Supplementary Experimental Procedures

Genotyping

βTCFDN mice were genotyped by PCR for presence of the transgene with the human TCF7L2 forward primer 5'-TCGCCTGGCACCGTAGGACA-3' and reverse primer 5'-GGATGCGGAATGCCCGTCGT-3' using the Mouse Genotyping Kit (KAPA). The primers for *Ins2-rtTA* genotyping are: Forward, 5-TAGATGTGCTTTACTAAGTCAT-3; Reverse: 5-GAGATCGAGCGGGCCCTCGATG-3.

The generation of Adenovirus

The wild type human TCF7L2 (TCF7L2WT) and dominant negative TCF7L2 (TCF7L2DN) cDNA sequences were amplified by PCR with the Q5 High-Fidelity DNA Polymerase (New England Biolabs), with primers to append NotI and XhoI restriction sites at the 5' and 3' ends, respectively: TCF7L2WT-F, 5'-CGCGCGGCCGCAAAATGCCGCAGCTGAACG-3'; TCF7L2WT-R, 5'- CGGCTCGAGTTCTAAAGACTTGGTGACGAGC-3'; TCF7L2DN-F, 5'-CGCGCGGCCGCAAAATGGAAACGAATCAAAACAGCTCCTC-3'; TCF7L2DN-R, 5'-CGGCTCGAGTTCTAAAGACTTGGTGACGAGC-3'. The PCR products, purified by the Gel/PCR DNA Extraction Kit (FroggaBio) followed by agarose gene separation, were A-tailed with Taq Polymerase (New England Biolabs) according to the manufacturer's protocol, followed by ligation into the pGEM-T Easy vector (Promega) and routine amplification in *E. coli*. Clones that possess accurate sequences were verified by DNA sequencing (ACGT Corporation, Toronto). The TCF7L2DN and TCF7L2WT were then subcloned into the pShuttle-IRES-hrGFP-2 vector (Agilent Technologies) via the NotI/XhoI sites. The PmeI-linearized TCF7L2WT, TCF7L2DN, and empty shuttle construct (GFP control) were then recombined into the pAdEasy-

1 vector via transformation into BJ5183-AD-1 cells (Agilent Technologies) and subsequent homologous recombination. Recombinant DNA was then amplified in XL10-Gold *E. coli* cells (Agilent Technologies), linearized with PacI (New England Biolabs), then transfected into AD-293 cells, using Lipofectamine 2000 (Life Technologies). Primary virus particles were harvested 3 weeks following transfection through 4 cycles of freeze-thaw and used in further infect AD-293 cells for further amplification. After 3 successive rounds of virus amplification, adenovirus particles were purified using the Vivapure AdenoPACK 20 (Sartorius) and diluted in PBS. Adenovirus titers were determined by taking the absorbance at 260 nm. Virus infection efficiency was monitored by GFP expression.

Glucose stimulated insulin secretion (GSIS) and GLP-1 stimulated insulin secretion

Ins-1(832/13) cells seeded on a 12-well plate were washed with the KRB buffer (129 mM NaCl, 5 mM $NaHCO_3$, 4.8 mM KCl, 1.2 mM KH_2PO_4, 1.2 mM $MgSO_4$, and 2.5 mM $CaCl_2$), followed by the incubation with the KRB buffer with 2.8 mM glucose and 0.1% BSA for 30 min. Insulin secretion from Ins-1 cells was then performed in the KRB buffer containing 2.8 mM or 16.7 mM glucose with or without GLP-1 (7-37) (100 nM) at 37°C in a humidified incubator for 20 min. The supernatants were collected and detected by insulin enzyme immunoassay kit (EMD Millipore, Billerica, MA) according to the manufacturer's instructions. For assessing insulin secretion from mouse islets, ten hand-picked islets were utilized per each assay. The islets were pre-incubated with KRB containing 2.8 mM glucose for 30 min, followed by 20 min incubation with either 2.8 mM or 16.7mM glucose containing KRB, in the presence or absence of 100 nM GLP-1 (7-37). The supernatant fractions were collected for insulin level measurement, as we have reported previously (1).

The measurement of β-cell and α-cell mass and immunohistochemistry study

The pancreas was isolated and fixed in 10% paraformaldehyde, dehydrated, and embedded in paraffin. Pancreatic tissue sections were stained for insulin, glucagon, Pdx-1 and Nkx6.1. Pancreatic images were visualized using ScanScope CS (Aperio Technologies) and analyzed by using the ImageScope program (Aperio Technologies) as we described previously (2; 3). Briefly, entire pancreatic tissue areas from each of the pancreatic sections containing 4-6 segments, stained for insulin or glucagon were outlined, and the number of strong positive signals within those areas, representing insulin- or glucagon-positive cells, was determined as the function of the positive pixel count algorithm (Aperio Technologies). This algorithm was also used to detect the number of negative signals, which represented the total pancreatic area. Total β- and α-cell mass for each pancreas was determined as the product of the total cross-sectional β- or α-cell areas over total tissue area and the weight of the pancreas before fixation. The following primary antibodies were used: goat anti-Pdx-1 (gift of Dr. Chris Wright, 1:10,000), Nkx6.1 (F55A10-c, DSHB, 1:2000), insulin (Dako, 1:1000). After washing with PBS-Tween, sections were incubated with fluorescence-conjugated secondary antibody for 45 minutes at room temperature. Secondary Antibodies: anti-guinea pig IgG-Alexa 488 (Jackson Immunoresearch), anti-mouse and anti-goat IgG Alexa 647 (Life Technologies). Slides were counterstained with DAPI (Biotium), prior to mounting with fluorescence mounting medium (DAKO). Pdx-1 and Nkx6.1 positive islets cell counting was conducted utilizing the program Imagine Pro Plus.

Cell proliferation assay

Cell proliferation on Ad-TCF7L2WT and Ad-TCF7L2DN infected Ins-1 cells was determined with the MTT method, utilizing the 96 well cell culture plates (4). Approximately 10,000 Ins1

cells were distributed into each well. At indicated time intervals, the cells were incubated with the MTT dye for 1 h. Cell numbers were then determined by colorimetric measurement.

Supplementary Figure legend

Figure S1. The establishement of βTCFDN mouse model. A) A schematic of P_{TRE3G}-TCF7L2DN fusion gene. IRES, internal ribosome entry site. pA, polyadenylation site. rtTA, Tet-On Advanced transactivator. **B-C)** Visualization of mCherry expression by fluorescent microscope (B) and detection of TCF7L2DN expression by Western blotting (C) for Ins-1 cells co-transfected with P_{TRE3G}-TCF7L2DN and Tet-On3G (i.e. rtTA). **D)** Genotyping results of $TCF7L2DN_{Tet}$ founders. **E-F)** Detection of mCherry expression in *βTCFDN* mouse islets (E) and dispersed islet cells (F).

Figure S2. Inducing TCF7L2DN expression in adult *βTCFDN* mice generates only modest metabolic defect. A) Western blotting shows the detection of TCF7L2DN in *βTCFDN* generated by mating $TCF7L2DN_{Tet}$ founder 4 with *Ins2-rtTA*. **B)** TCF7L2DN expression cannot be detected when adult *βTCFDN* mice were fed with diet that does not contain doxycycline (Doxy). **C)** Reduced endogenous *TCF7L2* mRNA expression in *βTCFDN* islets, assessed by qPCR. **D-E)** The lack of significant defect in *βTCFDN* mice in response to OGTT and IPITT. **F)** Plasma insulin levels before and 15 min after oral glucose challenge. **G-H)** Assessment of β-cell **(G)** and α-cell **(H)** mass. **I)** qPCR assessment for a panel of β-cell specific genes. Panel A, B/C, D-H, and I were performed with four sets of mice, respectively. For panels D-F, n=4. For panel C and I, n=3. Mice were fed with Doxycycline (Doxy) at the age of 6 w. The age of mice for each panel were indicated.

Figure S3. Inducing TCF7L2 expression in *βTCFDN* mice immediately after weaning generates only modest metabolic defects. A) Body weight comparison at the age of 5 wks. **B)** No significant defect in response to OGTT. **C)** Comparison of blood glucose levels before and 15 min after oral glucose gavage. **D)** Comparison of serum insulin levels before and 15 min after glucose gavage. **E)** Assessment of β-cell mass. For panel A-D, n=4. For panel E, n=3.

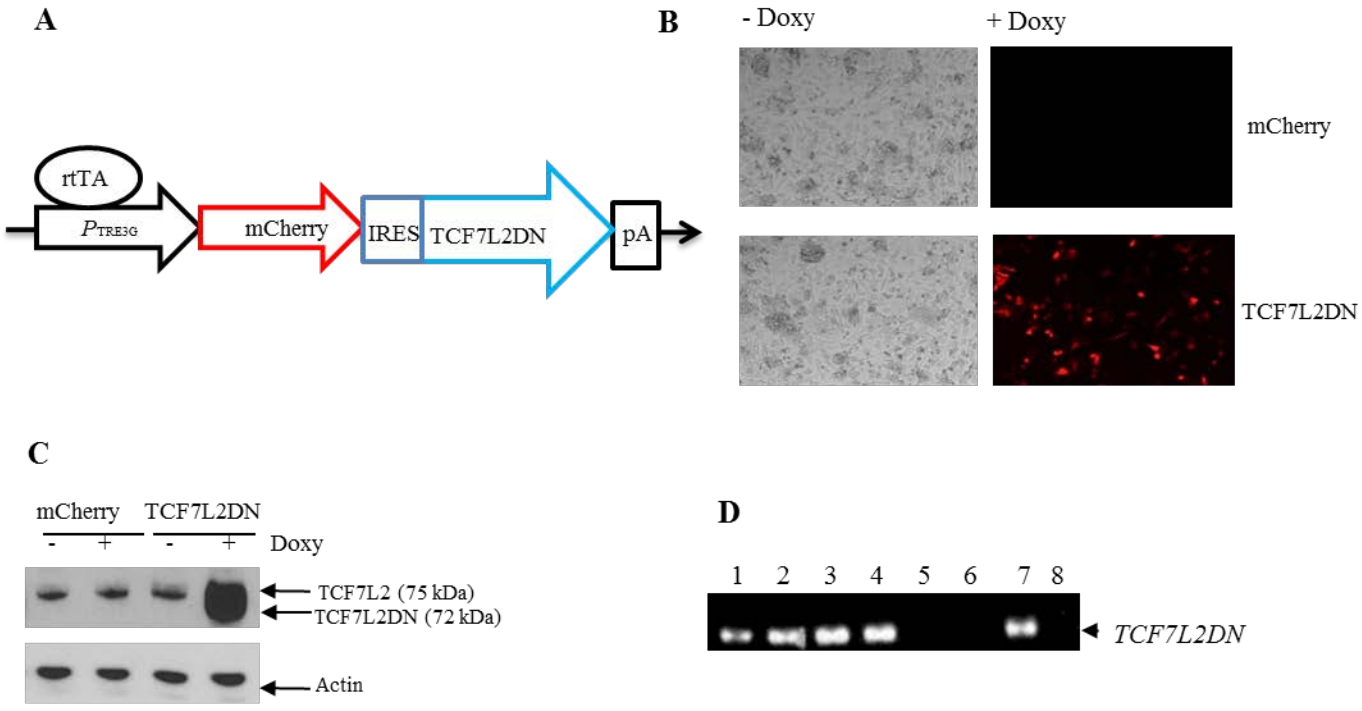
Supplementary Method References

1. Shao W, Wang Z, Ip W, Chiang YT, Xiong X, Chai T, Xu C, Wang Q, Jin T: GLP-1(28-36) improves beta-cell mass and glucose disposal in streptozotocin-induced diabetic mice and activates cAMP/PKA/beta-catenin signaling in beta-cells in vitro. Am J Physiol Endocrinol Metab 2013;304:E1263-1272
2. Soltani N, Qiu H, Aleksic M, Glinka Y, Zhao F, Liu R, Li Y, Zhang N, Chakrabarti R, Ng T, Jin T, Zhang H, Lu WY, Feng ZP, Prud'homme GJ, Wang Q: GABA exerts protective and regenerative effects on islet beta cells and reverses diabetes. Proceedings of the National Academy of Sciences of the United States of America 2011;108:11692-11697
3. Wang Q, Brubaker PL: Glucagon-like peptide-1 treatment delays the onset of diabetes in 8 week-old db/db mice. Diabetologia 2002;45:1263-1273
4. Wang P, Branch DR, Bali M, Schultz GA, Goss PE, Jin T: The POU homeodomain protein OCT3 as a potential transcriptional activator for fibroblast growth factor-4 (FGF-4) in human breast cancer cells. The Biochemical journal 2003;375:199-205

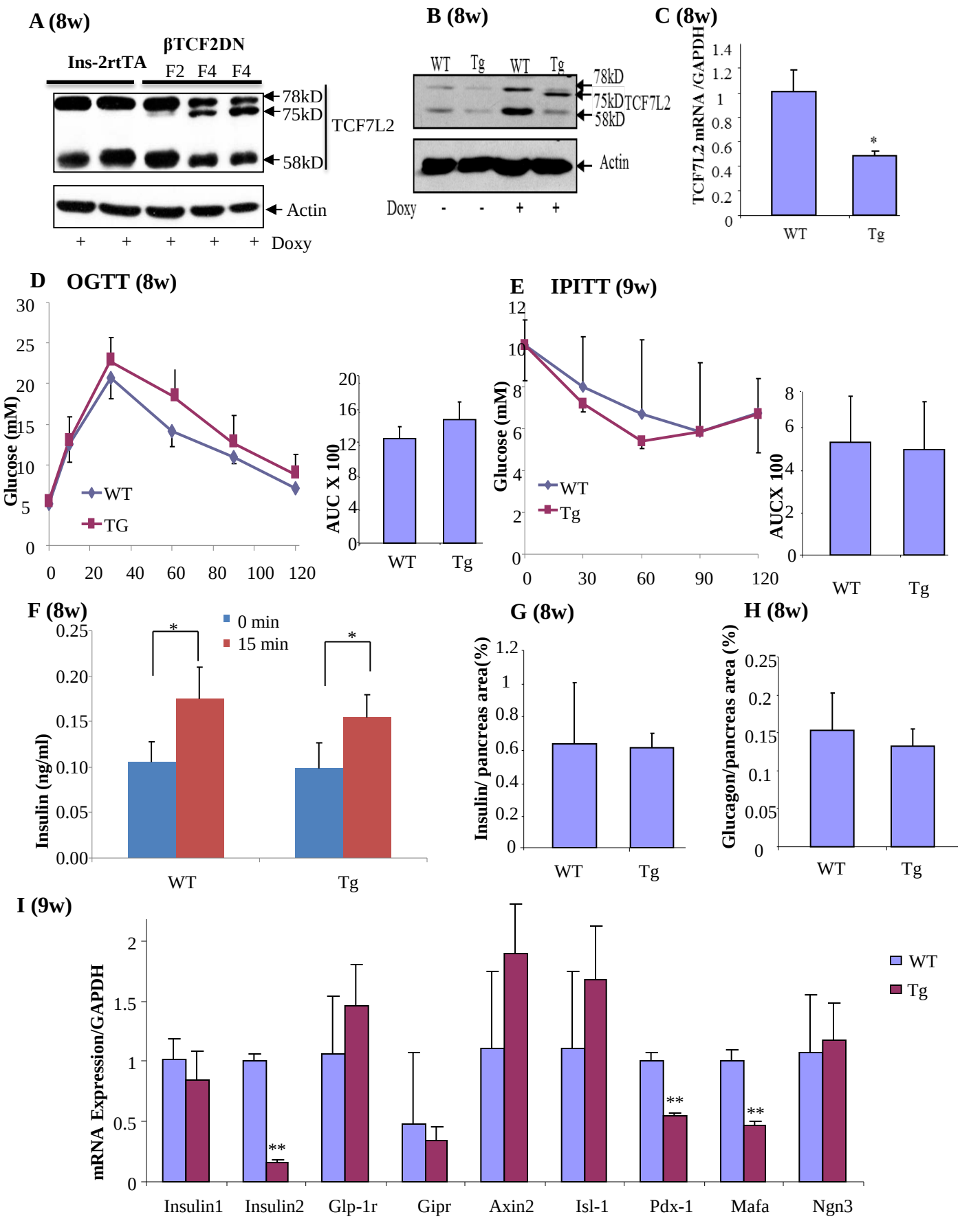
Supplementary Table 1 Nucleotide primers utilized in this study

		Product (bp)
Insulin1	Forward GCAAGCAGGTCATTGTTCCA Reverse CCAAGGTCTGAAGATCCCCG	263
Insulin2 (rat)	Forward GCAAGCAGGTCATTGTTCCA Reverse CTTGTGGGTCCTCCACTTCG	209
Insulin 2 (mouse)	Forward CATCAGCAAGCAGGAAGCCTATCT Reverse GCTGGTGCAGCACTGATCT	337
Axin2	Forward TCCTGACCAAACAGACGACG Reverse ACCTCTGCTGCCACAAAACT	212
Glp-1r	Forward GGGTCTCTGGCTACATAAGGA Reverse AAGGATGGCTGAAGCGATGAC	178
Gipr	Forward AACCATCTTGATCAATTTCCTCATC Reverse TTTCAAAGGCCAGTTTGGCG	218
Mafa for mouse	Forward ACAGAAAGAAGTCGGGTGCG Reverse GCACATTCTGGAGAGCGAGA	224
Mafa for rat	Forward GTATCCATGTCCGTGCGGG Reverse CTTGTACAGGTCCCGCTCCT	237
Pdx-1	Forward CAGTGGGCAGGAGGTGCTTA Reverse TCCACTTCATGCGACGGTTT	208
Ngn3	Forward TCGGGAGAACTAGGATGGCG Reverse GTTTGCTGAGTGCCAACTCG	252
Isl-1	Forward TGAGGGTTTCTCCGGATTTG Reverse TGAAGCCTATGCTGCACTTG	171
Tcf7l2 (mouse)	Forward GCATCCCTCACCCGGCCATC Reverse GCCACCTGCGCCCGAGAATC	243
Gapdh (rat)	Forward AGCTCATTTCCTGGTATGACAA Reverse GGTATTCGAGAGAAGGGAGGG	258
Gapdh (mouse)	Forward GACCACAGTCCATGCCATCA Reverse TGAAGTCGCAGGAGACAACC	335

Supplementary Figure 1



Supplementary Figure 2



Supplementary Figure 3