Supporting Information

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SI Materials and Methods

Animal Models. Lewis rats with induced diabetes were used as islet recipients. For the isogeneic transplant model, Lewis rats were also used as islet donors. In the allogeneic model, Sprague–Dawley rats were used as donors (Harlan). Diabetes was induced by a single i.v. infusion of Streptozotocin (STZ; Sigma) at 85 mg/kg body weight. Rats were considered diabetic if nonfasting blood glucose was > 25 mmol/L for at least three consecutive days. To metabolically bridge the animals between diabetes induction and device implantation, slow releasing insulin (Linplant, LinShin) was applied s.c. and discontinued at the time of graft implantation.

Islet Isolation and Culture. Pancreatic islets were isolated from 9 to 10 wk old male Lewis or Sprague–Dawley rats, respectively. Digestion solution (Collagenase NB8, Serva) was injected in situ through the pancreatic common bile duct. Islets were purified by discontinuous Histopaque gradient centrifugation. Purified islets were maintained in complete CR medium [CMRL: RPMI medium (1:1) supplemented with 10% FBS] for 5–8 d at 37 °C in a 5% CO_2 incubator before integration into the implantable devices.

For experiments with GHRH agonist, islets were divided into two treatment groups: (*i*) culture media with vehicle (DMSO) as a control group, and (*ii*) culture media containing GHRH agonist JI-36 (10^{-6} M). Islets were cultured for 48 h, and medium change was performed at 24 h.

Determination of Islet Equivalent. Triplicate samples of 50–100 islets were stained with dithizone (Sigma Aldrich) and sized using an eyepiece reticle and inverted microscope as described (1). Samples of 2,500 islet equivalents per animal were embedded in alginate and integrated into the device. For GHRH experiments, a minimal mass model of 1,000 islet equivalents per animal was used.

1. Ludwig B, et al. (2010) Agonist of growth hormone-releasing hormone as a potential effector for survival and proliferation of pancreatic islets. *Proc Natl Acad Sci USA* 107: 12623–12628.

Glucose Tolerance Test. Rats were fasted overnight before the test. A glucose solution was given i.v. at 0.5 g/kg body weight, and blood glucose was recorded before injection and at 10, 30, 60, 120, and 180 min following injection of glucose.

Histology and Immunohistochemistry. For histological assessments, explanted islet slabs were dehydrated, processed in paraffin and sectioned at 4 μm. For Toluidine-Blue staining, sections were incubated in 0.1% Toluidine-Blue (Sigma-Aldrich) in 1% NaCl for 1 min. For immunohistochemistry, primary antibodies were mouse anti-human insulin (Biogenex), rabbit anti-human glucagon (ab18461. Abcam), and monoclonal anti-a-smooth muscle actin (SMA; clone 1A4; Sigma product A2547). The signal was amplified using the VENTANA amplification kit and visualized using avidin-biotin labeling and 3,3′-diaminobenzidine. All slides were counterstained with hematoxylin. Staining with isotype control antibodies was performed to confirm specificity of staining.

Measurement of Insulin Secretion by Static Challenge with Glucose. For static insulin secretion in response to glucose challenge (GSIR), islet slabs were equilibrated in Krebs-Ringer bicarbonate buffer (KRB: 137 mM NaCl/4.7 mM KCl/1.2 mM KH_2PO_4/1.2 mM $MgSO_4$-$7H_2O$/2.5 mM $CaCl_2$-$2H_2O$/25 mM $NaHCO_3$/0.25% BSA) at 2.8 mM glucose at 37 °C (5% CO_2) and subsequently stimulated with 16.7 mM glucose for 1 h. Secreted insulin and insulin content was measured by ELISA (Millipore) and values normalized to islet number.

Statistical Analysis. In all experiments, statistical differences between experimental groups versus appropriate controls were determined using one-way analysis of variance (ANOVA). Data are presented as mean ± SEM. Statistical significance was tested by analysis of variance with Bonferroni's or Student *t* test. Differences were considered significant at values of $P < 0.05$.