Supplementary Information for:

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Intravital imaging of islet Ca²⁺ dynamics reveals enhanced β cell connectivity after bariatric surgery in mice

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This PDF file includes:

Supplementary text Figures S1 to S5 Legends for Movies S1A to S1F



Percentage of pancreatic area occupied by and ratio of α to β cells. (a) Percentage of pancreatic area occupied by β cells measured by immunofluorescent staining of pancreatic islets using anti-insulin antibody against total pancreatic area. (b) Ratio of α to β cells in pancreatic islets of VSG (n=4) and Sham (n=5) mice measured by immunofluorescent staining of pancreatic islets using anti-glucagon (red) and anti-insulin (green) antibody. (c) Percentage of pancreatic area occupied by α cells measured by immunofluorescent staining of pancreatic islets using anti-glucagon antibody against total pancreatic area. All pancreatic biopsies were obtained 10 weeks following surgery. Each point represents an average of all islets (10-30/ slide) present in three permeabilised pancreatic slices, separated by 600µm. Total pancreatic area is measured in each slide and percentage is calculated accordingly.*P<0.05, by unpaired Student's t-test. Data are expressed as means ± SEM.

Supplementary Figure 2:



Immunofluorescence staining of pancreatic islets. Anti-glucagon (red) and anti-insulin (green) antibodies where used in VSG (n=4) and Sham (n=5) mice. All pancreatic biopsies were obtained 10 weeks following surgery (age: 32 weeks). For further details please see Results and Methods Sections.

VSG

2

Supplementary Figure 3:



Sham



VSG



In vivo measurement of mitochondrial membrane potential. Mean Intensity measurements in ACE islets of sham (n=7 islets, 1 mouse) and VSG (n=4 islets, 3 mice) 15 mins following IV injection of TMRM (0.5mg/kg). For further details please see Results and Methods Section. Source data are provided as a Source Data file.

Supplementary Figure 4:



Liraglutide-treated and weight matched group. (a) Body weight monitoring following Liraglutide (n=8 animals) or weight matched control (n=9 (b). Glucose tolerance test. Glucose was administered via intraperitoneal injection (1 g/kg) after mice were fasted overnight and blood glucose levels measured at 0, 15, 30, 60 and 90 min. post injection, after two weeks of treatment and (c) six weeks of treatment (d) Categorisation of Ins1Cre:GCaMPffl/fl islets in Liraglutide-treated (n=6 animals, n=7-10 islets) and weight matched (n=3 animals, n-4-5 islets) mice on weeks 0 and 6. Categories: 0. No activity, 1. Oscillations, 2. Partial Wave, 3. Wave, 4. Super Wave (e) Percentage of significantly connected cell pairs Liraglutide-treated (n=6 animals, n=7-10 islets) and weight matched (n=3 animals, n-4-5 islets) mice. Data are means \pm SEM and *p<0.05 by Student t-test. Data are expressed as means \pm SEM. Source data are provided as a Source Data file.

Supplementary Figure 5:



Metabolomics and Lipidomics analysis. (a) Metabolomics and (b) Lipidomics in plasma 12 weeks following VSG (n=3) or sham (n=3) procedures and post 3-day Exendin9 wash out period. Source data are provided as a Source Data file.