Supplementary Materials

Hypoxylonol F isolated from *Annulohypoxylon annulatum* improves insulin secretion by regulating pancreatic β-cell metabolism

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Contents

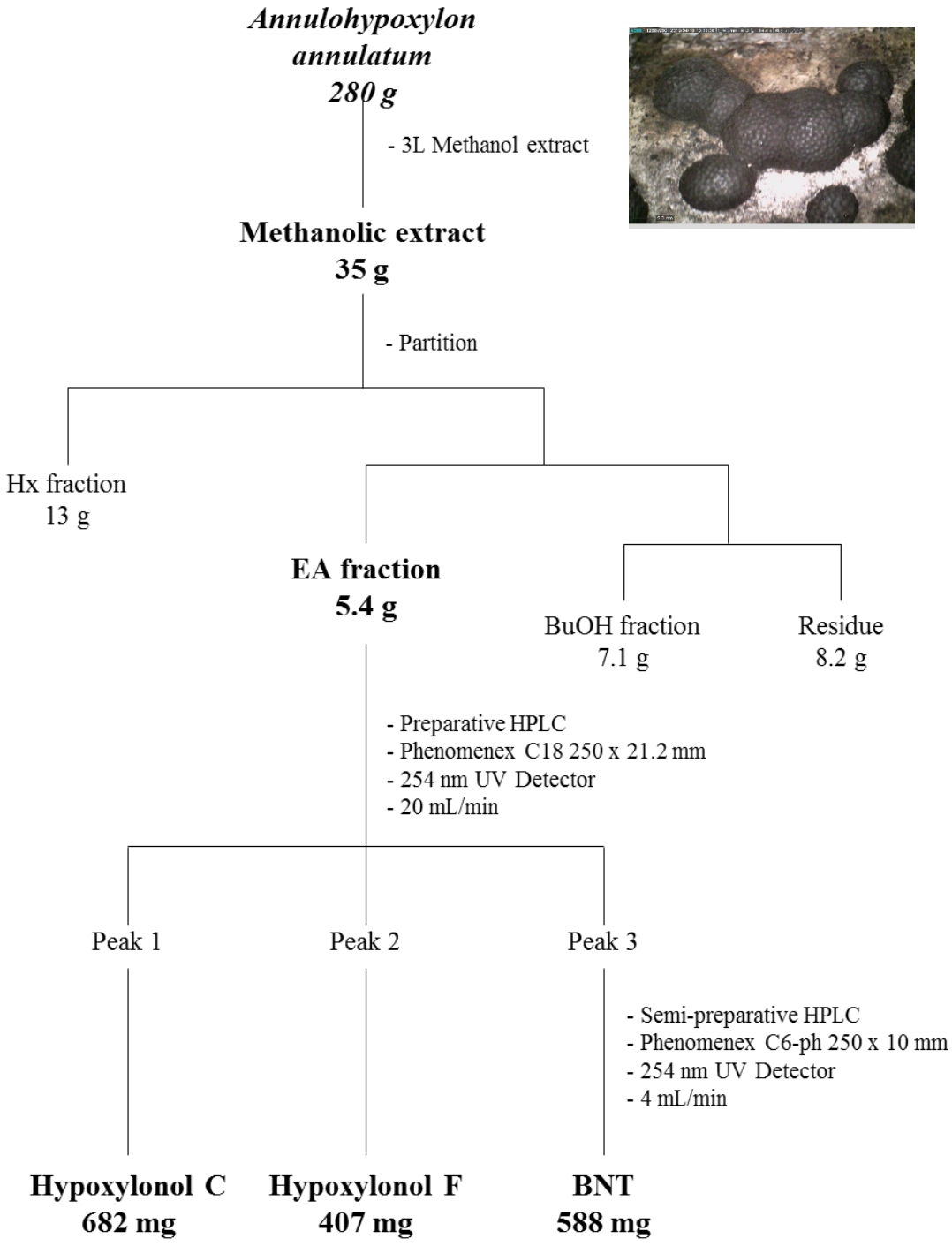
Characterization data

Hypoxylonol C (1).
Yellow amorphous powder. ^{1}H NMR (400 MHz, acetone-d_6) δ 12.61 (s, 1H, OH-9), 8.65 (s, 1H, OH-4), 7.56 (t, 1H, *J* = 8.0 Hz, H-11), 7.49 (dd, 1H, *J* = 8.0, 1.2 Hz, H-12), 7.28 (d, 1H, *J* = 8.0 Hz, H-6), 6.84 (dd, 1H, *J* = 8.0, 1.2 Hz, H-10), 6.68 (d, 1H, *J* = 8.0 Hz, H-5), 5.59 (dd, 1H, *J* = 8.5, 4.2 Hz, H-1), 5.48 (m, 1H, H-3), 5.14 (brd, 1H, OH-3), 4.28 (s, 1H, OH-1), 4.11 (dd, 1H, *J* = 13.8, 5.5 Hz, H-6b), 3.38 (dd, 1H, *J* = 16.5, 5.6 Hz, H-7), 2.47 (dt, 1H, *J* = 13.0, 4.3 Hz, H-2), 2.31 (dd, 1H, *J* = 16.4, 14.0 Hz, H-7), 2.14 (ddd, 1H, *J* = 13.0, 8.5, 3.2 Hz, H-2); ^{13}C NMR (100 MHz, acetone-d_6) δ 206.2 (C-8), 163.8 (C-9), 155.9 (C-4), 144.3 (C-12d), 139.6 (C-12a), 138.3 (C-12c), 137.9 (C-12b), 137.7 (C-11), 136.6 (C-6a), 123.7 (C-6), 120.8 (C-3a), 119.0 (C-12), 117.2 (C-10), 115.7 (C-8a), 114.4 (C-5), 65.5 (C-3), 62.8 (C-1), 49.9 (C-6b), 43.7 (C-7), 42.4 (C-2); HRFABMS *m/z* 335.0925 $[M-H]^-$ (calcd for $C_{20}H_{15}O_5$, 335.0919).

Hypoxylonol F (2).
Yellow amorphous powder. ^{1}H NMR (400 MHz, acetone-d_6) δ 12.60 (s, 1H, OH-9), 7.53 (t, 1H, *J* = 8.0 Hz, H-11), 7.48 (dd, 1H, *J* = 8.0, 1.2 Hz, H-12), 7.29 (d, 1H, *J* = 8.0 Hz, H-6), 6.82 (dd, 1H, *J* = 8.0, 1.2 Hz, H-10), 6.69 (d, 1H, *J* = 8.0 Hz, H-5), 5.38 (d, 1H, *J* = 8.5 Hz, H-1), 5.11 (d, 1H, *J* = 8.5 Hz, H-3), 4.09 (dddd, 1H, *J* = 13.8, 5.4, 23.9, 0.7 Hz, H-6b), 3.39 (dd, 1H, *J* = 16.4, 5.5 Hz, H-7), 2.47 (dt, 1H, *J* = 12.6, 4.3 Hz, H-2), 2.34 (dd, 1H, *J* = 16.3, 13.8 Hz, H-7), 2.24 (dt, 1H, *J* = 12.6, 8.5 Hz, H-2); ^{13}C NMR (100 MHz, acetone-d_6) δ 206.3 (C-8), 163.6 (C-9), 155.7 (C-4), 144.2 (C-12d), 139.4 (C-12a), 139.0 (C-12c), 137.5 (C-12b), 137.1 (C-11), 136.6 (C-6a), 123.8 (C-6), 121.0 (C-3a), 120.9 (C-12), 117.0 (C-10), 115.8 (C-8a), 114.6 (C-5), 67.0 (C-3), 65.2 (C-1), 50.1 (C-6b), 43.7 (C-7), 43.3 (C-2); HRFABMS *m/z* 337.1074 $[M+H]^+$ (calcd for $C_{20}H_{17}O_5$, 337.1076).

4,5,4',5'-Tetrahydroxy-1,1'-binaphthyl, BNT (3).
Yellow amorphous powder. ^{1}H NMR (400 MHz, acetone-d_6) δ 11.04 (br d, 4H, OH-4, 5), 7.16 (d, 2H, *J* = 8.0 Hz, H-7), 7.07 (t, 2H, *J* = 8.0 Hz, H-2), 6.83 (d, 2H, *J* = 8.0 Hz, H-3), 6.72 (dd, 2H, *J* = 8.0, 1.3 Hz, H-6), 6.59 (d, 2H, *J* = 8.0, 1.3 Hz, H-8); ^{13}C NMR (100 MHz, acetone-d_6) δ 155.4 (C-5), 154.8 (C-4), 137.3 (C-8a), 131.2 (C-1), 129.8 (C-7), 127.6 (C-2), 119.2 (C-8), 115.8 (C-4a), 109.7 (C-6), 109.3 (C-3); HRFABMS *m/z* 317.0778 $[M-H]^+$ (calcd for $C_{20}H_{13}O_4$, 317.0773)

Scheme S1. Fractionation schemes of *Annulohypoxylon annulatum*



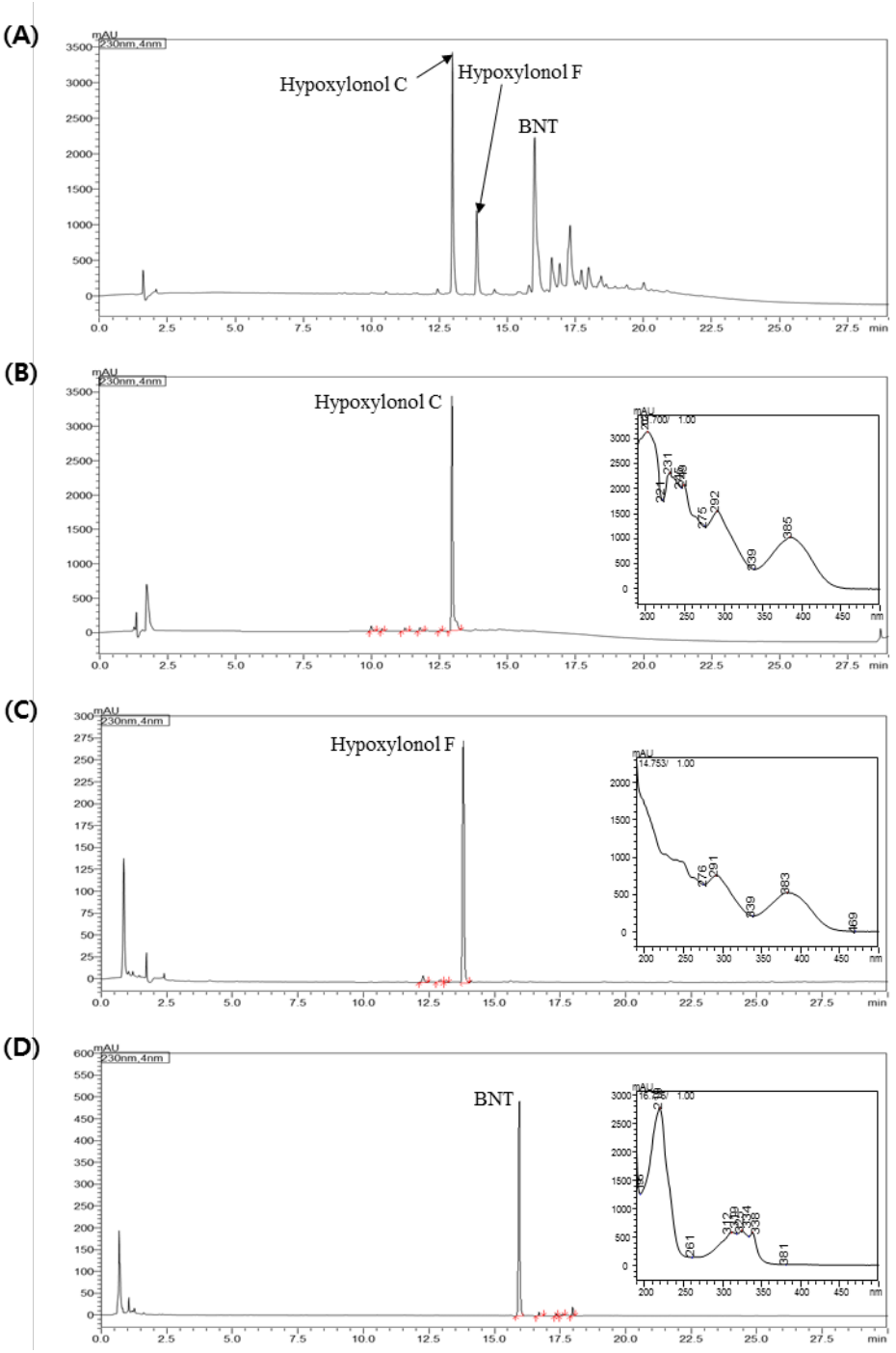


Figure S1. HPLC chromatograms of *Annulohypoxylon annulatum* extract (A), Hypoxylonol C (B), Hypoxylonol F (C), BNT (D).

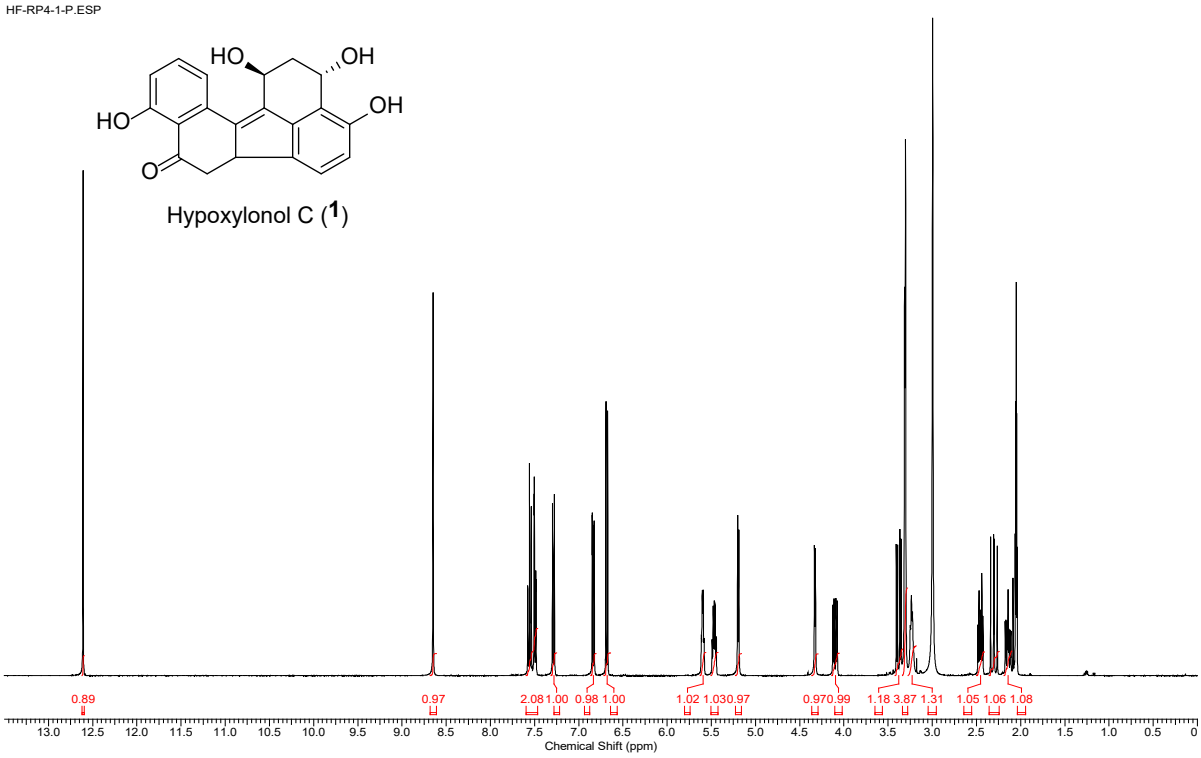


Figure S2. ^{1}H NMR NMR spectrum of hypoxylonol C (**1**)

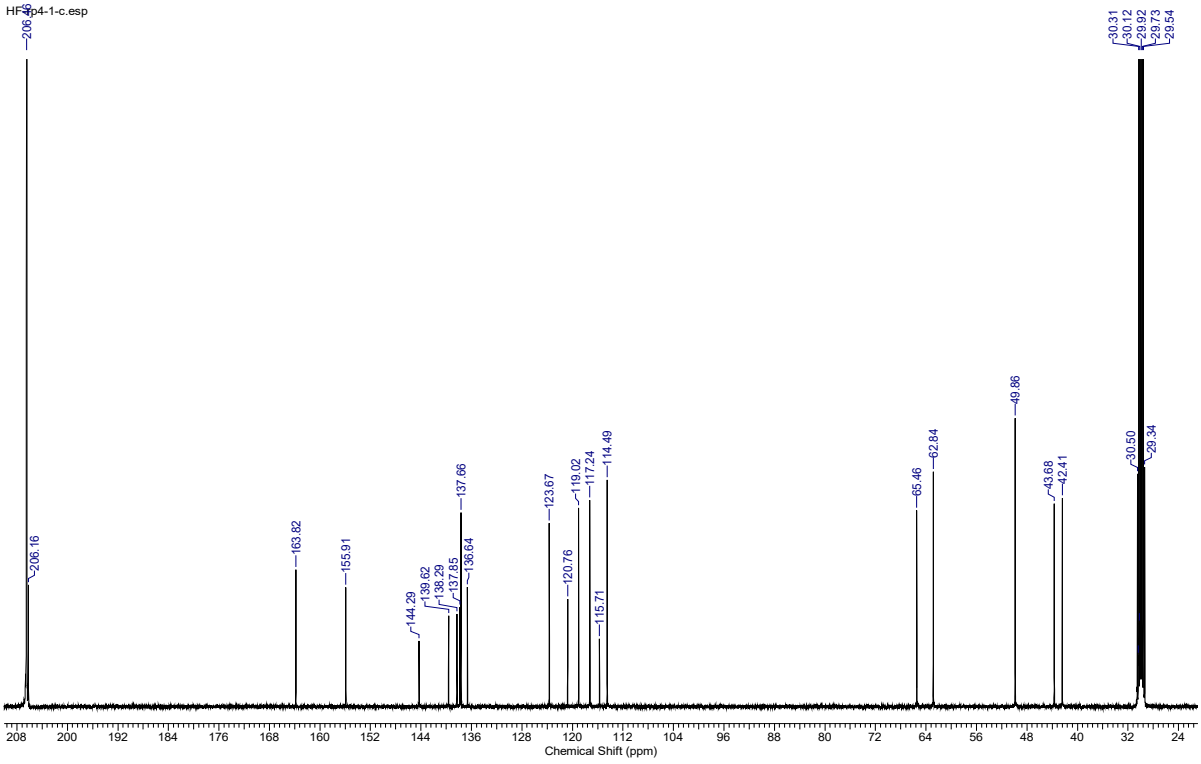
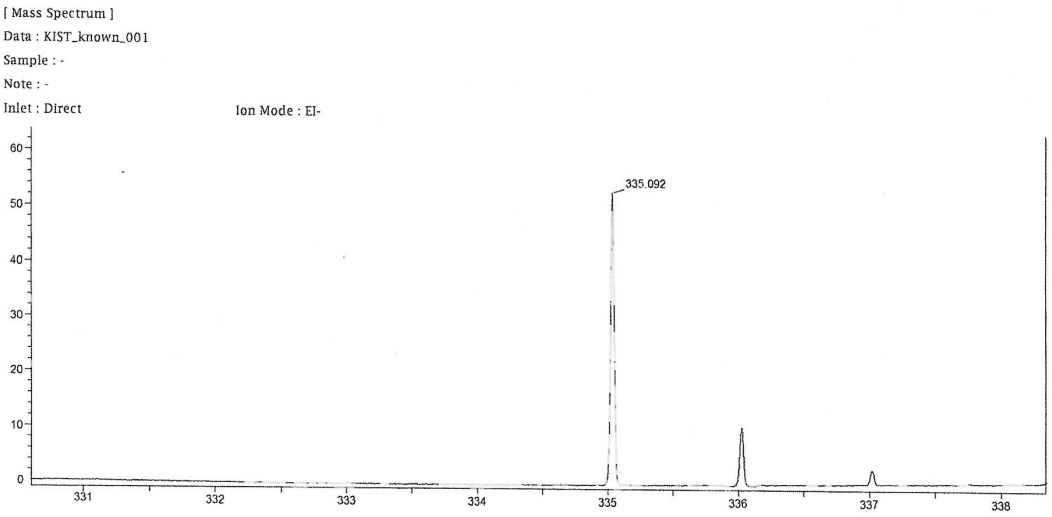


Figure S3. ^{13}C NMR NMR spectrum of hypoxylonol C (**1**)



[Theoretical Ion Distribution]

Molecular Formula : C20 H15 O5 [M-H]⁻

(m/z 335.0919, MW 335.3341)

Base Peak : 335.0919, Averaged MW : 335.3145(a), 335.3316(w)

m/z	INT	
335.0919	100.0000	**********
336.0942	21.4650	**********
337.0968	2.1852	**
338.0991	0.2468	
339.1104	0.0214	

Figure S4. HRMS of hypoxylonol C (**1**)

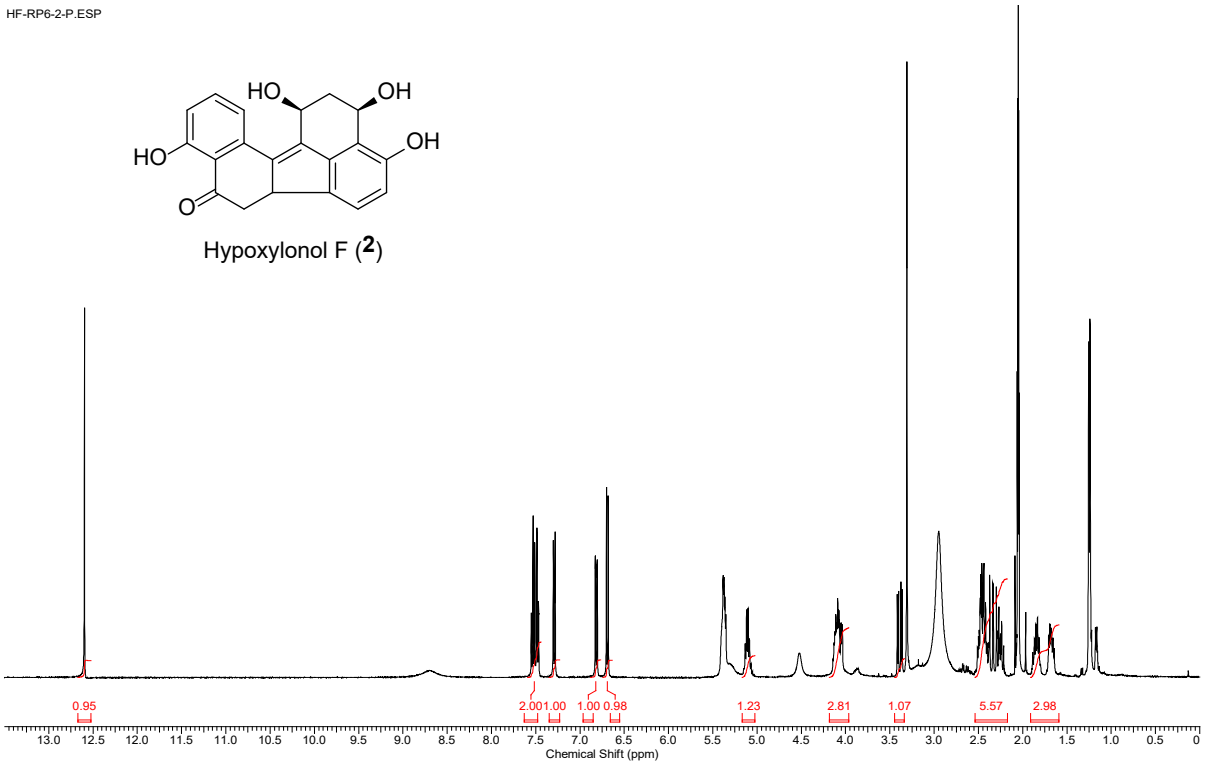


Figure S5. ^{1}H NMR NMR spectrum of hypoxylonol F (**2**)

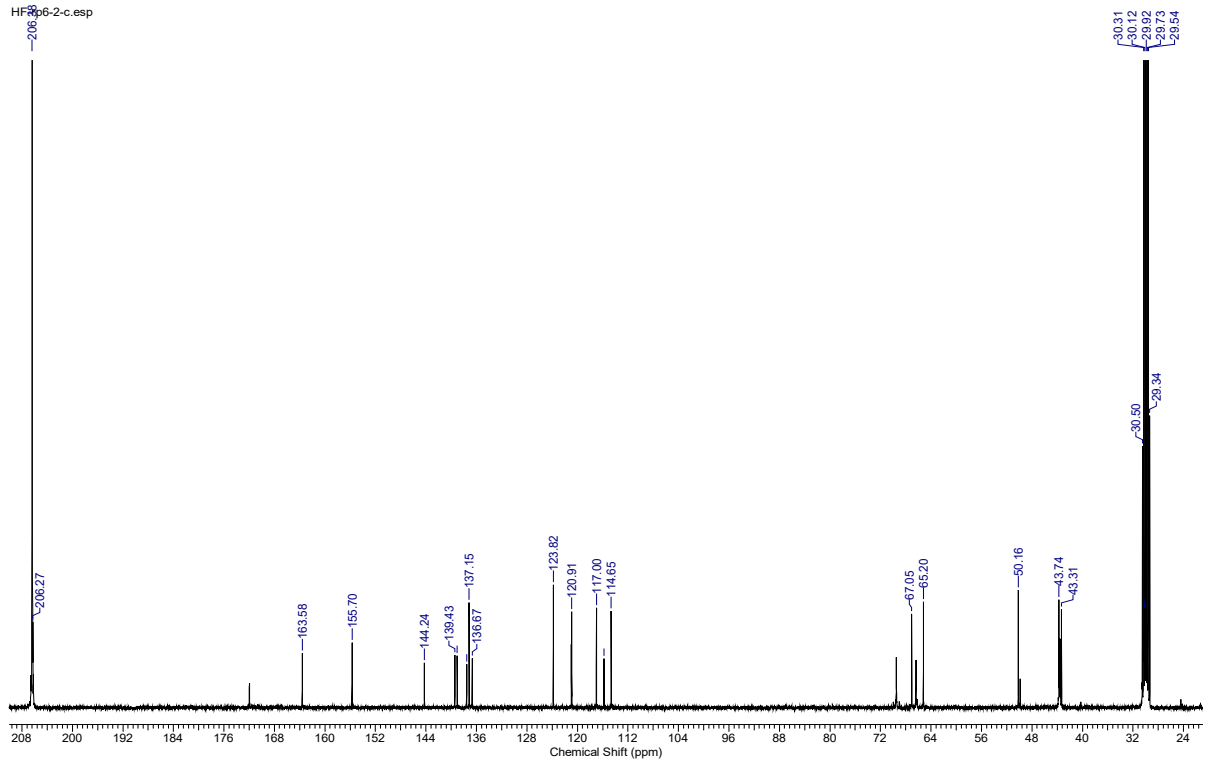
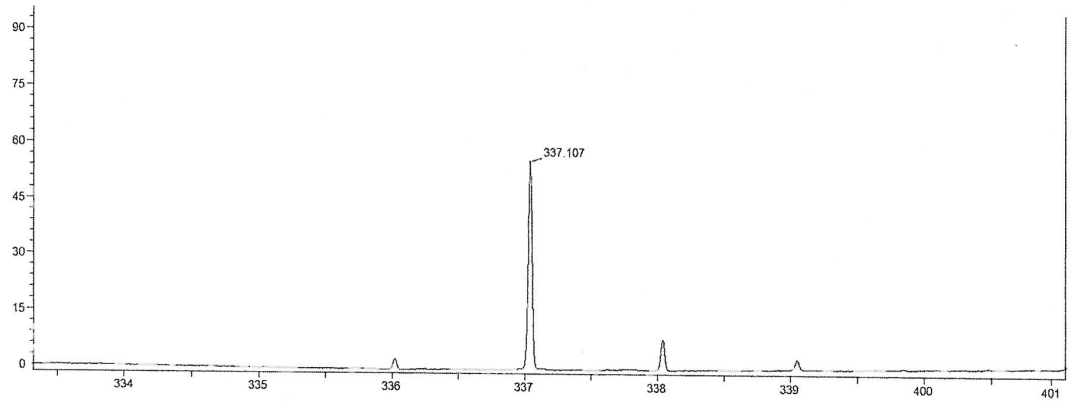


Figure S6. ^{13}C NMR NMR spectrum of hypoxylonol F (**2**)

[Mass Spectrum]
Data : KIST_known_002
Sample : -
Note : -
Inlet : Direct
Ion Mode : EI+



[Theoretical Ion Distribution]
Molecular Formula : C20 H17 O5 $[M+H]^+$
(m/z 337.1074, MW 337.3511)
Base Peak : 337.1074, Averaged MW : 337.3445(a), 337.3496(w)

m/z	INT	
337.1074	100.0000	**********
338.1081	21.9650	**********
339.1088	2.2852	**
400.1094	0.1975	
401.1102	0.0182	

Figure S7. HRMS of hypoxylonol F (**2**)

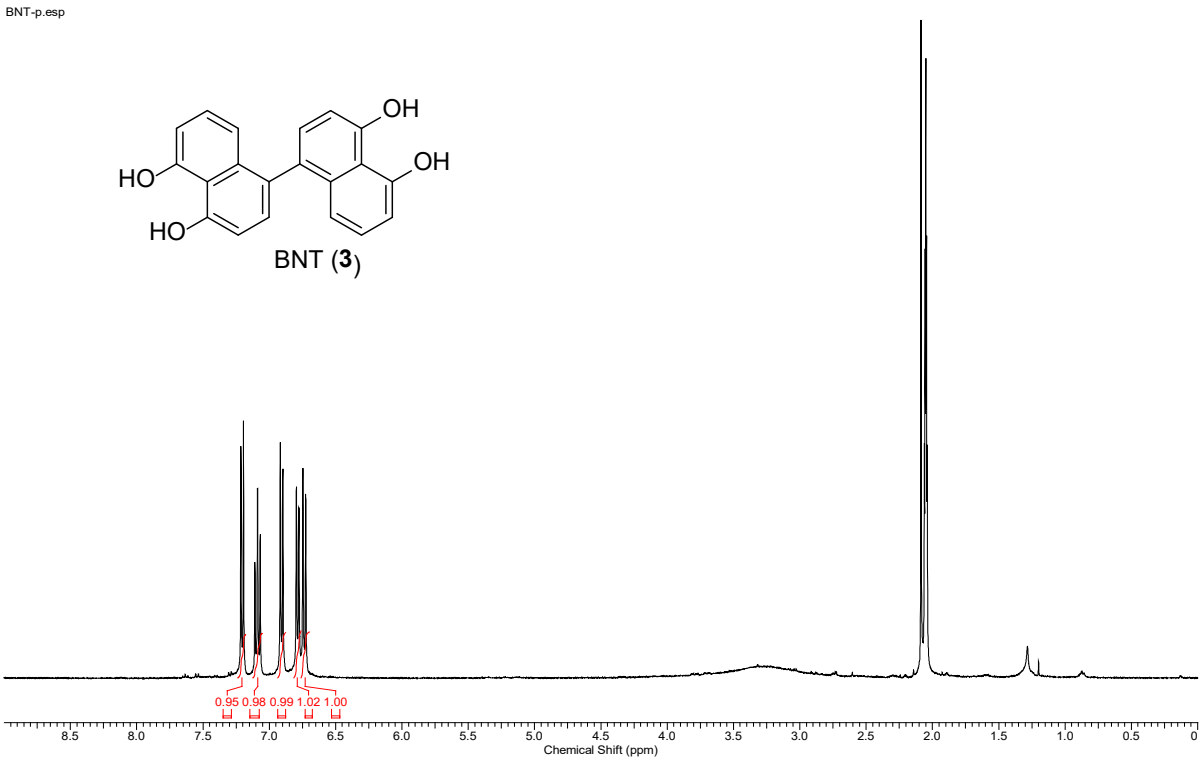


Figure S8. ^{1}H NMR NMR spectrum of BNT (**3**)

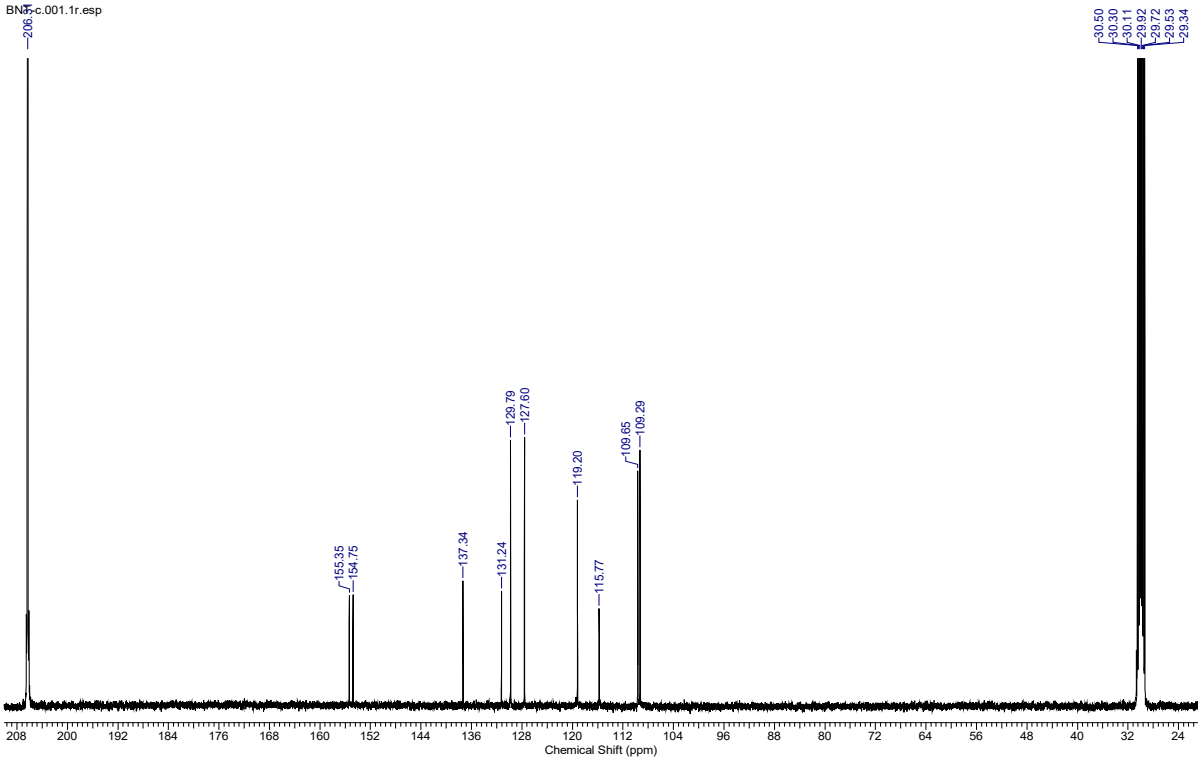
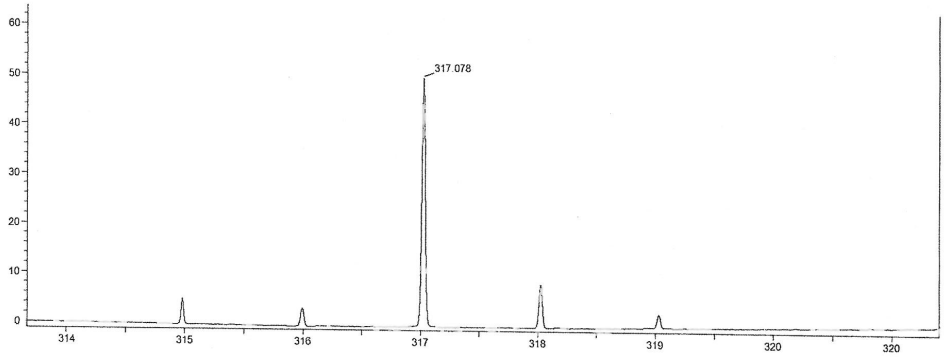


Figure S9. ^{13}C NMR NMR spectrum of BNT (**3**)

[Mass Spectrum]
Data : KIST_known_003
Sample : -
Note : -
Inlet : Direct
Ion Mode : EI+



[Theoretical Ion Distribution]
Molecular Formula : C20 H13 O4 $[M+H]^+$
(m/z 317.0778, MW 317.3199)
Base Peak : 317.0778, Averaged MW : 317.3045(a), 317.3156(w)

m/z	INT	
317.0778	100.0000	**********
318.0792	20.8460	**********
319.0808	1.9912	**
320.0821	0.2237	
321.0834	0.0197	

Figure S10. HRMS of BNT (**3**)

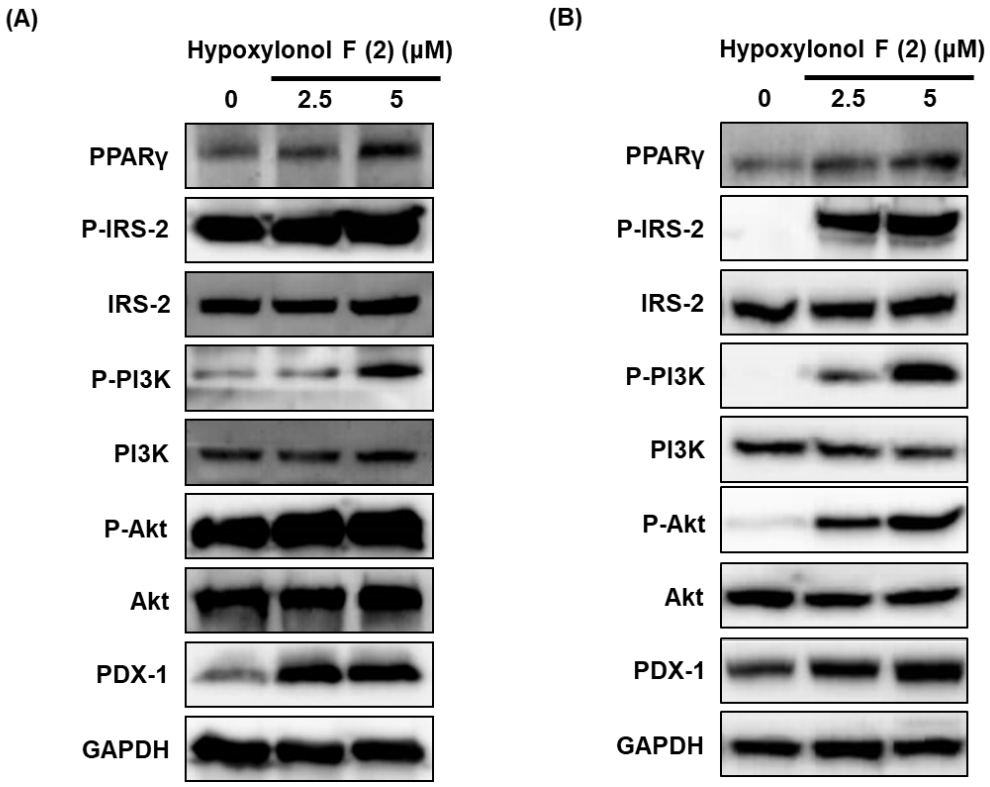


Figure S11. Effect of hypoxylonol F (**2**) on the protein expression levels of PPARγ, P-IRS-2, IRS-2, P-PI3K, PI3K, P-Akt (Ser473), Akt, and PDX-1 in INS-1 cells.