

Cav β 3 regulates Ca²⁺-signalling and insulin expression in pancreatic β cells in a cell-autonomous manner

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Supplementary Figure Legend

Supplementary Figure 1. Depolarization-induced Ca^{2+} entry in wild-type and Cav β 3-KO β -cells. (A) Mean Fura-2 (F340/F380) ratiometric traces in the presence of 2 mM extracellular Ca^{2+} before and after addition of 25 mM potassium in wild-type (black) and Cav β 3-KO (red) cells. Application of 2 μM nimodipine reduced Ca^{2+} -entry. (B) Resting Ca^{2+} , peak amplitude and area under the curve of the potassium-induced Ca^{2+} -influx, shown as Tukey's box and whiskers with the boxes extend from the 25th to the 75th percentile and the line inside the box shows the median. The inter-quartile ranges (IQR) represent the difference between the 25th and 75th percentiles. Whiskers are extended to the most extreme data point that is no more than 1.5 \times IQR from the edge of the box and outliers beyond the whiskers are depicted as dots. The indicated P-values were calculated by Mann-Whitney test and the number of measured cells (x) per experiment (y) are indicated as (x/y) in panel B.

Supplementary Figure 2. Cav β 3 inhibits frequency of low (3 mM) glucose induced Ca^{2+} oscillations. (A) Representative Fura-2 (F340/380) ratiometric traces in the presence of extracellular Ca^{2+} from wild-type (black) and Cav β 3-KO (red) β -cells in the presence of 3 mM extracellular glucose. (B) Numbers of glucose-evoked Ca^{2+} oscillations per minute (left) and the mean peak amplitude per cell (right) from experiments in A. (C) Representative Fura-2 (F340/380) ratiometric traces in the presence of extracellular Ca^{2+} from wild-type β -cells pretreated with vehicle (+vehicle, black) or 10 μM xestospongin C (+Xest. C, gray) for 20 min and xestospongin C was maintained during the whole experiment. (D) Numbers of glucose-evoked Ca^{2+} oscillations per minute (left) and mean peak amplitude per cell (right) from experiments in C. Data in B and C are shown as Tukey's box and whiskers with the boxes extend from the 25th to the 75th percentile and the line inside the box shows the median. The inter-quartile ranges (IQR) represent the difference between the 25th and 75th percentiles. Whiskers are extended to the most extreme data point that is no more than 1.5 \times IQR from the

edge of the box and outliers beyond the whiskers are depicted as dots. The indicated P-values were calculated by Mann-Whitney test and the number of measured cells (x) per experiment (y) are indicated as (x/y). **(E)** Co-immunoprecipitation of the IP3R3. Immunoprecipitations were performed with antibodies against Cav β 3 and anti-IP3-receptor type 3 (IP3R3). Eluted protein complexes were subjected to Western blot using a second independent anti-IP3R3 antibody.

Supplementary Figure 3. Full scans for Western blots (uncropped images) which are shown in main Figures 1B, 1C, 4C, 5E,7K and S2E.