.88 (each 3H, s, H-12¹, OCH₃), 4.22 (1H, m, H-17), 4.48 (2H, m, H-18), 4.49 (1H, d, J = 7.2, H-21), 5.16 (1H, t, J = 7.4, H-22), 6.19 (1H, d, J = 11.6, H-3²), 6.30 (1H, d, J = 17.8, H-3²), 6.28 (1H, s, H-13²), 8.00 (1H, dd, J = 17.8, 11.4, H-3¹), 8.57, 9.39, 9.52 (each 1H, s, H-20, 5, 10). FAB-MS m/z 871 $[\text{M} + \text{H}]^+$ [12].

Pheophorbide a (11). $\text{C}_{35}\text{H}_{36}\text{N}_4\text{O}_5$, deep green needles (CHCl_3), mp 115–116°C. UV (λ_{max} , nm): 229, 274, 330, 372, 408, 506, 536, 608, 665. IR (ν_{max} , cm^{-1}): 3400, 1740, 1700, 1620. ^1H NMR (400 MHz, CDCl_3 , δ , ppm, J/Hz): -1.63 (1H, br.s, NH, D₂O exchangeable), 1.64 (3H, t, J = 7.6, H-8²), 2.22 (1H, m), 2.35 (1H, m), 2.50 (1H, m), 2.65 (1H, m), 3.22, 3.40 (each 3H, s, H-7¹, 2¹), 3.52 (2H, q, J = 7.6, H-8¹), 3.70, 3.88 (each 3H, s, H-12¹, OCH₃), 4.22 (1H, m, H-17), 4.47 (2H, m, H-18),

6.17 (1H, d, J = 11.4, H-3²), 6.28 (1H, s, H-13²), 6.30 (1H, d, J = 17.8, H-3²), 8.00 (1H, dd, J = 17.8, 11.4, H-3¹), 8.57, 9.39, 9.52 (each 1H, s, H-20, 5, 10). FAB-MS *m/z* 593 [M + H]⁺ [12].

Pheophytin b (12). C₅₅H₇₂N₄O₆, deep green needles (CHCl₃), mp 118–119°C. UV (λ_{\max} , nm): 233, 280, 330, 411, 435, 536, 608, 665. IR (ν_{\max} , cm⁻¹): 3500, 1730, 1700, 1665, 1616. ¹H NMR (400 MHz, CDCl₃, δ , ppm, J/Hz): -1.49 (1H, br.s, NH, D₂O exchangeable), 0.77 (3H, d, J = 6.4, H-38), 0.78 (3H, d, J = 6.4, H-39), 0.83 (6H, d, J = 6.6, H-36, 37), 1.58 (3H, s, H-40), 1.00–1.90 (21H, m, H-24–35), 1.63 (3H, t, J = 7.6, H-8²), 2.21 (1H, m), 2.35 (1H, m), 2.50 (1H, m), 2.64 (1H, m), 3.38 (3H, s, H-2¹), 3.69 (3H, s, H-7¹), 3.88 (2H, q, J = 7.6, H-8¹), 3.90 (3H, s, OCH₃), 4.07 (1H, m, H-17), 4.45 (2H, m, H-18), 4.53 (2H, m, H-21), 5.14 (1H, t, J = 7.2, H-22), 6.21 (1H, d, J = 11.6, H-3²), 6.23 (1H, s, H-13²), 6.37 (1H, d, J = 17.8, H-3²), 8.01 (1H, dd, J = 17.8, 11.6, H-3¹), 8.54, 9.66, 10.38 (each 1H, s, H-20, 5, 10), 11.01 (1H, s, CHO). FAB-MS *m/z* 885 [M + H]⁺ [13].

Pheophorbide b (13). C₃₅H₃₄N₄O₆, deep green needles (CHCl₃), mp 118–119°C. UV (λ_{\max} , nm): 233, 280, 330, 411, 435, 536, 608, 665. IR (ν_{\max} , cm⁻¹): 3500, 1730, 1700, 1665, 1616. ¹H NMR (400 MHz, CDCl₃, δ , ppm, J/Hz): -1.49 (1H, br.s, NH, D₂O exchangeable), 1.63 (3H, t, J = 7.6, H-8²), 2.21 (1H, m), 2.35 (1H, m), 2.50 (1H, m), 2.64 (1H, m), 3.38 (3H, s, H-2¹), 3.69 (3H, s, H-7¹), 3.88 (2H, q, J = 7.6, H-8¹), 3.90 (3H, s, OCH₃), 4.07 (1H, m, H-17), 4.45 (2H, m, H-18), 6.21 (1H, d, J = 11.6, H-3²), 6.23 (1H, s, H-13²), 6.37 (1H, d, J = 17.8, H-3²), 8.01 (1H, dd, J = 17.8, 11.6, H-3¹), 8.54, 9.66, 10.38 (each 1H, s, H-20, 5, 10), 11.01 (1H, s, CHO). FAB-MS *m/z* 607 [M + H]⁺ [13].

Aristophyll-C (14). C₅₃H₇₀N₄O₅, deep green needles (CHCl₃), mp 247–248°C. UV (λ_{\max} , nm): 282, 360, 412, 480, 512, 550, 642, 702. IR (ν_{\max} , cm⁻¹): 1740, 1725. ¹H NMR (400 MHz, CDCl₃, δ , ppm, J/Hz): -0.18, 0.12 (each 1H, br.s, NH, D₂O exchangeable), 0.78, 0.80 (each 3H, d, J = 7.0, H-38, 39), 0.83 (6H, d, J = 6.8, H-36, 37), 1.01–1.62 (21H, m), 1.62 (3H, s, H-40), 1.66 (3H, t, J = 7.6, H-8²), 1.75 (3H, d, J = 7.0, H-18¹), 2.04 (1H, m, H-17¹), 2.45 (2H, m, H-17¹, 17²), 2.73 (1H, m, H-17²), 3.15 (3H, s, CH₃-7), 3.37 (3H, s, CH₃-2), 3.62 (2H, q, J = 7.6, H-8¹), 3.75 (3H, s, CH₃-12), 4.36 (1H, q, J = 7.0, H-18), 4.52 (2H, m, H-21), 5.22 (2H, m, H-17, 22), 6.22 (1H, d, J = 11.4, H-3²), 6.30 (1H, d, J = 18.0, H-3²), 7.89 (1H, dd, J = 18.0, 11.5, H-3¹), 8.56, 9.40, 9.56 (each 1H, s, H-20, 5, 10). FAB-MS *m/z* 843 [M + H]⁺ [14].

13²-Hydroxy-(13²-S)-pheophytin a (15). C₅₅H₇₄N₄O₆, deep green needles (CHCl₃), mp 205–206°C. UV (λ_{\max} , nm): 225, 410, 505, 611, 665. IR (ν_{\max} , cm⁻¹): 3400, 1740, 1700, 1620. ¹H NMR (400 MHz, CDCl₃, δ , ppm, J/Hz): -1.82 (1H, br.s, NH, D₂O exchangeable), 0.77, 0.78 (each 3H, d, J = 6.4, H-38, 39), 0.83 (3H, d, J = 6.4, H-36), 0.86 (3H, d, J = 6.4, H-37), 1.60 (3H, s, H-40), 1.62 (3H, d, J = 7.4, H-18¹), 2.21–2.34 (2H, m), 2.56 (1H, m), 2.93 (1H, m), 3.27, 3.43 (each 3H, s, H-7¹, 2¹), 3.61 (3H, s, H-12¹, OCH₃), 3.75 (3H, s, H-13⁴, OCH₃), 4.16 (1H, m, H-17), 4.52 (2H, m, H-18), 4.57 (1H, d, J = 7.2, H-21), 5.20 (1H, t, J = 7.2, H-22), 6.20 (1H, d, J = 11.6, H-3²), 6.30 (1H, d, J = 17.8, H-3²), 8.03 (1H, dd, J = 17.8, 11.6, H-3¹), 8.64, 9.50, 9.63 (each 1H, s, H-20, 5, 10). FAB-MS *m/z* 887 [M + H]⁺ [15].

11,13-Dehydrolanuginolide (16). C₁₇H₂₂O₅, colorless needles (MeOH), mp 168–170°C. UV (λ_{\max} , nm): 210. IR (ν_{\max} , cm⁻¹): 1770, 1655. ¹H NMR (400 MHz, CDCl₃, δ , ppm, J/Hz): 1.29 (3H, s, H-15), 1.84 (3H, s, H-14), 3.30 (1H, dd, J = 7.2, 3.6, H-7), 4.32 (1H, dd, J = 9.2, 6.8, H-6), 4.60 (1H, m, H-8), 5.33 (1H, m, H-1), 5.80, 6.43 (each 1H, d, J = 3.4, H-13). ESI-MS *m/z* 330 [M + Na + H]⁺ [16].

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