Improving Islet Survival by Microencapsulation in Hydrogels Modified with Anti-Apoptotic Peptides

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Statement of Purpose: The aim of our research is to develop a biocompatible microencapsulation technique to facilitate pancreatic islet transplantation for the treatment of type I diabetes. Transplantation of pancreatic tissue has been demonstrated to be an efficacious method of restoring glycemic control in type I diabetic patients. In particular, islet transplantation has been very attractive because it does not require major surgery and islet tissue may be modulated prior to implantation to reduce the risk of rejection. Microencapsulation can protect the islet tissue through a mechanical barrier from the immunoreactive cells and antibodies and therefore enables islet transplantation in the absence of immunosuppression. Yet encapsulated islet graft survival is limited because of a lack of biocompatibility, inefficient immunoprotection from small inflammatory factors and hypoxia.[1] Our research utilizes enzyme-crosslinked hydrogels to encapsulate the islet tissue. Poly(ethylene glycol) molecules are modified with peptides that can be cross-linked at the presence of tissue transglutaminase (TGase) to form a biocompatible protective network around the islets. Presented on the hydrogels are also peptides inhibiting islet cell surface receptor for IL-1β, a cytokine that plays an important role in the inflammatory process causing the islet death after transplantation. Microencapsulated β-cells and islets in peptide-modified hydrogels are tested for viability and insulin secretion both *in vitro* and in *vivo*.

Methods: For this work, β-cell line INS-1 cells are used and pancreatic islets are isolated from 8-10 weeks old mice and used within 48 hrs after isolation. For hydrogel formation, 4-armed PEGs conjugated with peptide substrates for tissue transglutaminase (Ac-GQQQLG-PEG, Ac-FKG-PEG and Maleimide-FKG-PEG) are synthesized as previously reported.[2] IL-1 receptor (IL-1r) inhibitory peptide (Ac-CFEWTPGWYQPY-NH_2)[3] and a control peptide (Ac-CGGG-NH_2) are manually synthesized and conjugated to the hydrogels through the maleimide group (Mal) attached at the N-terminal of Mal–FKG-PEG. Density of IL-1r inhibitory peptide in the hydrogels can be adjusted by varying the ratio between Ac-FKG-PEG and Mal-FKG-PEG. These two peptides are also immobilized to maleimide-presenting cell culture plates for culturing cells and checking cell survival and functions. Encapsulation of β-cells and islets is carried out by adding cell suspension to a buffered solution containing the three different peptide-conjugated PEGs at defined ratios, followed by addition of a solution containing transglutaminase from guinea pig liver. *In vitro* cell viability assays are performed using the nuclei dyes Yo-Pro-1 and propidium to detect apoptosis and necrosis. For checking the function of encapsulated cells, glucose-sensitive insulin secretion analysis is carried out using the rat insulin ELISA kit from Mercodia AB. These islets are transplanted into diabetic mice to test the effects of encapsulation on long-term glycemic control.

Results/Discussion: To test if the presence of IL-1R inhibitory peptide affects the survival and function of islet cells, the 2D model system is used for the study prior to the use of 3D hydrogel network. INS-1 cells are cultured on surfaces presenting the IL-1R inhibitory peptide and a control peptide. Viability assay data show that the presence of IL-1r inhibitory peptide (IL-1r IP) significantly reduces the β-cell death induced by IL-1β combined with TNF-α and IFN-γ. (**Figure 1**) GSIS analysis shows that the insulin secretion function of β-cells is not affected by the IL-1R inhibitory peptide.

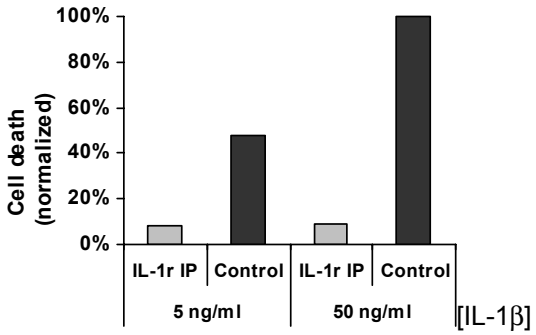

Figure 1. INS-1 cell apoptosis reduced by IL-1r inhibitory peptide (IL-1r IP)

Conclusions: Microencapsulation can provide a mechanical protective barrier for transplanted tissues from immunoreactive cells and antibodies. The microenvironment inside the capsule can also be modified to facilitate the trapped tissues to survive and function properly *in vivo* following transplantation. Our research will focus on inhibiting apoptosis of islet cells with an IL-1R inhibitory peptide presented on hydrogel capsules. Preliminarily results demonstrate this approach can improve the survival of β-cells without detrimental effects on insulin release function. Our work implies microcapsules presenting oligopeptide inhibitors of pro-inflammatory factors would improve islet engraftment in a controllable manner.

References:

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