Iridoid glycoside isolated from *Wendlandia glabrata* and the role of its enriched fraction in regulating AMPK/PEPCK/G6Pase signaling pathway of hepatic gluconeogenesis

Pranamika Sarma[1,2], Simanta Bharadwaj[1], Deepsikha Swargiary[1,3], Semim Akhtar Ahmed [1,3], Yunus Sheikh[1], Sagar Ramrao Barge[1], Prasenjit Manna[3,4], Narayan Chandra Talukdar[1,5], Jayanta Bora[6], Jagat Chandra Borah[1,3 *]

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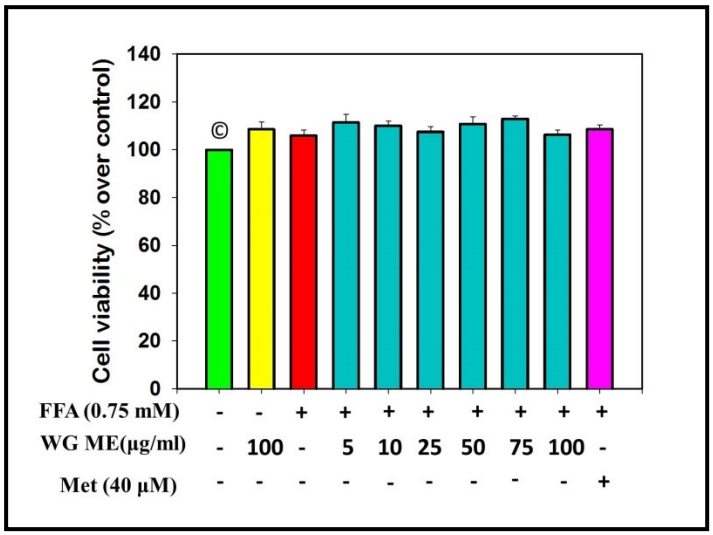
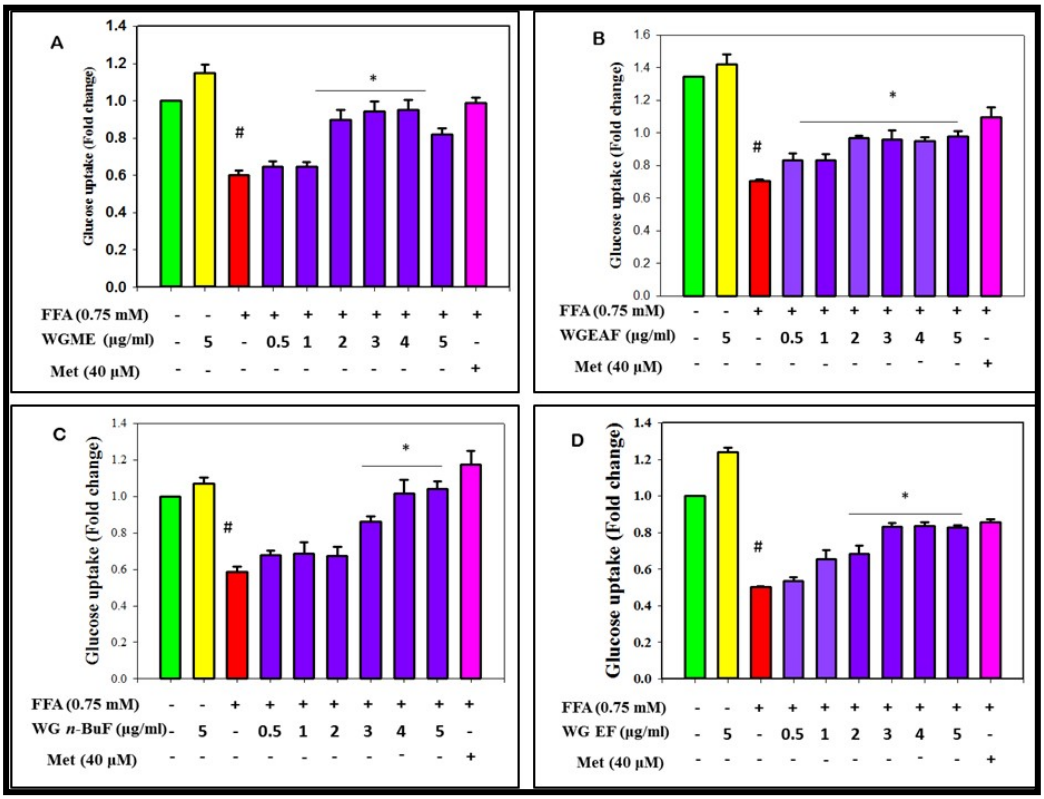
Supplementary Data :

Supplimentary S1. Effect of the WG Crude methanol extract on the cell viability in palmitate (designated as free fatty acid,FFA) treated hepatocytes.



Supplementary S2. Effect of the WG A)Crude methanol extract B) Ethyl acetate fraction C)*n*-Butanol fraction D) Enriched fraction on Glucose uptake in palmitate (PA) treated hepatocytes. Cells were pre-treated with different concentrations (0.5-5 µg/mL for 2 h followed by PA (0.75 mM) exposure for the next 18 h. "#" denotes the significant difference from untreated/control ($p\#<0.05$) and "*" denotes the significant difference from PA-treated groups ($p^*<0.05$).



Supplementary S3 : Spectral data of the isolated compounds 3-6

Procyanidin A2: Compound **3**, identified as Procyanidin A2, was isolated as a white powder. The HRMS data showed a deprotonated molecular ion peak $[M-H]^-$ at 575.19788 and a sodiated peak [M+Na]+ at 599.11359, corresponding to molecular formula $C_{30}H_{25}O_{12}$ (exact mass 577.1346).^{1}H NMR (600 MHz,MeOH-D4) : 4.43 (d,3.36), 3.66(s), 6.03 (d,2.28), 6.09(d,2.28), 6.82(d,3.12), 6.99(d,1.86), 7.01(dd,1.86,8.28), 4.66(s), 4.08(d,3.36), 2.80(m), 2.98(m), 6.11(s), 7.04(d,2.10), 6.84(d,3.0), 7.03(m). ^{13}C NMR: 101.0, 28.5, 156.5, 152.8, 150.9, 95.0, 96.8, 80.3, 66.6, 105.8.

Cinnamtannin B-1: Compound **4** found to be Cinnamtannin B-1. HRMS mass spectral data presents deprotonated [M − H]– ion at m/z value 863.30469 and a [M + Na]+ ion at 887.17603, referring to the molecular formula $C_{45}H_{36}O_{18}$ (864.7500 g/mol). UV-vis absorption λ_{max} MeOH at 280 nm.^{1}H NMR and ^{13}C NMR data (600 MHz, MeOH-D4) .^{1}H NMR : 3.30(d,3.42),4.17(d,3.35) 5.99(br s),3.32(d,2.79),3.88(br), 4.41 (br s), 4.58 (br s), 5.28 (br s), 5.73 (br s), 5.82 (br s), 5.99(d,2.24), 6.04(d,2.21),6.84(dd ,1.22,7.58), 6.85(7.59),6.87 (d, 4.75),7.13(br s), 7.05(br s), 7.34 (s) ,^{13}C NMR : 156.41, 155.35 , 154.38, 152.76, 150.41, 149.69 , 145.20, 144.88 , 144.49, 131.79, 131.05 , 130.39, 119.98 , 119.19 , 115.34 , 114.77 , 114.39 , 107.48, 105.00, 103.59, 102.89 , 98.55, 96.92 , 95.19, 95.05, 94.66 , 78.88, 77.47, 71.18, 66.13 , 65.77 , 36.90 , 28.47, 27.47.

Rutin: Compound **5** Rutin, was isolated as a yellow solid. The UV spectrum showed two maxima (λ_{max} MeOH) 256 and 357 nm. HRMS analysis presented m/z values of deprotonated ion at 609.14697 which predicted the molecular formula $C_{27}H_{30}O_{16}$.^{1}H NMR: 2.48 (d, 3.10), 3.06-4.36(m), 5.14(d, 2.01), 6.18(d, 0.92), 6.37(d, 0.94), 6.81(d, 1.01), 7.51(m). ^{13}C NMR : 177.01, 164.15, 161.46,

156.01, 148.47, 145.47,1 33.46, 121.06, 121.02, 116.43,115.37,107.84, 104.34,101.36,98.82,93.82, 100.92, 76.85-67.32 .

Isoorientin -6″-O-glycoside: Comound **6**, Isolated as yellow solid, showed m/z values in mass spectra as deprotonated molecular ion peak at 595.2200 and sodium adduct ion at 619.1245. UV-Vis absorption spectrum matches with that of isoorientin with λ_{max} MeOH values 250 nm and 350 nm.1H NMR (600 MHz,MeOH-D4) 1H NMR: 7.78(m), 6.91(d, 3.16), 6.89(d, 7.50), 6.42(s), 5.28(d, 7.70), 3.73-3.92 (m).13C NMR: 178.28,162.02,161.72, 156.96,148.40,146.10, 133.75, 121.97,115.96,111.711,101.01,100.00, 98.32, 93.13,60.50-78.70 . Spectral data indicate towards the molecular formula $C_{26}H_{28}O_{16}$ with mass 596.5 g/mol.

Supplementary S4: Behavioural observation of swiss albino mice in WG EFr (2000 mg/kg BW) treated and control group (n=6, 14 days)

Observation	Control group	Test group
Lethality	Not found	Not found
Convulsions	Not found	Not found
Straub tail	Not found	Not found
Sedation	Not found	Not found
Excitation	Normal	Normal
Jumps	Not found	Not found
Loss of balance	Not found	Not found
Abnormal writhes	Not found	Not found
Piloerection	Not found	Not found
Stereotypies	Not found	Not found
Head twitches	Not found	Not found
Scratching	Not found	Not found
Abnormal respiration	Not found	Not found
Loss of righting reflex	Not found	Not found
Loss of corneal reflex	Not found	Not found
Defecation	Normal	Normal
Salivation	Normal	Normal
Lacrimation	Not found	Not found
Aggressiveness	Normal	Normal

Supplementary S5: Table for body weight and feed consumption of Test animals

Group	Initial average body weight per group (g)	Final average body weight per group (g)	Average feed consumption per group (g)
Normal control	29.54 ±1.73	29.60±2.98	3.50 ± 0.05
Diabetic control	28.80± 2.07	32.81±4.04	4.2 ±0.20
Positive control	27.15± 1.22	28.78±1.98	3.7 ± 0.09
EF (10)	28.86± 2.02	26.18±1.13	3.3 ± 0.07
EF (25)	28.22± 1.20	27.33±1.59	3.8 ± 0.12
EF(50)	27.67± 1.21	26.30±1.13	3.6 ± 0.02

Supplimentary S6.Full length blots