Supporting Information

Neurodifferentiating Potential of 8-Prenylnaringenin and Related Compounds in Neural Precursor Cells and Correlation with Estrogen-Like Activity

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Fig. 1S Differentiation-inducing activity (DIA) of naringenin (1), 8-prenynaringenin (2), and liquiritigenin (7) (c = 10 μM). Retinoic acid in combination with valproic acid [c(RA/VPA) = 10/50 μM] was used as a positive control.
Figure 2S

Photographs of immunostaining of the control (unaffected cells), positive control RA/VPA (10 µm/50 µM), 8-prenylarigenin (2) and 6-prenylarigenin (3).

Chemical experimental procedures and analysis

GC/MS: Hewlett Packard HP 6890/HP MS 5973; column HP-5MS J & W Scientific; NMR: Jeol JNM-ECS-400; IR: Nicolet 380 FT-IR ATR (Firma Thermo Scientific).

Compound structures were numbered according to Grayer and Veitch (R. J. Grayer, N. C. Veitch In: Flavonoids: chemistry, biochemistry, and applications (Eds.: O. M. Andersen, K. R. Markham), CRC Press: Boca Raton; 2006.)

GC/MS method

The GC program starts with 80°C and heats with a rate of 14°C/min to 250°C, afterwards with a rate of 1.5°C/min to 300°C. This temperature was held for a period of 22 min. Flavonoids were derivatized with N,O-bis(trimethylsilyl)trifluoroacetamid at room temperature, and the mass spectra showed the characteristic splitting \([M^+ + xTMS-15] (100\%).\)
**Microwave synthesis**

Microwave irradiation was carried out with a CEM DiscoverS class single-mode synthesis system interfaced with a laptop pc running CEM synergy software monitoring the reaction. The temperature was checked by an external infrared sensor in the floor of the cavity. Once the target temperature is reached, the microwave system automatically starts to count down the hold time. For reactions, CEM Vials (10 mL) with snap-on caps were used. The pressure was monitored by a sensor outside the snap-on caps. The upper pressure limit was set to 18 bar. Temperature/pressure recording are attached to the CEM synergy reaction files.

**8-Prenylnaringenin**

5,7-Dihydroxy-2-(4-hydroxy-phenyl)-8-(3-methyl-but-2-enyl)-chroman-4-one.

R<sub>f</sub> 0.19 silica gel 60 F<sub>254</sub>, n-hexane/ethyl acetate (2/1); IR (ATR) ν = 3307 (br), 2913 (w), 1599 (m), 1516 (w), 1504 (w), 1435 (w), 1373 (w), 1343 (w), 1224 (w), 1168 (w), 1072 (s), 1014 (w), 985 (w), 949 (w), 899 (w) 878 (w), 831 (w), 621 (w), 558 (m), 527 (w), 441 (w) cm<sup>-1</sup>; <sup>1</sup>H-NMR (d<sub>6</sub>-acetone, 400 Hz) δ (ppm) = 1.59 (s, 3H, 4'<H>), 1.59 (s, 3H, 5'<H>), 2.76 (dd, 1H, J = 17.40 Hz, 3-H), 3.11 (dd, 1H, J = 16.94 Hz, 3-H), 3.20 (d, 2H, J = 7.33 Hz, 1''-H), 5.17 (t, 1H, J = 7.33 Hz, 2''-H), 5.43 (dd, 1H, J = 12.82 Hz, 2-H), 6.02 (s, 1H, 6-H), 6.88 (d, 2H, J = 8.70 Hz, 3'-H & 5'-H), 7.40 (dd, 2H, J = 8.70 Hz, 2'-H & 6'-H), 12.13 (s, 1H, OH); <sup>13</sup>C-NMR (d<sub>6</sub>-acetone, 100 Hz) δ (ppm) = 17.82 (C-4'), 22.21 (C-1'), 25.85 (C-5'), 43.38 (C-3), 79.67 (C-2), 96.34 (C-6), 103.18 (C-4a), 108.22 (C-8), 116.10 (C-3' & C-5'), 123.68 (C-2'H), 128.77 (C-2' & C-6'), 130.94 (C-1'), 131.09 (C-3'), 158.59 (C-4'), 161.01 (C-8a), 162.90 (C-5), 165.07 (C-7), 197.49 (C-4); MS (GC) R<sub>T</sub> = 26.71 min, m/z<sub>silylated</sub> (%) = 556 (8), 543 (22), 542 (46), 541 (100), 513 (20), 485 (14), 350 (11), 349 (36), 321 (21), 309 (32), 293 (11), 73 (24).
8-Geranylnaringenin

2,3-Dihydro-5,7-dihydroxy-2-(4-hydroxyphenyl)-8-((E)-3,7-dimethylocta-2,6-dienyl)chromen-4-one.

R<sub>f</sub> 0.64 silica gel 60 F<sub>254</sub>, n-hexane/ethyl acetate (1/1); IR (ATR) ν = 3305 (br), 2913 (w), 1633 (w), 1598 (w), 1519 (m), 1373 (w), 1217 (w), 1168 (w), 1069 (s), 830 (m), 637 (w), 621 (w), 555 (w), 528 (w);<sup>1</sup>H-NMR (d<sub>6</sub>-acetone, 400 MHz) δ (ppm) = 1.54 (s, 3H, 10′′-H), 1.59 (s, 6H, 4′′-H & 9′′-H), 1.89 - 2.07 (m, je 2H, 5′′ -H & 6′′ -H), 2.75 (dd, 1H, 2<sup>2</sup>J = 17.1 Hz, 3′′-H), 3.13 (dd, 1H, 2<sup>3</sup>J = 17.1 Hz, 3′′-H), 3.22 (d, 2H, 2<sup>1</sup>J = 7.33 Hz, 1′′-H), 5.04 - 5.07 (m, 1H, 7′′ -H), 5.19 - 5.23 (m, 1H, 2′′-H), 5.54 (dd, 1H, 2<sup>2</sup>J = 12.7 Hz, 2-H), 6.02 (s, 1H, 6-H), 6.89 (d, 2H, 2<sup>1</sup>J = 8.89 Hz, 3′-H & 5′ -H), 7.40 (d, 2H, 2<sup>1</sup>J = 8.60 Hz, 2′-H & 6′ -H), 8.95 (s, 1H, 7-OH), 12.13 (s, 1H, 5-OH).<sup>13</sup>C-NMR (d<sub>6</sub>-acetone, 100 MHz) δ (ppm) = 16.19 (C-4′′), 17.69 (C-9′′), 22.13 (C-1′′), 25.79 (C-10′′), 27.36 (C-5′′), 40.47 (C-6′′), 43.46 (C-3), 79.71 (C-2), 96.35 (C-6), 103.28 (C-4a), 108.32 (C-8), 116.11 (C-3′ & C-5′), 123.56 (C-2′′), 125.15 (C-7′′), 128.81 (C-2′ & C-6′), 131.08 (C-1′), 131.55 (C-8′′), 134.93 (C-3′′), 158.54 (C-4′), 161.12 (C-8a), 162.98 (C-5), 164.90 (C-71), 197.52 (C-4); MS (GC) <sup>1</sup>R<sub>T</sub> = 36.92 min m/z (%) = 625 (4), 624 (7), 611 (25), 610 (51), 609 (100), 555 (15), 513 (10), 502 (13), 501 (29), 485 (18), 429 (13), 364 (20), 363 (68), 310 (16), 309 (49), 291 (13), 207 (22), 73 (35), 4 (39).

8-Farnesylnaringenin

2,3-Dihydro-5,7-dihydroxy-2-(4-hydroxyphenyl)-8-((2E,6E)-3,7,11-trimethyldecada-2,6,10-trienyl)chromen-4-one.

R<sub>f</sub> 0.66 silica gel 60 F<sub>254</sub>, n-hexane/ethyl acetate (1/1); IR (ATR) ν = 3270 (m, br), 2963 (m), 2913 (m), 1633 (m), 1596 (s), 1517 (w), 1434 (m), 1376 (w), 1219 (w), 1169 (w), 1069 (s), 984,2 (w), 950 (w), 878 (w), 831 (s), 638 (w), 559 (m), 527 (w) cm<sup>-1</sup>; <sup>1</sup>H-NMR (d<sub>6</sub>-acetone, 400 Hz) δ (ppm) = 1.54 & 1.56 (s, je 3H, 9′′-H & 14′′-H), 1.60 (s, 3H, 15′′-H), 1.63 (s, 3H, 4′′-H), 1.89 - 2.08 (m, je 2H, 5′′-H, 6′′-H, 11′′-H), 2.73 (dd, 1H, 2<sup>2</sup>J = 17.10 Hz, 3′′-H), 3.09 (dd, 1H, 2<sup>2</sup>J = 17.10 Hz, 3′′-H), 3.23 (d, 2H, 2<sup>1</sup>J = 7.33 Hz, 1′′-H), 5.03 – 5.10 (m, 2H, 7′′-H and 12′′-H), 5.20 – 5.23 (m, 1H, 2′′-H), 5.40 (dd, 1H, 2<sup>2</sup>J = 13.90 Hz, 3′′-H), 6.03 (s, 1H, 6-H), 6.89 (d, 2H, 2<sup>1</sup>J = 8.79 Hz, 3′-H & 5′-H), 7.38 (d, 2H, 2<sup>1</sup>J = 8.79 Hz, 2′-H & 6′-H); <sup>13</sup>C-NMR (d<sub>6</sub>-acetone, 100 Hz) δ

(ppm) = 16.08 (C-9´´), 16.18 (C-4´´), 17.66 (C-14´´), 22.11 (C-1´´), 25.79 (C-15´´), 27.12 (C-6´´),
27.34 (C-11´´), 40.34 & 40.40 (C-5´´ & C-10´´), 43.40 (C-3), 79.62 (C-2), 96.34 (C-6) 103.20 (C-4a),
108.26 (C-8), 116.06 (C-3´ & C-5´), 123.54 (C-2´´), 124.95 (C-7´´), 125.06 (C-12´´), 128.69 (C-2´ &
C-6´), 130.95 (C-1´), 131.49 (C-13´´), 134.78 (C-8´´), 135.32 (C-3´´), 158.43 (C-4´), 161.02 (C-8a),
162.85 (C-5), 164.86 (C-7), 197.37 (C-4); MS (GC) R_T = 49.44 min, m/z silylated (%) = 692 (1), 677 (8),
281 (28), 253 (12), 209 (15), 208 (21), 207 (100), 191 (10), 4 (42).

6-Prenylnaringenin
5,7-Dihydroxy-2-(4-hydroxy-phenyl)-6-(3-methyl-but-2-enyl)-chroman-4-one.
R_f 0.16 silica gel 60 F_254, n-hexane/ethyl acetate (2/1); IR (ATR) ν = 3120 (w), 2961 (w), 2912 (w),
1651 (m), 1633 (w), 1614 (w), 1584 (s), 1557 (m), 1538 (w), 1516 (w), 1504 (w), 1486 (w), 1463 (w),
1446 (w), 1326 (w), 1336 (m), 1310 (w), 1246 (m), 1215 (m), 1185 (m), 1150 (w), 1075 (w), 1015 (w), 992 (m), 935 (w), 903 (m), 867 (m), 829 (m), 819 (w), 780 (w), 757 (w), 724 (m), 701 (m), 650 (w), 635 (w), 614 (w), 574 (m), 561 (w), 533 (s), 464 (w), 443 (w), 436 (m), 414 (m) cm^{-1}; ^1H-NMR (d_6-acetone, 400 Hz) δ (ppm) = 1.62 (s, 3H, 4´´-H), 1.74 (s, 3H, 5´´-H), 2.70 (dd, 1H, J = 16.94 Hz, 3-
H), 3.16 (dd, 1H, J = 16.94 Hz, 3-H), 3.23 (d, 2H, J = 7.33 Hz, 1´´-H), 5.22 (t, 1H, J = 6.87 Hz, 2´´-
H), 5.41 (dd, 1H, J = 12.82 Hz, 2-H), 6.02 (s, 1H, 8-H), 6.88 (d, 2H, J = 8.70 Hz, 3´-H & 5´-H), 7.37 (d, 2H, J = 8.70 Hz, 2´-H & 6´-H), 12.46 (s, 1H, 5-OH); ^13C-NMR (d_6-acetone) δ (ppm) = 17.80 (C-
5´´), 21.59 (C-1´´), 25.82 (C-4´´), 43.58 (C-3), 79.85 (C-2), 95.26 (C-8), 103.06 (C-4a), 108.97 (C-6),
116.12 (C-3´ & C-5´), 123.56 (C-2´´), 128.96 (C-2´ & C-6´), 130.88 (C-1´), 131.19 (C-3´), 158.66 (C-
4´), 161.97 (C-8a), 162.23 (C-5), 164.79 (C-7), 197.31 (C-4); MS (GC) R_T = 28.10 min, m/z silylated (%)
= 556 (2), 543 (22), 542 (47), 541 (100), 73 (8).

8-Prenylnarinegenin-C
5-Hydroxy-2-(4-hydroxy-phenyl)-8,8-dimethyl-2,3-dihydro-8H-pyrano[2,3-f]chromen-4-one.
R_f 0.48 silica gel 60 F_254, n-hexane/ethyl acetate (2/1); IR (ATR) ν = 3299 (w, br), 1650 (w), 1644 (w),
1634 (w), 1615 (w), 1587 (s), 1558 (w), 1519 (s), 1506 (w), 1472 (w), 1455 (w), 1416 (w), 1370 (w),
1362 (w), 1355 (m), 1311 (w), 1275 (s), 1088 (m), 1069 (m), 919 (w), 897 (w), 881 (w), 863 (m), 833
(m), 822 (w), 797 (w), 771 (w), 694 (s), 668 (w), 632 (w), 604 (m), 569 (m), 555 (w), 516 (w), 443 (w), 429 (w), 408 (w) cm⁻¹; ¹H-NMR (d₆-acetone, 400 Hz) δ (ppm) = 1.39 (s, 3H, 7´´-H), 1.42 (s, 3H, 8´´-H), 2.77 (dd, 1H, J = 17.4 Hz, 3-H), 3.20 (dd, 1H, J = 16.94 Hz, 3-H), 5.50 (dd, 1H, J = 13.05 Hz, 2-H), 5.58 (d, 1H, J = 10.07, 5´´-H), 5.87 (s, 1H, 6-H), 6.49 (d, 1H, J = 10.07 Hz, 4´´-H), 6.90 (d, 2H, J = 8.7 Hz, 3´-H & 5´-H), 7.41 (d, 2H, J = 8.70 Hz, 2´-H & 6´-H), 8.59 (s, 1H, 4´-OH), 12.24 (s, 1H, 5-OH); ¹³C-NMR (d₆-acetone 100 MHz) δ (ppm) = 28.29 (C-8´´), 28.56 (C-7´´), 43.22 (C-3), 78.78 (C-6´´), 80.03 (C-2), 97.50 (C-6), 102.50 (C-8), 103.51 (C-4a), 116.17 (C-4´´), 116.19 (C-3´ & C-5´), 127.33 (C-5´´), 128.92 (C-2´ & C-6´), 130.50 (C-1´), 158.02 (C-8a), 158.69 (C-4´), 162.65 (C-7), 164.61 (C-5), 197.61 (C-4); MS (GC) Rₜ = 26.09 min, m/z silylated (%) = 482 (16), 468 (24), 467 (63), 290 (20), 276 (21), 275 (100), 73 (14).