Figure S1. Chemical structure of FX and changes in GSDMC, GSDMD, and GSDME in response to FX. (A) Chemical structure of FX. (B) Western blot analysis was performed to evaluate and quantified changes in GSDMC, GSDMD, and GSDME in response to FX treatment in pancreatic cancer cells. *P<0.05, **P<0.01, ***P<0.001 vs. 0 μM). FX, fucoxanthin; ns, not significant; GSDM, gasdermin.

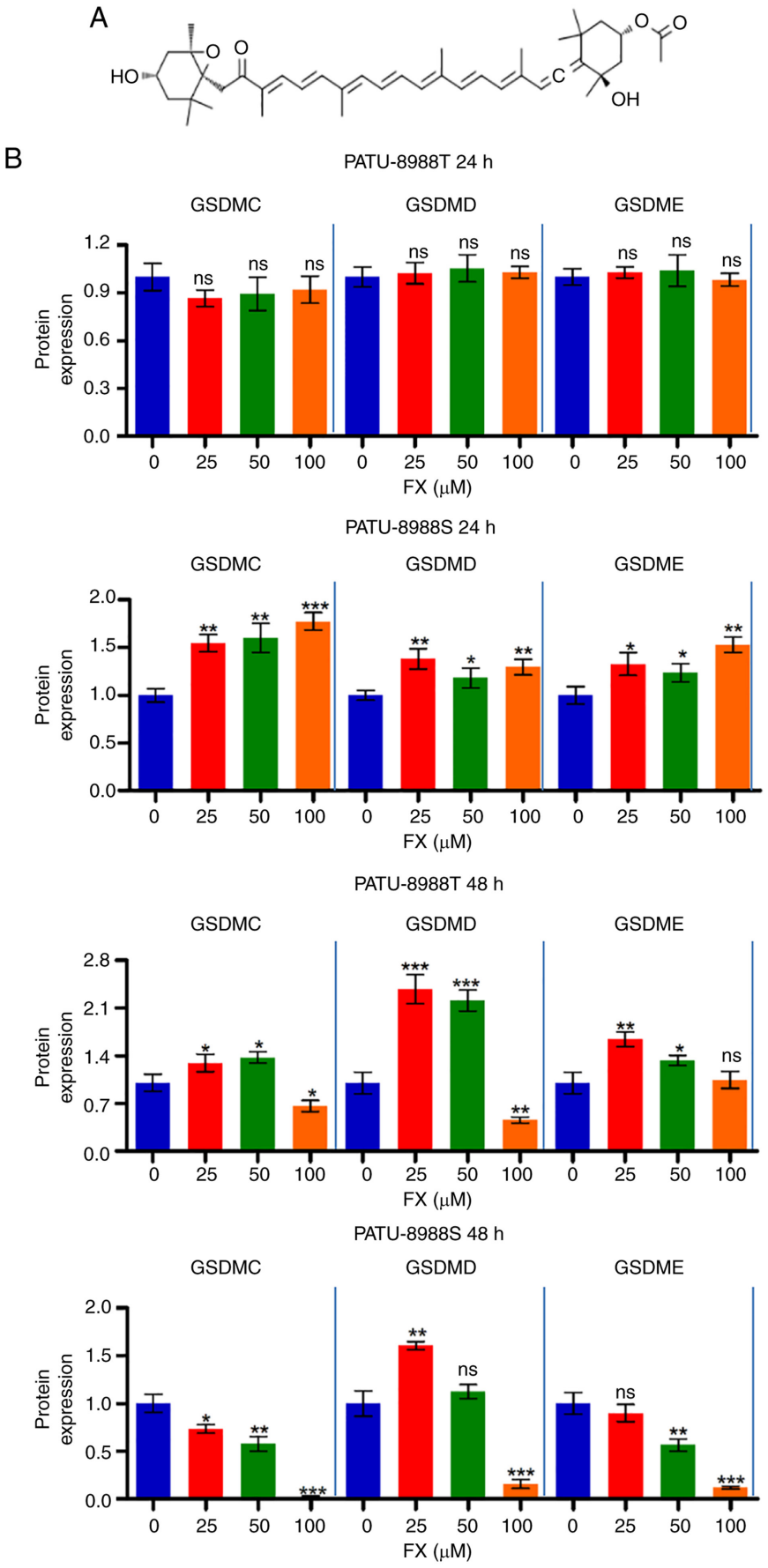

Figure S2. FX impairs mitochondrial homeostasis and causes oxidative stress in PC cells. (A) Changes in OCR in FX-treated PC cells were detected using XFe96 extracellular flux analyzer. (B) Changes in basal respiration, ATP production and maximal respiration in FX-treated PC cells. (C) ROS levels were detected in PC cells stained with DCFH-DA using flow cytometry. (D) Mitochondrial ROS production was measured in PC cells stained with MitoSOX via flow cytometry. (E) PC cells were stained with DCFH-DA and ROS levels were detected using fluorescence microscopy (magnification was 200 times). (F) PC cells were stained with Mitosox and mitochondrial ROS production was detected by fluorescence microscopy. (G) Changes in L-OPA1, S-OPA1 and S-OPA1/L-OPA1 in response to FX treatment in PC cells. *P<0.05, **P<0.01, ***P<0.001 vs. 0 μM. FX, fucoxanthin; PC, pancreatic cancer; OCR, oxygen consumption rate; ROS, Reactive oxygen species; L-OPA1, Long; S-OPA1, Short-OPA1; oligo, Oligomycin; FCCP, Carbonyl cyanide 4-(trifluoromethoxy)phenylhydrazone; Anti/Rot, Antimycin a1/ Rotenone; ns, no significant differences.

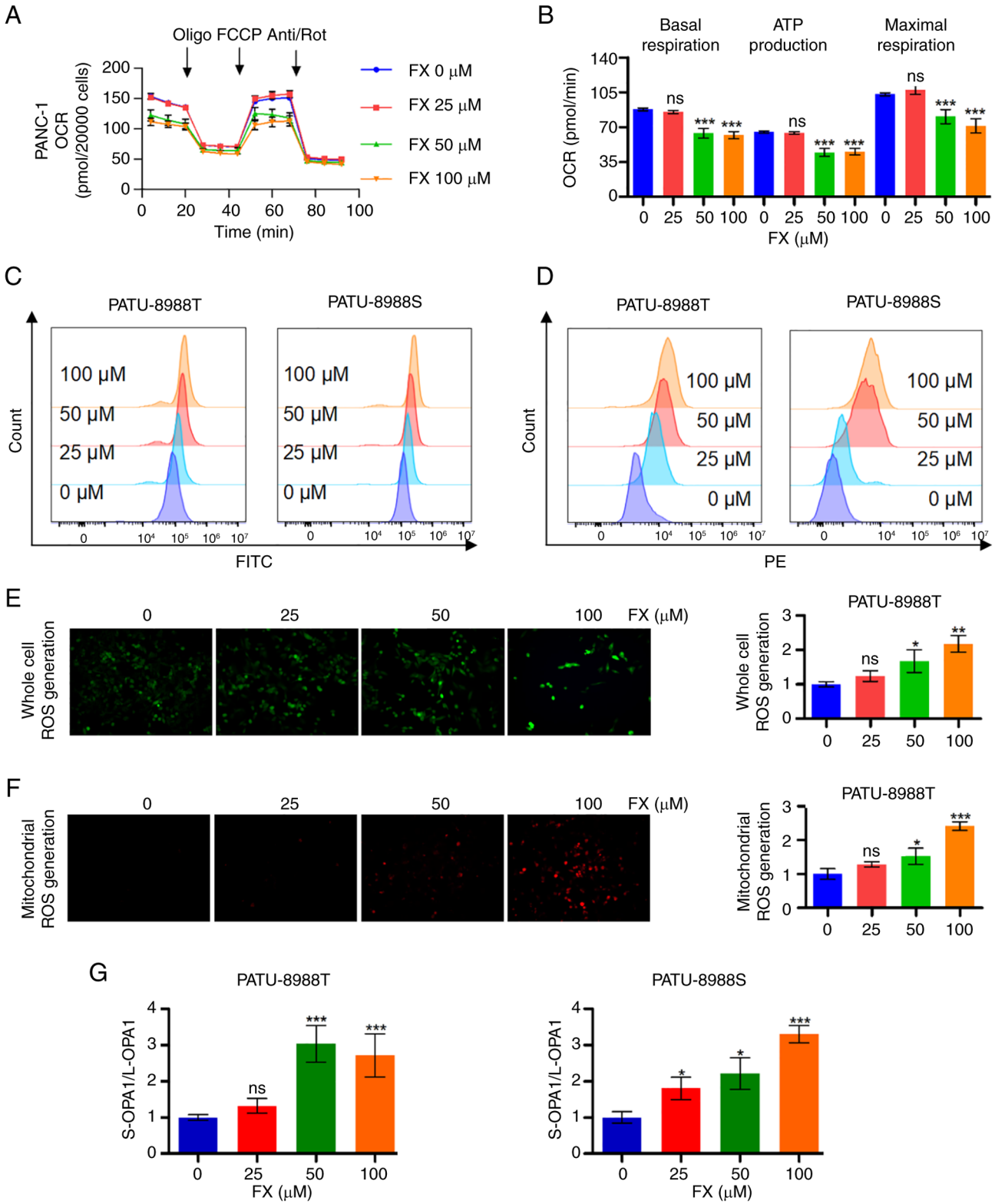

Figure S3. FX facilitates aerobic glycolysis in PC cells. (A) Changes in ECAR in FX-exposed PC cells were measured using the XFe96 extracellular flux analyzer. (B) Changes in glycolysis rate and capacity in FX-treated PC cells. *P<0.05, **P<0.01, ***P<0.001 vs. 0 μM). FX, Fucoxanthin; ECAR, Measurement of extracellular acidification rate; PC, Pancreatic cancer; Glc, Glucose; oligo, Oligomycin; 2-DG, 2-deoxy-D-glucose; ns, no significant.

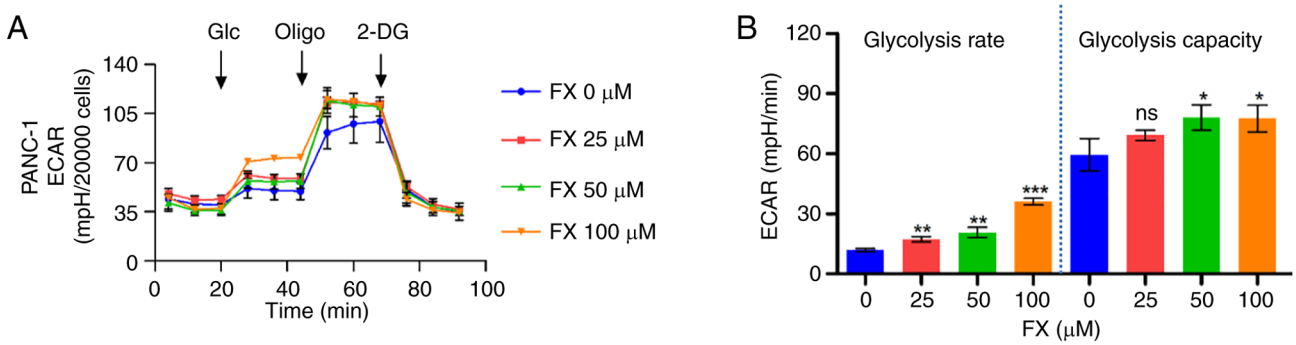

Figure S4. FX sensitizes PC cells to GL conditions by enhancing decrease in GSH/GSSG ratio. (A) Cell viability was detected by MTT assay following 24 h treatment with 25 μM FX alone or under GL conditions in the presence or absence of 7.5 mM GSH. (B) Change in GLS expression in PC cells following FX treatment was analyzed by western blotting. (C) The western blot results of GLS was quantified and showed. *P<0.05, **P<0.01, ***P<0.001 vs. 0 μM). FX, Fucoxanthin; PC, Pancreatic cancer; GL, Glucose limited; GSH, Reduced glutathione; GSSG, Oxidized glutathione; SLC31A1, Solute carrier family 31 member 1; sh, short hairpin; ns, no significant; OD, Optical density; GLS, Glutaminase.

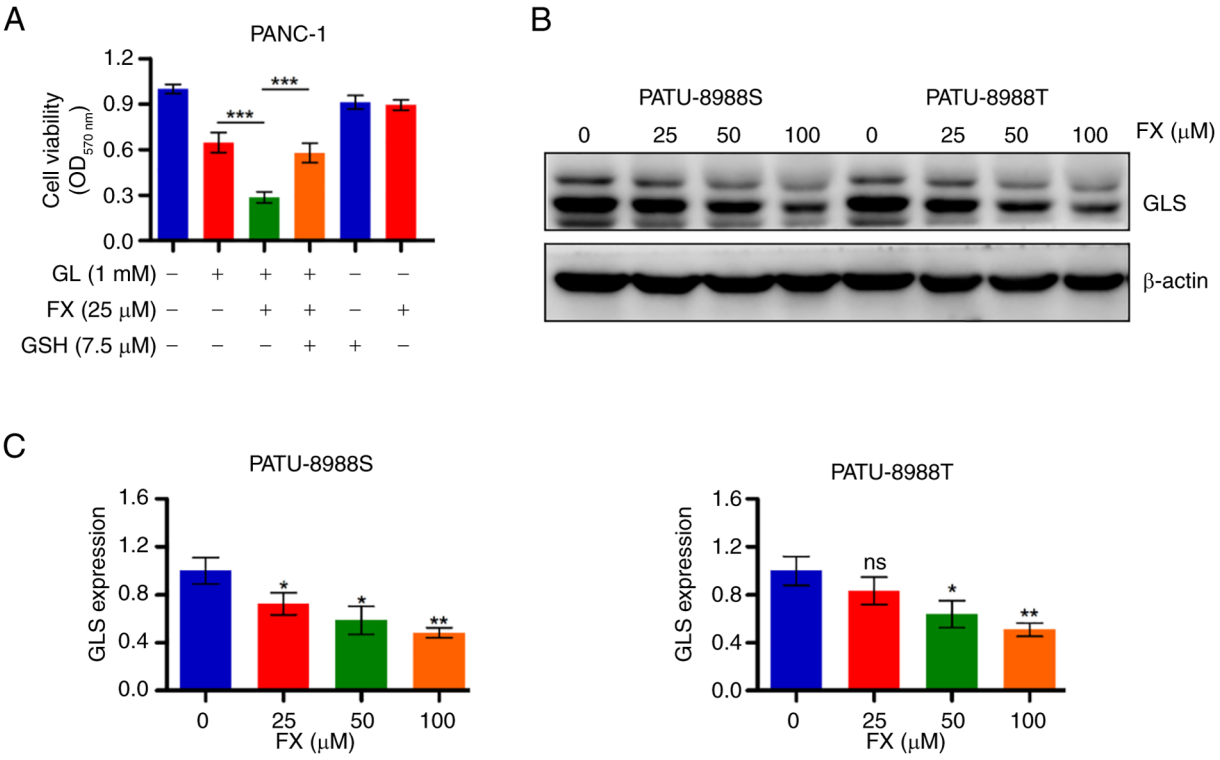

Figure S5. SLC31A1 is induced by FX in PC cells. (A) Changes in SLC31A1 expression in PC cells following 48 h FX treatment were analyzed by western blotting. (B) Efficiency of SLC31A1 silencing in PATU-8988T cells was confirmd by western blot assay. *P<0.05, **P<0.01, ***P<0.001 vs. 0 μM. SLC31A1, Solute carrier family 31 member 1; FX, Fucoxanthin; PC, Pancreatic cancer; sh, short hairpin; NC, Negative control.