Supplemental Figures and Tables

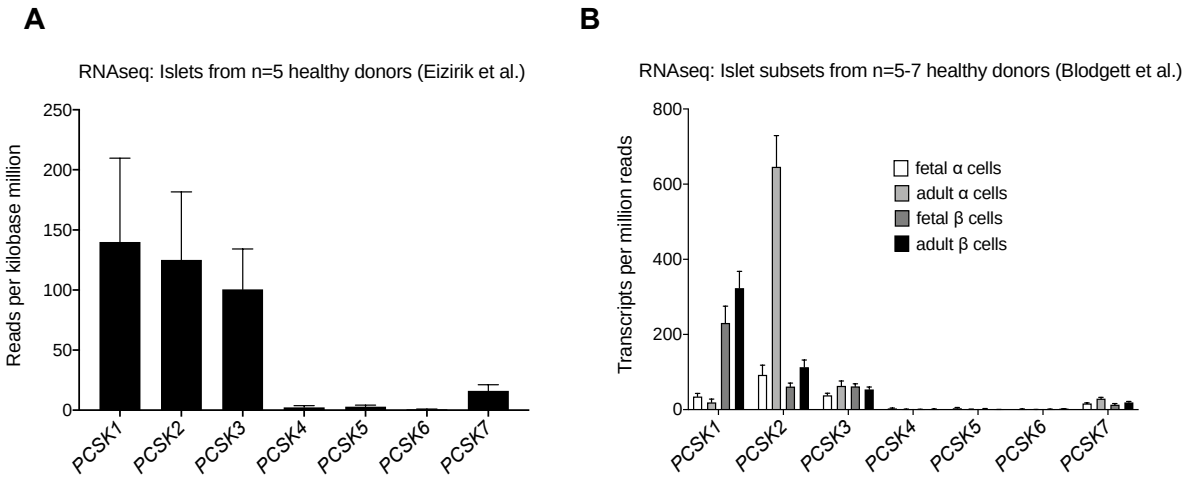

Figure S1 (A) RNA sequencing data of islets from healthy human donors (n=5) (taken from (1)). Data are shown as reads per kilobase million. (B) RNA sequencing data from islet subsets (i.e. fetal or adult α or β cells) from 5-7 healthy donors (taken from (2)). **Related to Figure 1.**

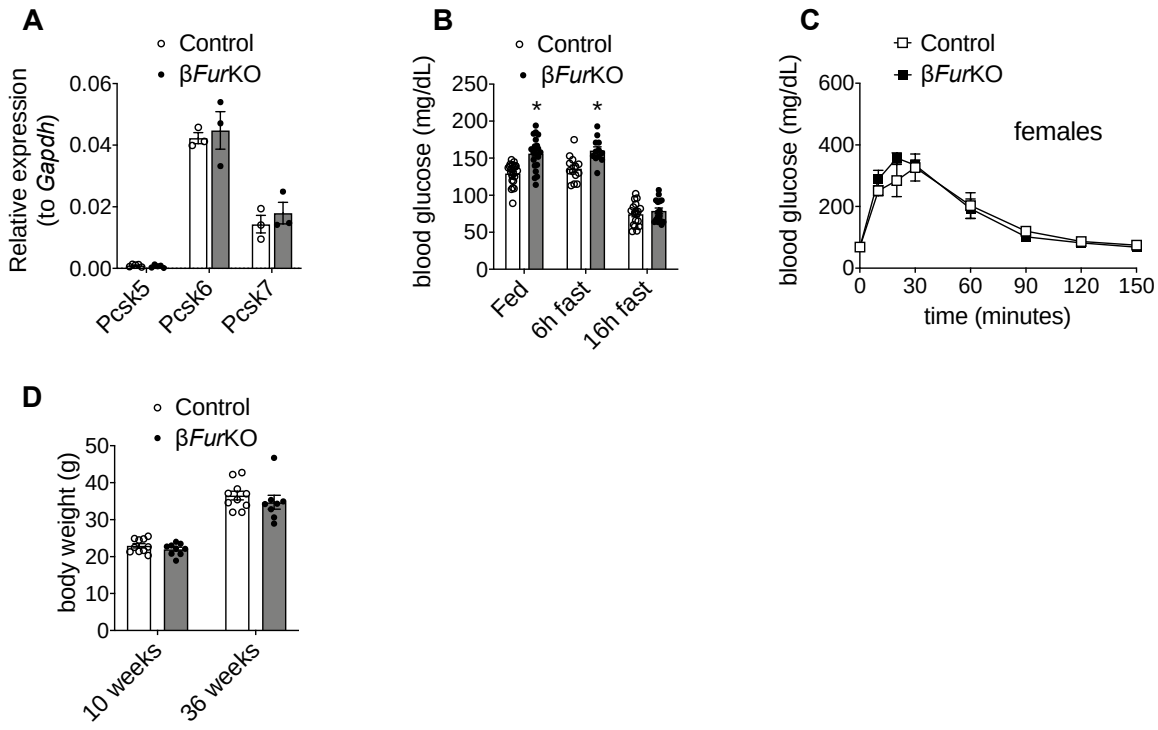

Figure S2 (A) qRT-PCR analysis of islet *Pcsk5*, *Pcsk6*, and *Pcsk7* mRNA from 12-week-old β*Fur*KO and control mice, n=3-5 per group, **P<0.01. (B) Blood glucose levels measured in 10-week-old

βFurKO and control mice, in a random fed state, and after a 6h or an overnight (ON, ~16h) fast, n=12-23 animals per group, *P<0.05. (C) IPGTT on 10-week-old female mice, (n=4)/genotype. (D) Body weight (g) of 10-week-old and 36-week-old male control and βFurKO mice (n=9-10/group). **Related to Figure 2.**

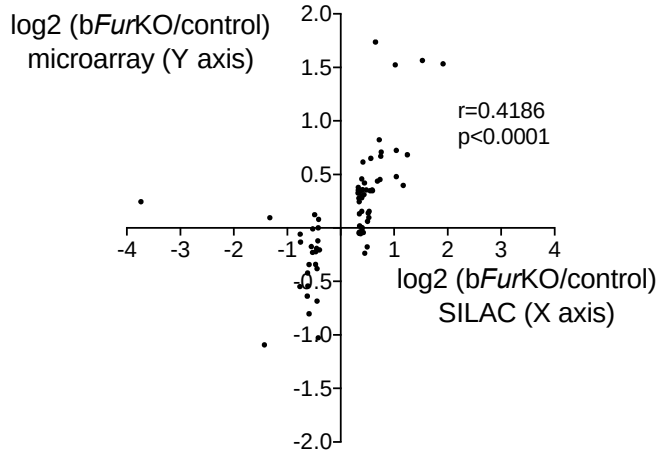

Figure S3 Correlation between SILAC and microarray data (r=0.4186, p<0.0001), for proteins that show at least a 25% increase or decrease in expression as quantified in the SILAC experiment. **Related to Figure 4.**

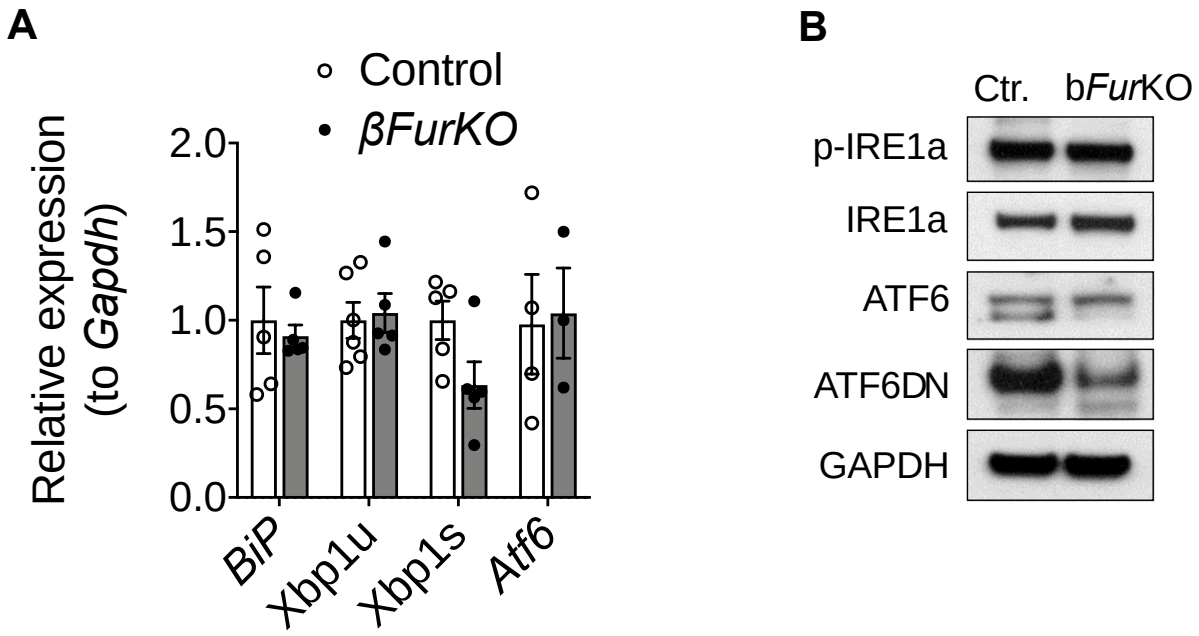

Figure S4 (A) qRT-PCR expression of genes involved in the unfolded protein response (UPR): binding immunoglobulin protein (*BiP*), unspliced and spliced X-box binding protein 1 (*Xbp1u*, *Xbp1s*) and Activating transcription factor 6 (*Atf6*), n=3-6 per group. (B) Protein analysis of total and phosphorylated Inositol-requiring protein 1 (IRE1α), and uncleaved and cleaved activating transcription factor 6 (ATF6, ATF6ΔN). GAPDH was used as a loading control. **Related to Figure 5.**

Figure S5 (A) Position of the CRISPR guide (gray) in mouse exon 2 of the *Pcsk3* gene, and the indels in the 3 different βTC3 clones. The first clone is compound heterozygous and contains a 2-bp deletion in the first allele (GC), and a 1-bp insertion in the second allele (T), leading to a frameshift on both alleles. In clones 2 and 3 we observed a 1-bp insertion (T) in all alleles tested, suggesting a homozygous mutation leading to a frameshift. (B) Protein immunoblotting of cell extracts from control and furin knockout βTC3 cells engineered using CRISPR Cas9 editing. GAPDH and Ponceau S staining were used as loading control. **Related to Figure 5.**

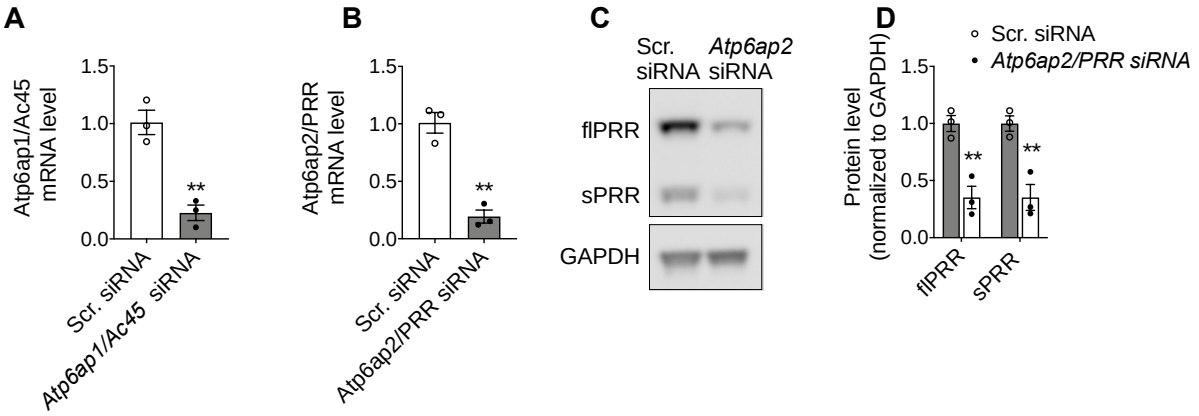

Figure S6 (A) Knockdown efficiency of *Atp6ap1/Ac45* in βTC3 cells measured by qRT-PCR, n=3 independent experiments. (B) Knockdown efficiency of *Atp6ap2/Prr* in βTC3 cells measured by qRT-PCR (B) and western blot (C), including western blot quantification (D), n=3 independent experiments. For western blot, GAPDH was used as a loading control. **P<0.01 determined by unpaired t-test. **Related to Figure 6.**

Table S1 Differentially expressed genes found by microarray, p<0.01, |fold change| ≥ 1.5.

Table S2 Ingenuity enrichment analysis for disease and functions, based on the microarray dataset.

Table S3 Ingenuity upstream regulator analysis, based on the microarray dataset.

Table S4 Proteins identified by SILAC. The highlighted proteins (gray) meet the p<0.0072 criteria.

Table S5 Ingenuity enrichment analysis for disease and functions, based on the SILAC dataset.

Table S6 Ingenuity canonical pathway analysis, based on the SILAC dataset.

Table S7 Ingenuity upstream regulator analysis, based on the SILAC dataset.

Table S8 General information on human islet donors used for qRT-PCR in Figure 1.

Table S9 Human and mouse primers used for qRT-PCR (listed in 5' - 3' direction), genotyping and CRISPR-Cas9 gene editing.

Supplemental Material

Intraperitoneal glucose (IPGTT), insulin (IPITT) tolerance tests and glucose stimulated insulin secretion (GSIS)

Male WT, Flox, Cre, Het and β*Fur*KO mice fed a normal chow diet were fasted overnight and injected with 2 mg/g body weight (BW) D-glucose (IPGTT) or 0.75 IU/g BW human insulin (ITT) in PBS, respectively. Blood glucose levels were monitored at indicated time-points using a Contour Glucometer (Roche). Mice were analyzed at ages 10 weeks, 20 weeks and 36 weeks as indicated. In a separate experiment, to establish secreted insulin levels, 24-week-old mice were fasted overnight and injected with a single intraperitoneal bolus of 3 mg/g D-glucose dissolved in PBS. Subsequently, plasma samples

were collected at indicated time points and analyzed for insulin using the Ultrasensitive Mouse Insulin ELISA (Mercodia).

Immunofluorescence of Dissociated Human Islets

Human pancreatic islets were obtained from the NIDDK-funded Integrated Islets Distribution Program (IIDP) at City of Hope. Dispersed human islet cells were stained as previously described (3). Briefly, human islets from 2 donors (1 healthy and 1 T2D) were dispersed to single-cell by a 6-8 min digestion with Accutase (ThermoFisher Scientific) at 37 °C with dispersal by a p1000 micropipettor every 45 seconds. Cells were seeded on chamber-slide wells (Sigma) coated with Geltrex (ThermoFisher Scientific) and cultured overnight in PIM(T)-medium (provided by IIDP) at 37°C in 5% CO2 incubator. Samples were fixed in 4% pre-warmed paraformaldehyde (PFA) for 30 min and acetone-methanol (1:3) for 3 min and then permeabilized 30 min in PBS with 0.2% Tween before being blocked with 1% Bovine Serum Albumin (BSA, Sigma) in PBS. Slides with sorted islets were immunostained with 1/500 furin antibody (ThermoFisher Scientific, PA1-062) and 1/50 insulin antibody (Dako, A0564) overnight at 4C. Secondary antibodies Alexa 488 anti-guinea pig and Cy3 anti rabbit from Jackson Immunoresearch (1/100) were added the next day and slides were mounted with ProLong Gold Antifade Mounting containing DAPI (ThermoFisher Scientific). Stained islet cells were examined with a Leica DM6 microscope and LAS X software.

Immunofluorescence (IF) on mouse pancreata

Pancreata were isolated and fixed in 4% paraformaldehyde (PFA) in PBS at 4°C overnight. Subsequently, they were dehydrated by a graded series of ethanol and butanol and embedded in paraffin. Sections were cut at 5µm thickness. Sections were deparaffinized, rehydrated and boiled for 20 minutes in Target Retrieval solution (Dako) to recover epitopes. Slides were blocked with 20% fetal calf serum (FCS) in PBS and incubated overnight with 1/10 anti-insulin (Dako) and 1/500 anti-Ki67 (Abcam) in Antibody Diluent (Dako) and subsequently incubated with Alexa antibodies (1/500) for 1h at room temperature. For Ki67 staining, six pancreatic sections per mouse, sampled every 200 µm, were stained

for insulin and Ki67, and all islets found on these sections were photographed using a Nikon C2 Eclipse confocal microscope, 20x magnification. The number of Ki67 $^{+}$ cells and the total number of β cells were counted using Image J software, and proliferation was calculated as (number of Ki67+ β cells/total number of β cells) x 100%.

β cell mass quantification

β cell mass of 24-week-old male mice was determined using a previously described protocol (4). Six pancreatic sections per mouse, sampled every 200 µm, were stained for insulin using standard IHC protocols. Briefly, sections were heated for 20 min in Target Retrieval Solution (Dako), blocked in 20% FCS and incubated with anti-insulin (1/10, Dako) in Antibody Diluent for 2h, followed by peroxidase-coupled secondary antibodies for 1h. 3'-3-Diamino-benzidine (DAB+, Dako) was used as substrate chromogen, after which sections were counterstained with hematoxylin and mounted. Six insulin-stained sections per mouse were photographed using a Zeiss Axioimager (Zeiss). Axiovision software (Micro Imaging, Heidelberg, Germany) was used to determine the relative insulin-positive area for every section. Subsequently, the β cell mass (mg) was calculated by multiplying relative insulin-positive area by the weight of the pancreas in mg.

Pancreatic insulin content

Total pancreatic insulin content was determined using the acid/ethanol extraction method. Briefly, pancreata were homogenized in acid/ethanol (0.12 M HCl in 75% ethanol) and after overnight incubation at -20°C, samples were centrifuged at 3,500 rpm for 15 min at 4°C to remove cell debris. Insulin content was determined using the Rat High Range Insulin ELISA (Mercodia).

Islet isolation

Islets were isolated by locally injecting the pancreas with 1 Wünsch unit/ml Liberase (Roche) in HANKS buffer as previously described (5). Injected pancreata were incubated for 18 min at 37°C in a shaker (220 rpm) and subsequently the islet fraction was separated from exocrine tissue using a Dextran

T70 gradient. Finally, islets were handpicked twice in HANKS buffer under a stereomicroscope to reach a pure islet fraction for further processing.

Islet Insulin Content and Release

For insulin secretion measurements, size-matched islets (n = 5 islets per tube, in triplicate for each condition per mouse) were placed in glass tubes containing HEPES Krebs solution 0.5% BSA supplemented with glucose: 5 mM (G5), 20 mM (G20), or G20 with 250 μM IBMX (Sigma). Supernatant was collected after 1 h incubation at 37ºC. The islets were sonicated for 3 min after adding acid ethanol (final concentration: 75% EtOH, 0.1 N HCl, 1% Triton). Samples were stored at 20ºC until quantification. The ELISA kit used for insulin determination was from Crystal Chem. Proinsulin levels were measured in the conditioned medium from islets incubated at 20mM glucose (samples from Figure 3L). Secreted proinsulin/insulin was normalised to total insulin content, similar to Figure 3L. The ELISA kit to quantify proinsulin was from Mercodia.

Cell culture and transfection

The mouse insulinoma cell line βTC3 was cultured in DMEM:F12 (1:1) supplemented with 10% heat inactivated fetal bovine serum, 100 U/ml penicillin, and 100 μg/ml streptomycin. For overexpression experiments, βTC3 cells were transfected with plasmids encoding mouse FURIN and/or mouse *ATP6AP1*/*AC45*-Flag using Lipofectamine 2000 (Life Technologies) according to the manufacturer's protocol. For knockdown experiments, βTC3 cells were transfected with SMARTpool siGENOME mouse *Atp6ap1/Ac45* (Dharmacon cat. #M-060210-01-0005) or *Atp6ap2/Prr* (Dharmacon cat. #M-063641-02-0005) using Lipofectamine RNAiMax (Life Technologies) according to the manufacturer's protocol. The Stealth siRNA Negative Control, siGENOME non-targeting siRNA control pools (Dharmacon cat. #D-001206-13-05) was transfected as a control. Cells were harvested 48 hours after transfection. PRR knockdown was verified by western blot, Atp6ap1/*Ac45* knockdown was confirmed by qRT-PCR.

Apoptosis assay

Apoptosis was measured in freshly isolated islets using the Cell Death ELISA kit (Roche) according to the manufacturer's protocol.

Cell proliferation assay

βTC3 cells were plated in triplicates at a density of 1×10^4 cells/well in 96-well plates and then incubated for 72 h. Cell proliferation was measured using the mitochondrial dye WST-1. Briefly, cells were incubated with 10 µl of WST-1 reagent per well for 2h, and the absorbance was measured at 450 and 600 nm. Three independent experiments were performed with the results presented ± SEM for the mean of all assays.

Electron Microscopy (EM)

Staining for the analysis of ultrastructures

Freshly isolated islets of 24-week-old male mice were washed with PBS, fixed with 4% formaldehyde/0.1% glutaraldehyde in 0.1 M sodium cacodylate buffer (pH 7.4), and pelleted at 13,000 rpm for 5 min. The fixative was aspirated, and the cell pellet was resuspended in warm 10% gelatin (sodium cacodylate buffer). The islets were then collected by centrifugation (4000 rpm for 1 min) and the gelatin-enrobed pellet was set on ice for 30 min, post-fixed in 1% osmium tetroxide for 2 h, dehydrated in increasing concentrations of ethanol, stained with 2% uranyl acetate and embedded in agar low viscosity resin.

After trimming the resin block containing the cell pellets, 70 nm sections were cut using a Reichardt Ultracut E ultramicrotome. The sections were collected as ribbons of 3-4 sections on a 75-mesh grid (Ted Pella). Every grid was then post stained with 3% uranyl acetate in water for 10 minutes and Reynold's lead citrate for 2 minutes. EM images were taken at 2500x magnification by a JEOL TEM1400 transmission electron microscope equipped with an Olympus SIS Quemesa 11 Mpxl camera at 80 kV. Random single sections from β cells (15 cells total) that possessed multiple dense-core granules and well-fixed cellular constituents (mitochondria, nuclear material and plasma membrane) were selected for the analysis. The plasma membranes, granules and nuclear borders of each β cell were manually marked in transmission electron microscopy (TEM) micrographs. Scanned images were

elaborated with the open-source software Microscopy Image Browser (MIB) to generate a mask of the cell compartments and the nucleus, and imported into ImageJ (National Institutes of Health, Bethesda, MD, USA) for image analysis. The immature secretory granules were distinguished from the mature secretory granules on the basis of the core electron-density and the presence of a distinctive, less pronounced halo as described by others (6).

Immunolabeling for the analysis of lysosomal pH (Tokuyasu method)

The islets of 20 week-old male mice were isolated as previously described and pre-incubated with 30 μM of DAMP (in HANKS buffer 0.02% BSA) at 37°C for 1h, and subsequently washed 2 times with the same buffer (100 rpm, 30 sec), then fixed for 1 hour at room temperature with 4% formaldehyde/0.2% glutaraldehyde in 0.1 M sodium cacodylate buffer (pH 7.4), and centrifuged at 5500 rpm for 1 min. The fixative was aspirated, and after 2 washes with HANKS buffer 0.02% BSA, the islet pellet was resuspended in warm 12% gelatin (0,1M phosphate buffer). The islets were then collected by centrifugation (13000 rpm for 5 min) and the gelatin-enrobed pellet was put on ice for 30 min. The solidified gelatin pellet with the islets aligned on the bottom was cautiously detached from the base of the Eppendorf tube, cut into smaller blocks, and submerged in a 2,3 M sucrose solution for 24h. The blocks were then mounted on cryo pins (#BS4620, Sapphire products limited, UK) and snap-frozen in liquid nitrogen. Ultrathin cryosections (70 nm thick) were cut using a Leica Ultracut UC7 with Leica FC7 cryo chamber and collected on glow discharged carbon-coated copper 200-mesh grids (#1800-F, Van Loenen Instruments, The Netherlands) with a 1:1 mixture of 2,3 M sucrose and 2% methylcellulose solution.

After floating on 2% gelatin in PBS at 37°C, grids were blocked with 0,15% glycine in PBS (3 times, 2 min each), 2% BSA in PBS for 15 minutes, and 0,1% BSA-c (#900099, Aurion, The Netherlands) in PBS (3 times, 2 min each). After blocking, grids were incubated with rabbit anti-DNP (1:400) in PBS 0,1% BSA-c for 1 hour. The primary antibody was detected with 15 nm gold protein A (#815.111,

Aurion, The Netherlands). The labeling was stabilized with 2% glutaraldehyde in PBS and washed with Milli Q water 6 times, 2 minutes each.

The lysosomes were imaged in a JEOL JEM1400-LaB6 Transmission Electron Microscope (TEM) equipped with an Olympus Quemesa 11Mpx camera at 25000x magnification. The gold particles of each lysosome were manually counted, and the lysosomal area was measured by using the Imagej software. The identification of the lysosomal ultrastructures was determined by the help of a few sections separately stained with the lysosomal marker LAMP-1 1/50 DSHB (Developmental Studies Hybridoma Bank), 1D4B, and then detected with a goat anti rabbit 6nm 1/30 (data not shown).

Western blot

Freshly isolated islets from 12 to 20 week-old male mice were lysed in Lysis buffer (Cell Signaling Technology) supplemented with protease and phosphatase inhibitor cocktails (Roche) by sonication on ice. βTC3 or HEK293T cells were lysed in 1x Ripa and extraction lysis buffer (Thermo Scientific) respectively, supplemented as above. Protein concentration was determined by BCA analysis (Thermo Scientific). Samples were boiled for 10 minutes in 4% β-Mercaptoethanol and 1x sample buffer (Thermo Scientific) and loaded on a 10% Bis/Tris gel with MES or MOPS running buffer for SDS-PAGE analysis. Proteins were transferred to a nitrocellulose blot, blocked with a blocking buffer (5% non-fat milk, 0.2% Triton X-100 in PBS) and subsequently incubated with primary antibody in the blocking buffer at 4°C overnight. Blots were washed in PBS 0.2% Triton X-100, incubated with peroxidase-conjugated secondary antibody (Dako) for 1h, and proteins were detected using the Western Lightning ECL System (PerkinElmer). For deglycosylation experiments, 100 µg of proteins from the cell lysate were boiled at 95°C for 5 min in sodium phosphate buffer 0.1% SDS and β-Mercaptoethanol (diluted 1:6 in water) in a total volume of 35ul. Once cooled down, the samples were supplemented with 0.8% NP-40 and 1 Unit of N-glycosidase F (Roche) in a final volume 50ul and incubated at 37°C overnight. The samples were then processed for western blot as above. The primary antibodies used were rabbit anti-ATF4, anti-phospho-p70S6K (Thr389), anti-p70S6K, anti-phospho-PERK (Thr980), anti-PERK, anti-phospho-4E-BP1 (Thr37/46), anti-4E-BP1, and anti-GAPDH from Cell Signaling

Technology; mouse anti-CHOP and, anti-eIF2α from Cell Signaling Technology; rabbit anti-phospho-IRE1α (Ser724) and anti-Renin R from Novus Biologicals; mouse anti-flag M2 from Sigma; mouse anti-ATF6 from Novus Biologicals; mouse anti-FURIN and rabbit anti-phospho-eIF2α (Ser51) were generated as previously described (7). To determine secreted sPRR levels in the culture medium, proteins were precipitated with methanol. Briefly, βTC3 cells were plated in 6-well plates at 80% confluence, washed and incubated overnight at 37°C and 5% CO2 in 1ml of serum-free DMEM:F12 medium. Sixteen hours after incubation the conditioned medium was transferred to ice cold tubes and centrifuged at 1000 rpm at 4°C for 10 minutes. Then, an equal amount (12.5μg) of bovine serum albumin (BSA) was added to each supernatant. After the addition of BSA, the samples were vigorously vortexed and 4 ml of cold methanol was added and mixed by hand shaking. After a 2 hour at -20°C, the samples were centrifuged at 1500 rpm at 4°C for 15 minutes. Then, the supernatants were removed and the pellets were dried for 2 hours. Dried pellets were dissolved in 25 μl of 2x sample buffer and analyzed by Western blot as described above.

Stable Isotope Labeling by Amino acids in Cell culture (SILAC)

Spike-in SILAC analysis was performed to compare relative abundance of proteins in islets from 12 week-old male Flox and β*Fur*KO mice, following established protocols. To prepare the spike-in standard, MIN6 cells were cultured for 12 passages in SILAC medium Dulbecco's modified Eagle medium (DMEM), high glucose (4,5 g/l) without L-glutamine, arginine and lysine (Silantes Basic Products cat. #280001300), supplemented with 15% fetal bovine serum (FBS), glutamine PS, heavy arginine (+10) and heavy lysine (+8) (Silantes Basic Products cat. no. 282986444). Light and heavy labeled cells were lysed in freshly prepared SDT-lysis buffer (2% SDS, 50 mM DTT, 100 mM Tris HCl pH 7.6), DNA was sheared by sonication, the sample was incubated at 95°C for 5' and centrifuged. To verify the labeling efficiency, 25 μg of the cell lysate of the light labeled cells (L) were mixed with equal amounts of heavy labeled cells (H) and digested with trypsin using the FASP protocol. After analysis of the sample on a Q Exactive mass spectrometer (Thermo Scientific), more than 99% of the peptides had a $\log_2$ normalized ratio between -1 and 1, demonstrating efficient labeling.

Sample preparation for proteomic analysis

Islets from 4 WT and 4 β*Fur*KO mice were isolated as previously described and were pooled per 2 mice of the same genotype to obtain at least 20 µg of proteins upon lysis in SDT-lysis buffer. Twenty µg of islet lysate was pooled with equal amounts of the spike-in standard and digested with trypsin using the FASP protocol (8). Using strong anion exchange (SAX) on a StageTip format (Thermo Scientific), 3 fractions (pH 3, 6 and 11) were obtained and analyzed on a Q Exactive mass spectrometer. The quality of the spike-in standard was analyzed as described before. For more than 90% of the proteins the difference in experimental sample and the standard was <5, suggesting that the spike-in standard was of good quality.

Mass spectrometry

Samples were analyzed by LC-MS/MS, coupling an Easy nLC1000 nanoflow HPLC system to the Q Exactive benchtop mass spectrometer (both Thermo Fisher Scientific) as described previously (9). Data were analyzed using the MaxQuant software environment (version 1.3.0.5) as previously described. Statistical evaluation of the data was performed with the freely available Perseus statistics software (version 1.2.0.7) and Microsoft Excel. Common contaminants and reverse decoy matches were removed from the protein identification list. At least 2 unique peptides per protein were required for protein identification. H/L ratios were reversed to L/H ratios. Only proteins that were identified and quantifiable in at least three biological replicates in each group were used for relative quantification. The average group means of the ratios were calculated and data are expressed as the log2 value of the ratio of ratios (β*Fur*KO/control). For statistical evaluation, a two-sided t-test was used. The p-value was corrected using false discovery rate (FDR) based multiple hypothesis testing. Both t-test and FDR based multiple hypothesis testing were carried out with the default settings of the Perseus statistics software.

Quantitative RT-PCR (qRT-PCR)

RNA from mouse islets or βTC3 cells was isolated using the Nucleospin RNA II (Macherey Nagel) kit according to the manufacturer's protocol. cDNA was synthetized using the iScript cDNA synthesis kit (Bio-Rad). Primers were designed using the ProbeFinder software (Roche Applied Sciences). qRT-PCR

was performed in triplicate with a MyiQ single-color real-time PCR detection system (Bio-Rad) using SYBR Green. RNA from human islets (~150 for each donor) was extracted with RNeasy Mini Kit (Qiagen) and was reverse transcribed using the High Capacity Reverse Transcription Kit (Applied Biosystems). qRT-PCR was performed with a ViiA 7 detection system from Life Technologies (Thermo Fisher Scientific) using SYBR Green PCR master mix (Bioline). Data is represented as 2ˆ-ΔCt compared to average of PC1/3 in healthy group. Primers for human and mouse genes are listed in **Table S9**.

Statistical analysis

Results are expressed as means ± SEM. Statistical analysis was performed by unpaired Student's t test or two-way ANOVA with post-hoc Bonferroni correction for pairwise time-specific differences between genotypes. GraphPad Prism 8 software was used for all analyses. A value of $p<0.05$ was considered significant.

Supplemental references

1. Eizirik DL, Sammeth M, Bouckenooghe T, Bottu G, Sisino G, Igoillo-Esteve M, et al. The human pancreatic islet transcriptome: expression of candidate genes for type 1 diabetes and the impact of pro-inflammatory cytokines. PLoS Genet. 2012 Mar 8;8(3):e1002552.
2. Blodgett DM, Nowosielska A, Afik S, Pechhold S, Cura AJ, Kennedy NJ, et al. Novel Observations From Next-Generation RNA Sequencing of Highly Purified Human Adult and Fetal Islet Cell Subsets. Diabetes. 2015 Sep;64(9):3172–81.
3. Furuyama K, Chera S, van Gurp L, Oropeza D, Ghila L, Damond N, et al. Diabetes relief in mice by glucose-sensing insulin-secreting human α-cells. Nature. 2019 Mar;567(7746):43–8.
4. Brouwers B, de Faudeur G, Osipovich AB, Goyvaerts L, Lemaire K, Boesmans L, et al. Impaired islet function in commonly used transgenic mouse lines due to human growth hormone minigene expression. Cell Metab. 2014 Dec 2;20(6):979–90.
5. Gysemans CA, Waer M, Valckx D, Laureys JM, Mihkalsky D, Bouillon R, et al. Early graft failure of xenogeneic islets in NOD mice is accompanied by high levels of interleukin-1 and low levels of transforming growth factor-beta mRNA in the grafts. Diabetes. 2000 Dec;49(12):1992–7.
6. Masini M, Marselli L, Bugliani M, Martino L, Masiello P, Marchetti P, et al. Ultrastructural morphometric analysis of insulin secretory granules in human type 2 diabetes. Acta Diabetol. 2012 Dec;49 Suppl 1:S247–52.

7. Scheuner D, Vander Mierde D, Song B, Flamez D, Creemers JWM, Tsukamoto K, et al. Control of mRNA translation preserves endoplasmic reticulum function in beta cells and maintains glucose homeostasis. Nat Med. 2005 Jul;11(7):757–64.

8. Geiger T, Wisniewski JR, Cox J, Zanivan S, Kruger M, Ishihama Y, et al. Use of stable isotope labeling by amino acids in cell culture as a spike-in standard in quantitative proteomics. Nat Protoc. 2011 Feb;6(2):147–57.

9. Dislich B, Wohlrab F, Bachhuber T, Müller SA, Kuhn P-H, Hogl S, et al. Label-free Quantitative Proteomics of Mouse Cerebrospinal Fluid Detects β-Site APP Cleaving Enzyme (BACE1) Protease Substrates In Vivo. Mol Cell Proteomics. 2015 Oct;14(10):2550–63.