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Interleukin (IL)-1α and IL-1β released from islet resident macrophages impair the success of islet transplantation

S. Wrublewsky[1], F. Pohlemann[1], L. Prates Roma[2], S. Rother[3], M. Menger[1], M. Laschke[1], E. Ampofo[1];
[1]Institute for Clinical and Experimental Surgery, Saarland University, Homburg, Germany, [2]Biophysics Department, Center for Human and Molecular Biology, Saarland University, Homburg, Germany, [3]Center for Molecular Signaling, Saarland University, Homburg, Germany.

Background and aims: Inhibition of interleukin (IL)-1 signaling has been shown to improve the outcome of clinical islet transplantation. However, the mode of action of IL-1α and IL-1β during this process is still elusive. Therefore, the aim of the present study was to investigate (i) the cellular source of IL-1α and IL-1β within the islets, (ii) the role of IL-1 signaling for the endocrine function of β-cells and (iii) the effect of IL-1α and IL-1β on islet transplantation.
Materials and methods: Islets were isolated from wild type (WT), IL-1α$^{-/-}$ and IL-1β$^{-/-}$ donor mice and exposed to IL-1α or IL-1β under hypoxic conditions for 24 h. The viability and cellular composition of these islets were determined by using flow cytometry, calcein / propidium iodide staining and immunohistochemistry. Secretion of insulin, IL-1α and IL-1β was analyzed by an enzyme-linked immunosorbent assay (ELISA). Expression of insulin, MafA and pancreatic duodenal homeobox protein (PDX)-1 was analyzed by quantitative real-time (qRT)-PCR and Western blot. Binding of IL-1α and IL-1β to heparan sulfate of islets was analyzed by surface-plasmon resonance (SPR) spectroscopy. *In vivo*, WT, IL-1α$^{-/-}$ and IL-1β$^{-/-}$ islets were transplanted into WT mouse dorsal skinfold chambers and their revascularization was assessed by intravital fluorescence microscopy. Moreover, we used the streptozotocin-induced diabetic mouse model to study the endocrine function of the grafts.
Results: We found that IL-1α and IL-1β are solely expressed by islet resident macrophages and both cytokines reduce insulin secretion. Additional analyses revealed that only IL-1α is capable of binding to heparan sulfate of islets, which, in turn, diminished the inhibitory effect of IL-1α on insulin secretion. The analysis of the underlying mechanism demonstrated that the loss of both cytokines increases insulin expression in a PDX-1- and MafA-dependent manner. By using the mouse dorsal skinfold chamber model, we found an improved insulin-driven revascularization of transplanted IL-1α$^{-/-}$ and IL-1β$^{-/-}$ islets when compared to controls. Accordingly, the transplantation of IL-1α$^{-/-}$ and IL-1β$^{-/-}$ islets under the kidney capsule of diabetic mice led to an accelerated restoration of normoglycemia.
Conclusion: The loss of IL-1α or IL-1β improves the revascularization, and thus, the engraftment of islets. Hence, the herein introduced anti-inflammatory approach represents a promising strategy to increase the success rates of clinical islet transplantation.
Disclosure: **S. Wrublewsky:** None.

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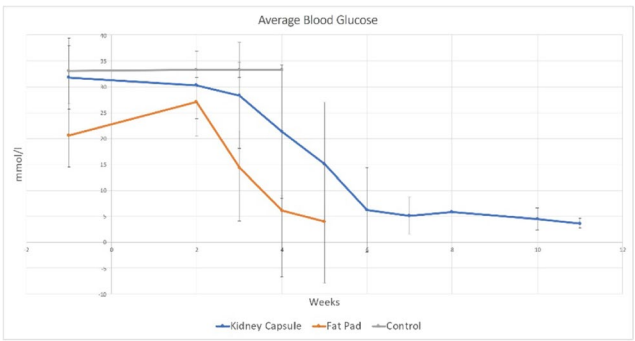
Stem-cell derived islets for treating diabetes

W. Rust, P. Dalal, A. Koval, J. Ratiu, S. Southard, P. Strumph, C. Welsch;
Seraxis Inc., Germantown, MD, USA.

Background and aims: We report the development of a novel human multipotent immortal stem cell line, SR1423 that efficiently differentiates to clusters of functional endocrine pancreatic cells. These Synthetic Replacement Endocrine (SRE) clusters contain cell types that recapitulate native islet function in vitro and potently control blood glucose in animal models of type 1 diabetes. Detailed characterization reveals significant distinctions between SRE, native islets and SC-beta cells. SRE clusters are manufactured following GMP requirements in closed vessels with large-scale formats and are being developed for near-term clinical studies.
Materials and methods: SRE was generated by transient expression of the Yamanaka factors in human islet cells harvested from a consented donor pancreas. Rather than screen for pluripotency, this cell line was selected for its ability to differentiate to the definitive endoderm in a first screen, and Pdx1+ pancreatic progenitors in a second screen. SRE does not meet criteria for pluripotency as it fails to express the master control gene for mesoderm specification, brachyury, in response to mesoderm-inducing agents. Comparison against a pluripotent stem cell database demonstrates poor overall differentiation capability for the mesoderm lineage. In contrast, SRE responds to basic differentiation protocols that drive endocrine pancreatic fate choice and generates highly pure populations of cells with endocrine cell characteristics. We therefore characterize SR1423 as a multipotent, and not pluripotent stem cell. In a prospectively designed islet replacement study male NSG mice with streptozotocin-induced diabetes were implanted with SRE clusters to the kidney capsule or gonadal fat pad (as a surrogate for the omentum) and disease progression was monitored.
Results: SRE resemble human islets in morphology and distribution of hormone expressing cells (data not shown). In this mouse model of STZ-induced type 1 diabetes (blood glucose > 11mmol/l), SRE implanted to the kidney capsule (n=17) or gonadal fat pad (n=10) resulted in euglycemia 3-4 weeks post-implant. Control mice (without SRE implant) in this model of type 1 diabetes (n=2) remained hyperglycemic.
Conclusion: These data are consistent with demonstration of proof of principle in this model with resolution of hyperglycemia after transplant of SRE. This is the first description of a novel cell type (non-native, non-SC-beta) with potential clinical therapeutic benefit. SRE clusters are currently in preparation for clinical studies.



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Glucose-dependent insulin production and insulin-independence in patients with type 1 diabetes infused with stem cell-derived, fully differentiated islet cells (VX-880)

T. Reichman[1], C. Ricordi[2], A. Naji[3], J.F. Markmann[4], B. Perkins[5], B.G. Bruinsma[6], G. Marigowda[6], D. Melton[6], F. Pagliuca[6], B. Sanna[6], L.S. Kean[7], C. Mathieu[8], A. Peters[9], P. Witkowski[10], M.R. Rickels[3];
[1]Toronto General Hosp., Toronto, ON, Canada, [2]Univ. of Miami, Miami, FL, USA, [3]Univ. of Pennsylvania Perlman School of Medicine, Philadelphia, PA, USA, [4]Massachusetts General Hosp., Boston, MA, USA, [5]Mount Sinai Hosp., Toronto, ON, Canada, [6]Vertex Pharmaceuticals Incorporated, Boston, MA, USA, [7]Boston Children's Hosp. and Dana-Farber Cancer Institute, Boston, MA, USA, [8]Katholieke Universiteit Leuven, Leuven, Belgium, [9]Univ. of Southern California, Los Angeles, CA, USA, [10]Univ. of Chicago, Chicago, IL, USA.

Background and aims: VX-880 is an allogeneic stem cell-derived, fully differentiated, pancreatic islet cell replacement therapy being evaluated in a Phase 1/2 clinical trial in patients with T1D and impaired awareness of hypoglycemia and severe hypoglycemia. We report results from the first 3 patients dosed with VX-880 with at least ~1 year of follow-up.
Materials and methods: The trial has 3 parts: Part A where 2 patients are enrolled sequentially and receive half the target dose, Part B where 5 patients are enrolled sequentially and receive the target (full) dose, and Part C where 10 patients are enrolled concurrently and receive the target dose. Following a single infusion of VX-880, patients are monitored for safety and tolerability (as assessed by adverse events [AEs] and clinical laboratory assessments), fasting and stimulated C-peptide, HbA1c, glycemic variability, interstitial glucose by continuous glucose monitoring, and exogenous insulin dose.
Results: Among the first 3 patients dosed with VX-880 and at least 1 year of follow up, 2 are insulin independent. Following VX-880 infusion at half target dose, Patient 1 in Part A, who was taking 34.0 units insulin/day and had HbA1c of 8.6% with 40.1% time-in-range (TIR) at baseline, became insulin independent at Day 270 and remains insulin independent at Month 18 (HbA1c 5.2%; 97.7%TIR). Patient 2 in Part A, who was taking 25.9 units insulin/day and had HbA1c of 7.5% with 35.9% TIR at baseline, had improved HbA1c (6.2%) and TIR (61.7.%) with a daily insulin dose of 27.8 units/day at Month 12. Patient 2 subsequently withdrew from the study for personal reasons. Patient 3, the first patient in Part B, received the full target dose of VX-880. This patient, who was taking 45.1 units insulin/day and had HbA1c of 7.6% with 53.8% TIR at baseline, became insulin independent at Day 180 and remains insulin independent at Month 12 (HbA1c 6.0%; 96.8 % TIR). VX-880 has been generally safe and well tolerated at both doses; the majority of adverse events (AEs) were mild or moderate in severity and there were no serious AEs considered related to VX-880. The safety profile was consistent with the immunosuppressive regimen and the perioperative period.
Conclusion: These results demonstrate that a single infusion of stem cell-derived islets (VX-880) can restore insulin production and glucose regulation, leading to insulin independence in patients with T1D. Based on this, VX-880 has the potential to be a functional cure for patients with T1D. Part B is fully enrolled and multiple additional patients have been dosed with the target dose. Longer-term and updated data from all patients dosed in Part A and Part B will be presented at the conference.
Clinical Trial Registration Number: NCT04786262
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Mesenchymal stem cell-laden composite islet porous microgel for diabetes treatment

J. Sun, L. Li;
Zhongda Hospital, School of Medicine, Southeast University, Nanjing, China.

Background and aims: As an alternative to continuous exogenous insulin administration of insulin, islet transplantation is shown to provide an alternative approach for controlling hyperglycemia in diabetes. However, islet transplantation faces significant challenges of low islet viability and immunologic insults. Thus, novel approaches capable of promoting the islet microenvironment for sustainability is highly desired. This study develops mesenchymal stem cell (MSC)-laden composite islet porous microgels (MGs) to address sustainability for the treatment of diabetes.
Materials and methods: The MGs are prepared via microfluidic droplet templates containing both MSCs and islets with a hybrid solution of poly (ethylene oxide) (PEO) and gelatin methacrylate (GelMA) for creating a porous structure. Islet viability and insulin secretion were measured in vitro. Changes of blood glucose, body weight and food intake were monitored in vivo in diabetic mice after omental transplantation. Histological analysis was performed after the removal of the implants.
Results: The 3D porous structure facilitated islet viability and function and allowed for insulin molecule penetration and rapid exchange of nutrients. The outstanding anti-apoptotic and immunomodulatory roles of co-encapsulated MSCs further provided a favorable environment for islet survival and function. In vivo experiments confirmed that MGs reversed hyperglycemia in diabetic mice, continuously regulated the dynamic balance of blood glucose levels within a normal range for 6 weeks, and rapidly secreted insulin in response to blood glucose stimulation. Histological analysis showed that the implants maintained hormone identities without significant inflammatory cell infiltration and was surrounded by neovascularization.
Conclusion: MSC-laden composite islet MGs demonstrated good hypoglycemic function, excellent biocompatibility and established a local immunomodulatory microenvironmenta, and diabetic mice treated by the MGs showed glycemic recovery, improvements in body weight and lower immune response. Overall, this strategy has promising applications in islet transplantation for diabetes treatment.
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Hydrogel encapsulation can protect functionality of transplanted islets in immunocompetent diabetic animals without immunosuppression

H. Stover;
Allarta Life Science Inc., Hamilton, ON, Canada.

Background and aims: Type 1 diabetes (T1D) is an autoimmune disorder characterized by the destruction of insulin-producing beta cells in the pancreas, leading to a chronic deficiency in insulin production. Despite the promise of islet cell transplantation to replace lost cells and cure T1D, the side effects of systemic immune suppression remain a major concern and long-term tight glycemic control remains elusive. Physical immune protection has the potential to enable allogeneic islet implantation and provide a functional cure. We have developed crosslinked, synthetic hydrogels that are robust, shape agnostic and non-immunogenic. More importantly, encapsulated rat and human donor islets remained viable and functional in vitro and in vivo, including in large animals.
Materials and methods: Micropipette aspiration, chemical stress tests (e.g., citrate), and in vivo persistence were used to investigate the hydrogels' mechanical robustness in different form factors, such as beads and strings. Protein release profiles and diffusion limits were evaluated using FITC-labeled dextran (10, 70, 250, and 500 kDa), IgG (150 kDa), and insulin (5.8 kDa). Different polymer modifications were tested for toxicity to human induced pluripotent stem cells (hiPSCs) and for fibrosis in healthy mice and pigs. Rat and human islet functionality after encapsulation were examined by glucose stimulated insulin secretion (GSIS). Marginal islet loads were tested for fibrosis and functionality (e.g., HbA1c, C-peptide, and blood glucose) following implantation into streptozotocin (STZ)-induced diabetic immunocompetent mice and healthy normal mixed breed pigs.
Results: The hydrogels are robust and easy to handle. They are effective at excluding IgGs and larger molecules, while allowing rapid insulin out-diffusion (~ 1 min $t_{½}$). Several novel immune-evasive modifications improved cell viability and non-immunogenicity. High viability (Fig. 1) and GSIS indices (avg. of 12x increase after stimulation) from rat and human islets demonstrate that the hydrogels maintained effective islet function in vitro. Capsules were retrieved intact after ~300 days in vivo. Encapsulated rat islets enabled rapid and persistent blood