

Anti-inflammatory effects of *Mitrephora sirikitiae* leaf extract and isolated lignans in RAW 264.7 cells

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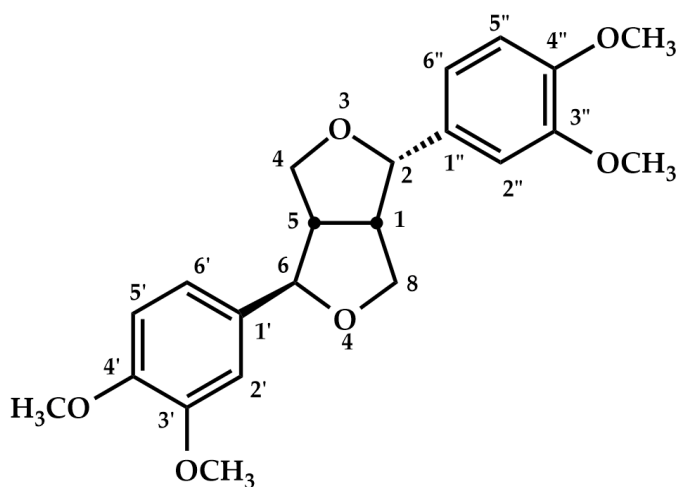
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Spectroscopic and physical data of isolated lignans 1-6 from *Mitrephora sirikitiae* Leaf extract.

1. (-)-Epieudesmin (1)



Physical characteristics: Colorless crystals from CH₂Cl₂/MeOH; m.p. 128-130 °C

$[\alpha]_D^{25}$: -123.66 (*c* 1.0, CHCl₃)

UV $\lambda_{\max}^{\text{MeOH}}$ nm (log ϵ): 231 (3.91), 279 (4.38)

FTIR (neat) ν_{\max} : 2918, 2915, 1607, 1590, 1463, 1265, 1160, 1142 cm⁻¹

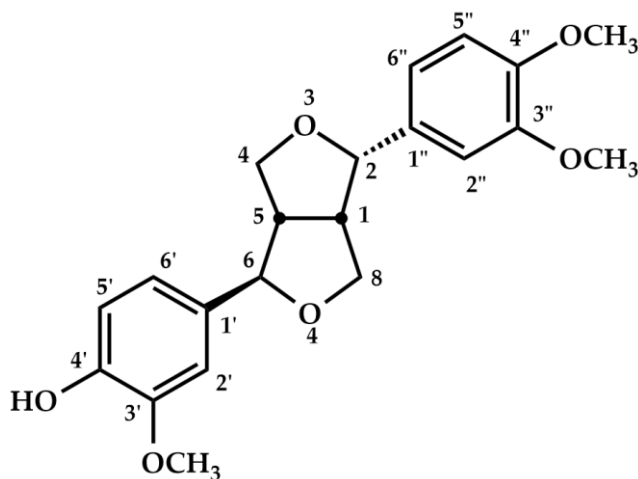
HR-ESI-MS *m/z*: 409.1621 [M+Na]⁺

EI-MS *m/z* (% relative intensity): 386 [M]⁺ (68), 355 [M-OCH₃]⁺ (6), 340 [M-OCH₃-CH₃]⁺ (9), 309 [M-2OCH₃-CH₃]⁺ (7), 220 (10), 205 (13), 194 (14), 189 (15), 177 (36), 165 (100), 151 (26)

¹H-NMR (500 MHz, CDCl₃) δ ppm (mult., J (Hz)): 6.97 (1H, *d*, 1.1 Hz, H-2''), 6.95 (1H, *d*, 1.9 Hz, H-2'), 6.93 (1H, *dd*, 8.2, 1.9 Hz, H-6'), 6.90 (1H, *d*, 8.4 Hz, H-6''), 6.88 (1H, *d*, 8.4 Hz, H-5''), 6.87 (1H, *d*, 8.2 Hz, H-5'), 4.91 (1H, *d*, 5.7 Hz, H-2), 4.48 (1H, *d*, 7.2 Hz, H-6), 4.16 (1H, *dd*, 9.4, 0.8 Hz, H-4), 3.94 (3H, *s*, OMe-3''), 3.93 (3H, *s*, OMe-3'), 3.92 (3H, *s*, OMe-4''), 3.91 (3H, *s*, OMe-4'), 3.88 (1H, *m*, H-8), 3.88 (1H, *m*, H-4), 3.39 (1H, *m*, H-1), 3.34 (1H, *t*, 8.4 Hz, H-8), 2.95 (1H, *m*, H-5)

¹³C-NMR (125 MHz, CDCl₃) δ ppm: 149.28 (C-3'), 148.90 (C-3''), 148.77 (C-4'), 148.07 (C-4''), 133.72 (C-1'), 131.00 (C-1''), 118.47 (C-6'), 117.75 (C-6''), 111.10 (C-5', C-5''), 109.23 (C-2'), 109.01 (C-2''), 87.64 (C-6), 82.07 (C-2), 71.04 (C-4), 69.74 (C-8), 55.97 (OMe-4'), 55.95 (OMe-4''), 55.93 (OMe-3', OMe-3''), 54.50 (C-5), 50.18 (C-1)

2. (-)-Phylligenin (2)



Physical characteristics: Colorless crystals from CH₂Cl₂/MeOH; m.p. 135-136 °C;

$[\alpha]_D^{24}$: -128.53 (*c* 1.0, CHCl₃)

UV λ_{max}^{MeOH} nm (log ϵ): 231 (3.95), 280 (4.38)

FTIR (neat) ν_{max} : 3430, 2961, 2939, 2844, 1607, 1589, 1515, 1469, 1460, 1447, 1460, 1417, 1382, 1347, 1278, 1264, 1256, 1236, 1193, 1157, 1143, 1127, 1076, 1023 cm⁻¹

HR-ESI-MS m/z : 395.1471 [M+Na]⁺

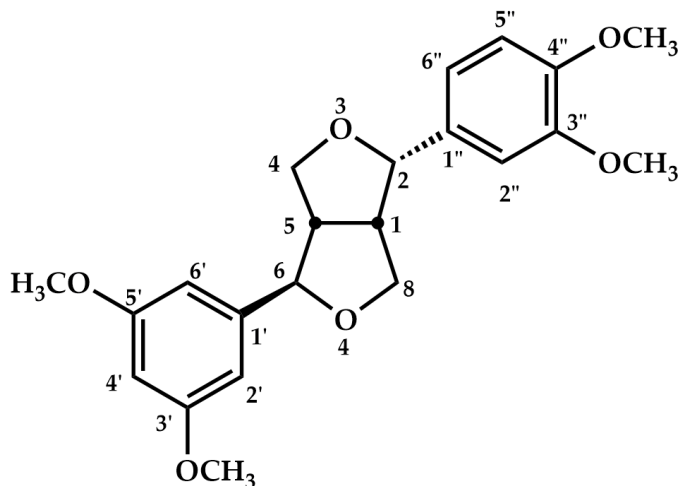
EI-MS m/z (% relative intensity): 372 [M]⁺ (68), 341 [M-OCH₃]⁺ (6), 326 [M-OCH₃-CH₃]⁺ (9), 273 (8), 205 (21), 194 (13), 189 (19), 177 (27), 165 (42), 151 (100)

¹H-NMR (500 MHz, CDCl₃) δ ppm (mult., J (Hz)): 6.99 (1H, *br s*, H-2'), δ 6.96 (1H, *d*, 1.7 Hz, H-2''), 6.91 (2H, *m*, H-6', H-6''), 6.93 (2H, *d*, 8.1 Hz, H-5', H-5''), 5.74 (1H, *s*, OH-4'), 4.92 (1H, *d*, 5.5 Hz, H-6), 4.48 (1H, *d*, 7.1 Hz, H-2), 4.18 (1H, *d*, 9.7 Hz, H-8), 3.99 (3H, *s*, OMe-3''), 3.93 (3H, *s*, OMe-3'), 3.92 (3H, *s*, OMe-4''), 3.89 (2H, *m*, H-4, H-8), 3.39 (2H, *m*, H-4, H-5), 2.95 (1H, *br dd*, 14.4, 7.2 Hz, H-1)

¹³C-NMR (125 MHz, CDCl₃) δ ppm: 148.82 (C-3'), 146.77 (C-3''), 147.98 (C-4'), 145.36 (C-4''), 133.00 (C-1'), 130.95 (C-1''), 119.22 (C-6''), 117.72 (C-6'), 114.28 (C-5''), 111.00 (C-5'), 108.91 (C-2'), 108.56 (C-2''),

87.77 (C-2), 82.66 (C-6), 71.02 (C-8), 69.73 (C-4), 55.95 (OMe-4''), 55.93 (OMe-3', OMe-3''), 54.52 (C-1), 50.16 (C-6)

3. 2-(3,4-Dimethoxyphenyl)-6-(3,5-dimethoxyphenyl)-3,7-dioxabicyclo[3.3.0]octane (3)



Physical characteristics: A pale yellow semisolid

$[\alpha]_D^{28.7}$: -17.68 (*c* 1.0, MeOH)

UV $\lambda_{\max}^{\text{MeOH}}$ nm (log ϵ): 230 (4.30), 280 (3.91)

FTIR (neat) ν_{\max} : 2930, 2850, 1593, 1512, 1461, 1419, 1258, 1232, 1156, 1136, 1061, 1021 cm^{-1}

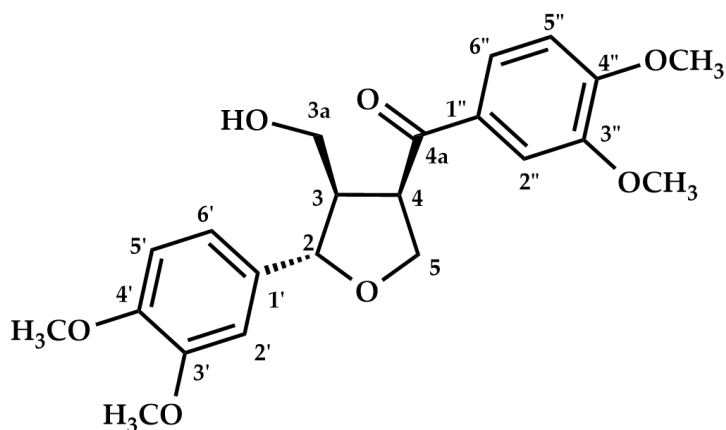
HR-ESI-MS m/z : 409.1621 $[\text{M}+\text{Na}]^+$

EI-MS m/z (% relative intensity): 386 $[\text{M}]^+$ (5), 384 $[\text{M}-2\text{H}]^+$ (34), 367 (14), 337 (41), 325 (82), 292 (32), 275 (20), 263 (27), 220 (32), 202 (41), 189 (100), 165 (85).

$^1\text{H-NMR}$ (400 MHz, CD_3OD) δ_{H} ppm (mult., *J* (Hz)): δ 6.96 (1H, *s*, H-2'), 6.91 (3H, *br s*, H-4', H-6', H-2''), 6.88 (1H, *d*, 8.2 Hz, H-5''), 6.82 (1H, *dd*, 8.2, 1.7 Hz, H-6''), 4.64 (1H, *d*, 7.4 Hz, H-6), 4.50 (1H, *d*, 8.4 Hz, H-2), 4.25 (1H, *dd*, 8.8, 4.4 Hz, H-4), 3.95 (1H, *dd*, 8.8, 7.6 Hz, H-4), 3.82 (3H, *s*, 5'-OMe), 3.81 (3H, *s*, 3'-OMe), 3.79 (6H, *s*, 3''-OMe, 4''-OMe), 3.30 (1H, *m*, H-8), 3.23 (1H, *dd*, 16.5, 5.9 Hz, H-8), 2.54 (1H, *m*, H-5), 1.88 (1H, *m*, H-1)

$^{13}\text{C-NMR}$ (100 MHz, CD_3OD) δ_{C} ppm: 150.51 (C-5'), 150.46 (C-3''), 149.98 (C-3', C-4''), 137.49 (C-1''), 136.25 (C-1'), 120.61 (C-6''), 120.02 (C-6'), 112.85 (C-4'), 112.71 (C-5''), 111.73 (C-2''), 111.32 (C-2'), 84.83 (C-6), 73.36 (C-2), 71.43 (C-4), 62.69 (C-8), 56.57 (3''-OMe), 56.50 (3'-OMe, 5'-OMe), 56.44 (4''-OMe), 53.57 (C-1), 50.77 (C-5)

4. Magnone A (4)



Physical characteristics: Colorless crystals from CH₂Cl₂/MeOH; m.p. 139-140 °C

$[\alpha]_D^{25}$: 18.54 (*c* 1.0, MeOH)

UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm (log ϵ): 230 (4.37), 276 (4.13), 305 (3.93)

FTIR (neat) ν_{max} : 3461, 2912, 2839, 1651, 1592, 1583, 1517, 1467, 1441, 1420, 1334, 1288, 1237, 1203, 1192, 1154, 1104, 1080, 1020 cm⁻¹

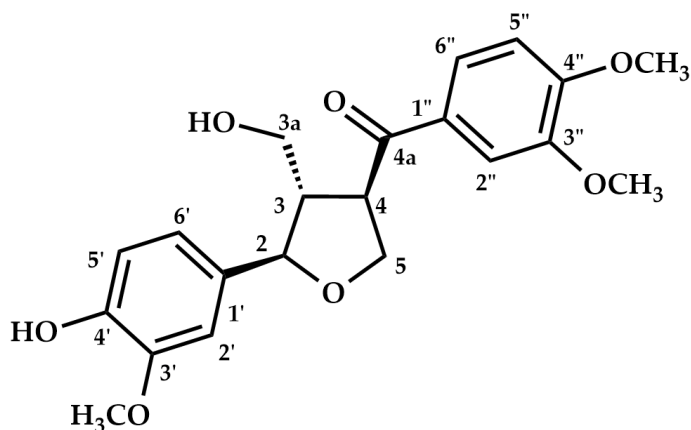
HR-ESI-MS *m/z*: 425.1584 [M+Na]⁺

EI-MS *m/z* (% relative intensity): 368 (68), 355 (6), 340 (9), 309 (7), 220 (10), 205 (13), 194 (14), 189 (15), 177 (36), 165 (100), 151 (26).

¹H-NMR (400 MHz, CDCl₃) δ ppm (mult., *J* (Hz)): 7.66 (1H, *dd*, 8.2, 2.0 Hz, H-6''), δ 7.63 (1H, *d*, 2.0 Hz, H-2''), 7.09 (1H, *d*, 1.9 Hz, H-2'), 6.99 (1H, *dd*, 8.2, 1.9 Hz, H-6'), 6.98 (1H, *d*, 8.3 Hz, H-5''), 6.89 (1H, *d*, 8.2 Hz, H-5'), 4.75 (1H, *d*, 9.0 Hz, H-2), 4.37 (1H, *dd*, 11.2, 10.9 Hz, H-5), 4.24 (2H, *m*, H-4, H-5), 4.01 (3H, *s*, OMe-4''), 4.00 (3H, *s*, OMe-4'), 3.97 (3H, *s*, OMe-3'), 3.93 (3H, *s*, OMe-3''), 3.84 (2H, *dd*, 10.9, 4.5 Hz, H-3a), 2.77 (1H, *m*, H-3)

¹³C-NMR (100 MHz, CDCl₃) δ ppm: 197.97 (C-4a), 153.70 (C-4''), 149.27 (C-4', C-3''), 148.95 (C-3'), 132.94 (C-1'), 129.75 (C-1''), 123.23 (C-6''), 119.36 (C-6', C-2''), 110.82 (C-5'), 110.72 (C-5''), 109.56 (C-2'), 83.82 (C-2), 70.90 (C-5), 61.51 (C-3a), 56.81 (OMe-4''), 56.06 (OMe-4'), 55.95 (OMe-3', OMe-3''), 52.14 (C-3), 49.72 (C-4)

5. Mitrephoran (5)



Physical characteristics: Colorless crystals were obtained from $\text{CH}_2\text{Cl}_2/\text{MeOH}$; m.p. 146-148 °C

$[\alpha]_{\text{D}}^{25}$: 14.00 (c 0.45, CHCl_3)

UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm (log ϵ): 230 (4.37), 276 (4.13), 305 (3.93)

FTIR (neat) ν_{max} : 3396, 3259, 2954, 2872, 2841, 1672, 1597, 1587, 1515, 1463, 1439, 1418, 1357, 1347, 1287, 1259, 1158, 1122, 1088, 1060, 1033, 1020, 1005, and 968 cm^{-1}

HR-ESI-MS m/z : 411.1420 $[\text{M}+\text{Na}]^+$

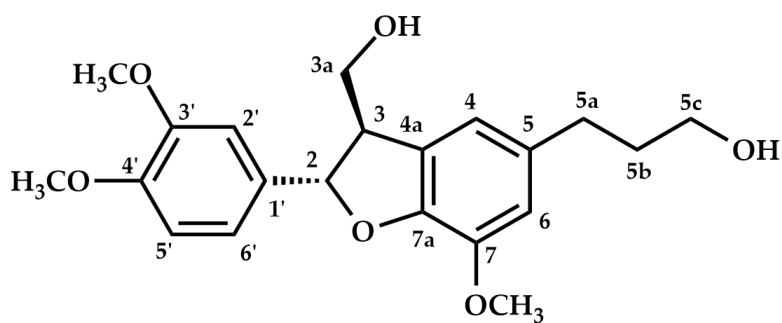
EI-MS m/z (% relative intensity): 388 $[\text{M}]^+$ (8), 218 (14), 196 (38), 180 (93), 165 (100), and 151 (60)

$^1\text{H-NMR}$ (400 MHz, CDCl_3) δ ppm (mult., J (Hz)): 7.67 (1H, dd, 8.4, 2.0 Hz, H-6''), 7.63 (1H, d, 2.0 Hz, H-2''), 7.08 (1H, br s, H-2'), 6.98 (1H, d, 8.4 Hz, H-5''), 6.94 (1H, m, H-6')*, 6.93 (1H, m, H-5')*, 4.73 (1H, d, 9.1 Hz, H-2), 4.37 (1H, m, H-5), 4.24 (2H, m, H-4, H-5), 4.02 (3H, s, 4''-OMe), 4.01 (3H, s, 3''-OMe), 3.99 (3H, s, 3'-OMe), 3.83 (1H, dd, 10.9, 4.4 Hz, H-3a), 3.73 (1H, dd, 10.9, 5.6 Hz, H-3a), 2.96 (1H, m, H-3)

* H-5' and H-6' were observed as broad singlets because of the virtual coupling effects.

$^{13}\text{C-NMR}$ (100 MHz, CDCl_3) δ ppm: 198.04 (C-4a), 153.70 (C-4''), 149.26 (C-3''), 146.88 (C-3'), 145.63 (C-4'), 132.31 (C-1'), 129.77 (C-1''), 123.23 (C-6''), 120.17 (C-6'), 114.04 (C-5''), 110.57 (C-2''), 110.12 (C-5'), 108.92 (C-2'), 83.91 (C-2), 70.85 (C-5), 61.44 (C-3a), 56.18 (4''-OMe), 56.06 (3''-OMe), 56.02 (3'-OMe), 52.21 (C-3), 49.67 (C-4)

6. 3',4'-O-Dimethylcedrusin (6)



Physical characteristics: A pale yellow semisolid; $[\alpha]_D^{28.7}$ -8.52 (c 0.1, CHCl₃)

UV $\lambda_{\max}^{\text{MeOH}}$ nm (log ϵ): 231 (4.19), 281 (3.73)

FTIR (neat) ν_{\max} : 3375, 2934, 1604, 1515, 1497, 1451, 1422, 1323, 1259, 1235, 1210, 1137, 1021 cm⁻¹

HR-ESI-MS m/z : 397.1624 [M+Na]⁺

EI-MS m/z (% relative intensity): 374 [M]⁺ (4), 356 [M-H₂O]⁺ (83), 341 [M-CH₃-H₂O]⁺ (100), 325 [M-OCH₃-H₂O]⁺ (39), 309 (15).

¹H-NMR (400 MHz, CD₃OD) δ ppm (mult., J (Hz)): 6.91 (1H, *br s*, H-2'), 6.88 (1H, *d*, 8.2, H-5'), 6.82 (1H, *dd*, 8.2, 1.7, H-6'), 6.72 (1H, *s*, H-6), 6.71 (1H, *s*, H-4), 5.52 (1H, *d*, 6.1 Hz, H-2), 3.84 (3H, *s*, 7-OMe), 3.81 (1H, *dd*, 11.0, 5.8 Hz, Hb-3a), 3.79 (3H, *s*, 4'-OMe), 3.77 (3H, *s*, 3'-OMe), 3.74 (1H, *dd*, 11.0, 7.3 Hz, Ha-3a), 3.56 (2H, *t*, 6.5 Hz, H-5c), 3.45 (1H, *m*, H-3), 2.61 (2H, *t*, 7.7 Hz, H-5a), 1.80 (2H, *tt*, 7.7, 6.5 Hz, H-5b)

¹³C-NMR (100 MHz, CD₃OD) δ ppm: 150.56 (C-3'), 150.25 (C-4'), 147.50 (C-7a), 145.22 (C-7), 136.99 (C-5), 136.19 (C-1'), 129.73 (C-4a), 119.43 (C-6'), 117.92 (C-4), 114.11 (C-6), 112.87 (C-5'), 110.70 (C-2'), 88.70 (C-2), 65.02 (C-3a), 62.23 (C-5c), 56.74 (7-OMe), 56.49 (4'-OMe), 56.42 (3'-OMe), 55.50 (C-3), 35.80 (C-5b), 32.89 (C-5a)