CRYOPRESERVATION OF PIG PANCREATIC ISLET WITH TREHALOSE CRYOPROTECTANT

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Introduction:

Pig pancreatic islets are being considered as an alternative source for clinical islet transplantation, but the technique for pig islet cryopreservation is not well established. Trehalose is a sugar that can withstand prolonged periods of desiccation of plants and animals, which is also a non-permeating cryoprotective agent, has been documented as less toxic and highly efficient during cryopreservation of various cells or organisms. However, it has never been applied to the cryopreservation of the pig pancreatic islets.

Materials and Methods:

In this study, the selective osmotic shock method was used to isolate a number of pancreatic islets of Large White pigs, and the isolated islets were in-vitro cultured for four days. We used the dithizone (DTZ) staining, the hoechst/propidium iodide (PI) double staining and the glucose for stimulating insulin secretion to detect the viability and function of islets. After thawing, the recovery rate and insulin release of islets and mRNA level of INS, GCG, Bcl-2 and Bax genes in islets were analysed.

Results and Discussion:

The results of DTZ staining and hoechst/PI double staining showed that the isolated pig islets using the selective osmotic shock method had intact morphology and preferable viability. And then the insulin release level showed that the islet function was sustained and it's favorable cryopreservation time was 1 day after in-vitro culture (p<0.01). Furthermore, we conformed that the optimal concentration of trehalose in the islet cryopreservation was 0.1 mM/L, resulting from the recovery rate, insulin release, and mRNA level (P<0.05). Finally, it was reported that the effect of trehalose and glycerol dealing with cryopreservation of pig pancreatic islets were not different. As a result, trehalose was the one potential and less toxic cryoprotectant for pig pancreatic islet cryopreservation.