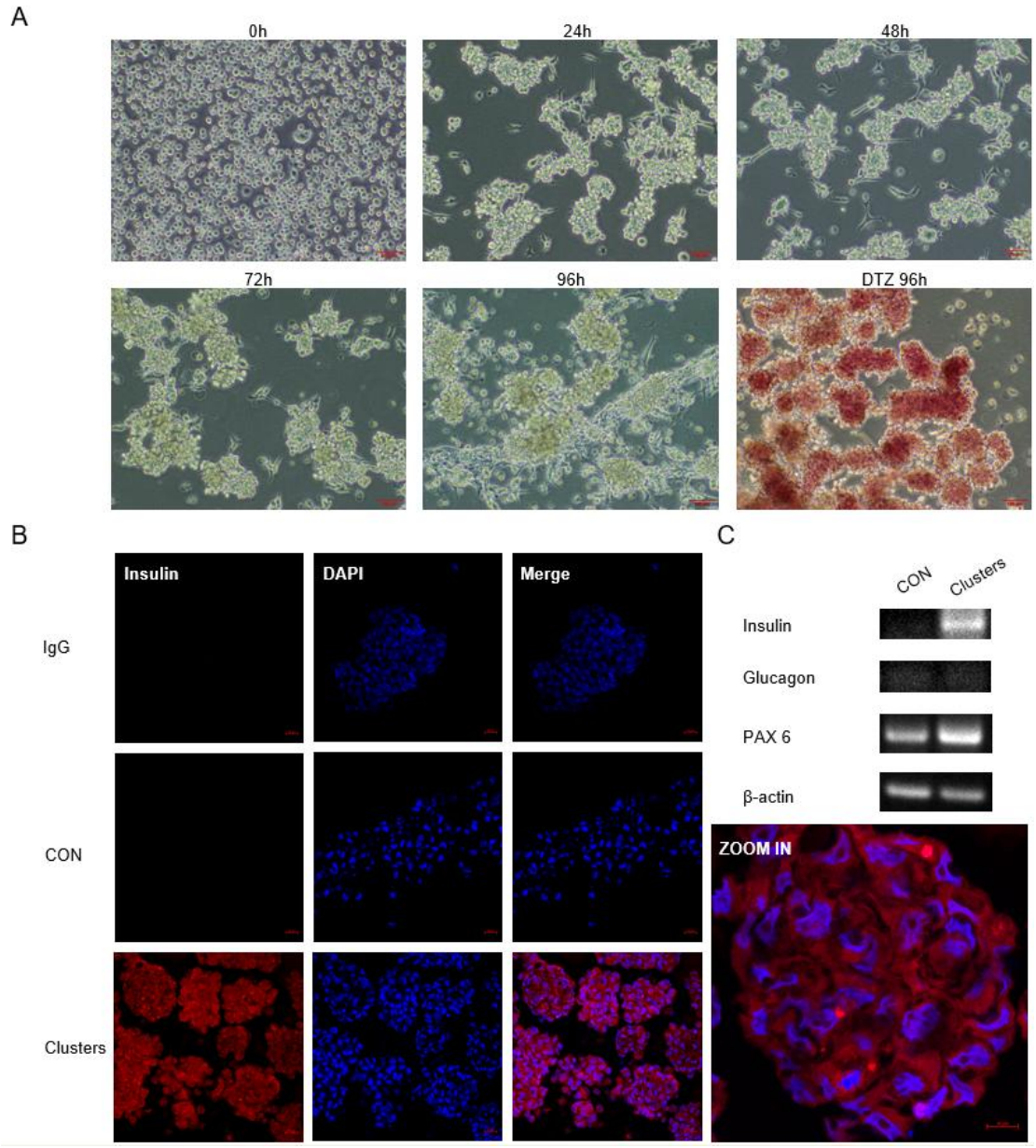
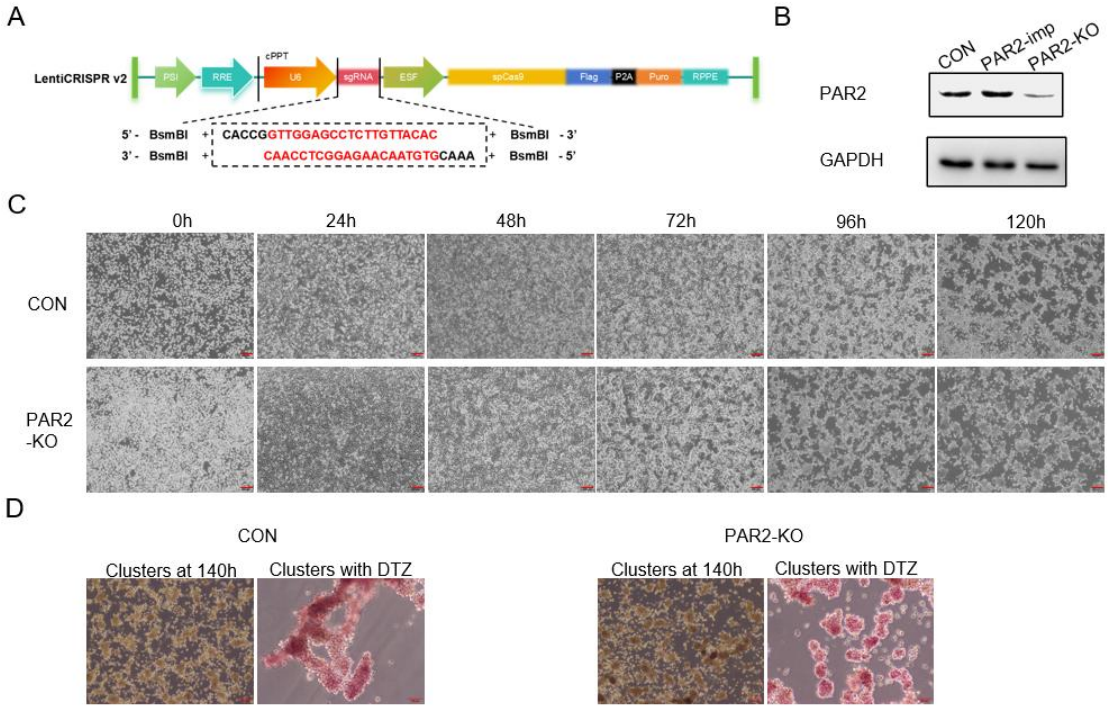
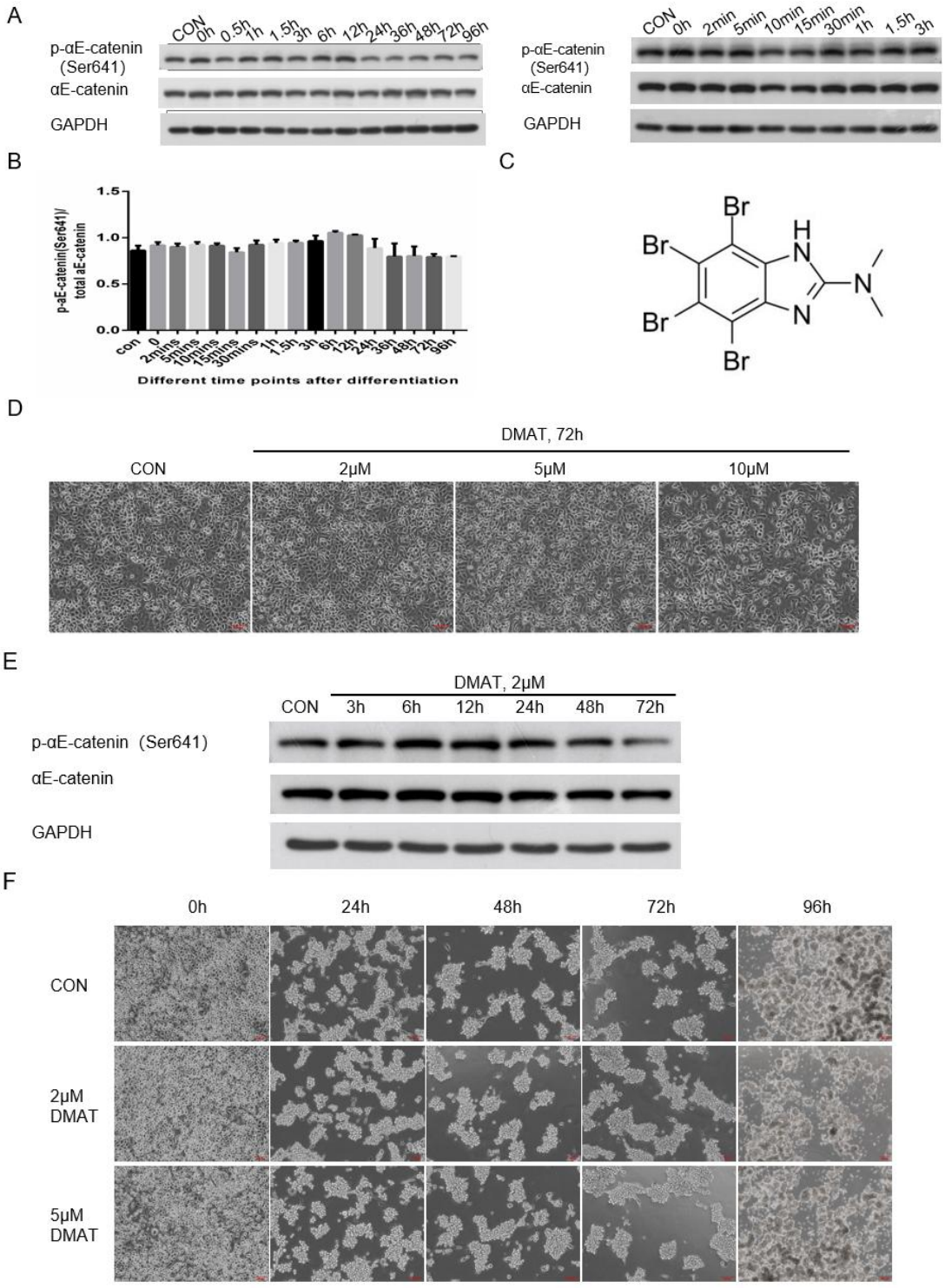


Supplementary Figure 1 Trypsin can induce the aggregation and differentiation of PANC-1 into functional islet-like clusters. (A) Representative images of PANC-1 at 5-time points after digestion with trypsin and representative image of DTZ staining of cell clusters formed at 96 h, with the objective lens magnification of 10X; (B) Representative immunofluorescence images of Insulin and DAPI signal in differentiated cell clusters, with the objective magnification of 20X, while the magnified of 63X; (C) PCR detection reflected the changes of expression of the transcripts for mature islet related genes, Insulin, Glucagon and PAX6 before and after differentiation. CON: Control.

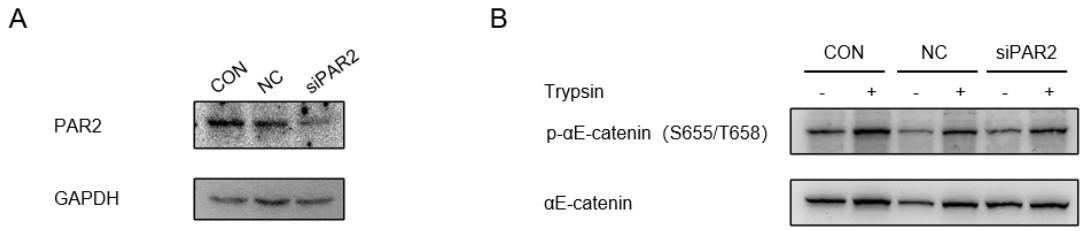


Supplementary Figure 2 PAR2 is not involved in the aggregation and differentiation of PANC-1 induced by trypsin. (A) The sequence of specific sgRNA targeting F2RL1 coding PAR2 and the construction diagram of core plasmid of CRISPR-Cas9 gene knockout system; (B) the expression of PAR2 protein was detected by western blot, reflecting knockout efficiency of F2RL1, PAR2-imp group: stable transgenic cells with F2RL1 gene knockout by CRISPR-Cas9 system, PAR2-KO: monoclonal cell line screening from stable transgenic cells. (C) Representative images of knockout cells at 5-time points after digestion with trypsin before and after F2RL1 knockout, with the objective lens magnification of 10X; (D) Representative images of DTZ staining of cell clusters formed at 96 h before and after differentiation upon knockout of F2RL1, with the objective lens magnification of 10X. CON: Control; KO: Knockout.



Supplementary Figure 3 Effect of Phosphorylation of αE-catenin at Ser641 and CK2 inhibitor DMAT on differentiation of PANC-1. (A) Representative western blot images reflecting protein expression of p-αE-catenin (Ser641) at different time points during PANC-1 differentiation induced by trypsin; (B) Statistical analysis of the gray value of western blot images using Image J and GraphPad Prism, reflecting the statistical differences of expression of p-αE-

catenin (Ser641); (C) Chemical structure of DMAT, the inhibitor of CK2; (D) Representative images of PANC-1 treated with 2, 5, 10 μM DMAT for 96 h, with the objective lens magnification of 10X; (E) Western blot images showed the expression of p-αE-catenin (Ser641) of PANC-1 cells treated with 2 μM DMAT for different time. (F) Representative images of PANC-1 after 72 h treatment with 2 μM and 5 μM DMAT at different time points after digestion with trypsin, with the objective lens magnification of 10X. CON: Control; KO: Knockout; DMAT:2-Dimethylamino-4,5,6,7-tetrabromo-1H-benzimidazole.



Supplementary Figure 4 PAR2 has no effects on the phosphorylation of αE-catenin (Ser655/T658) mediated by trypsin. (A) Representative western blot images reflecting the efficiency of siRNA knockdown of PAR2; (B) Representative western blot images reflecting the differences of trypsin-mediated αE-catenin (Ser655/Thr658) phosphorylation before and after knockdown. CON: Control; NC: negative control; siPAR2: siRNA knockdown of PAR2.