



HUMAN ISLET TRANSPLANT LABORATORY

Human Islet Isolation Procedure

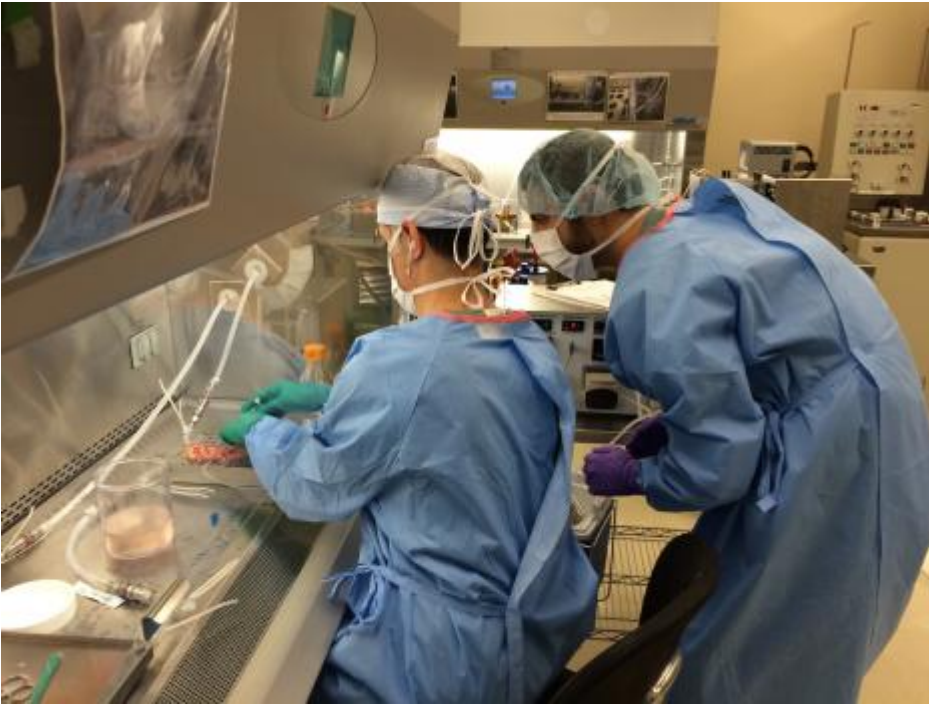
Photo Gallery
Craig Hasilo
February 7, 2014
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Pancreas retrieved from multi-organ donor is surface disinfected



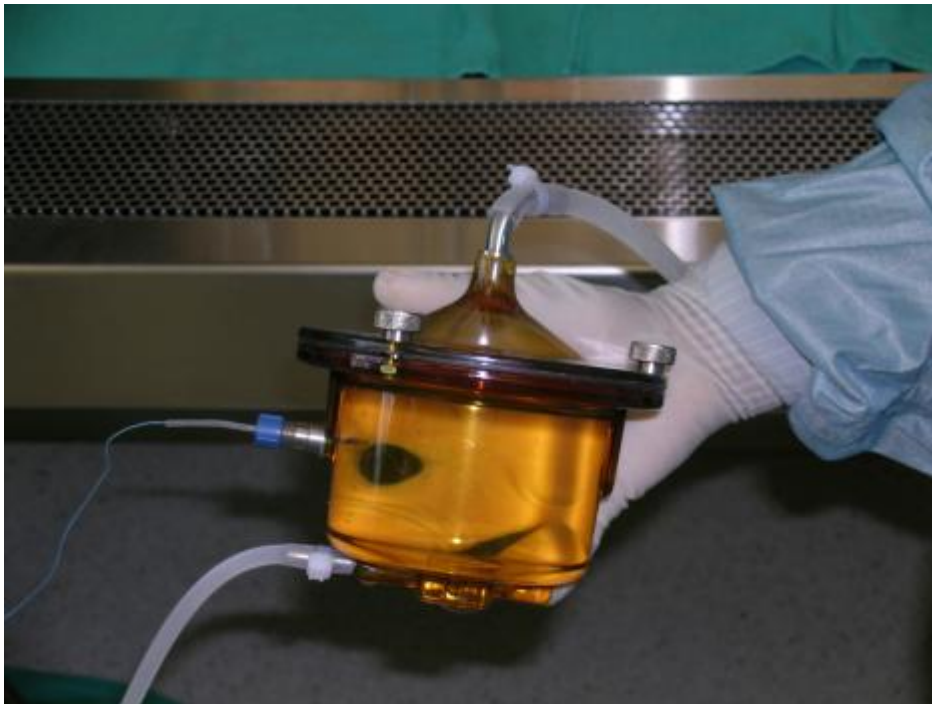
Excess fat and connective tissue is removed from the pancreas surface



The pancreas is carefully connected to the Perfusion pump



Digestive enzymes are pumped into the pancreas using the Perfusion pump



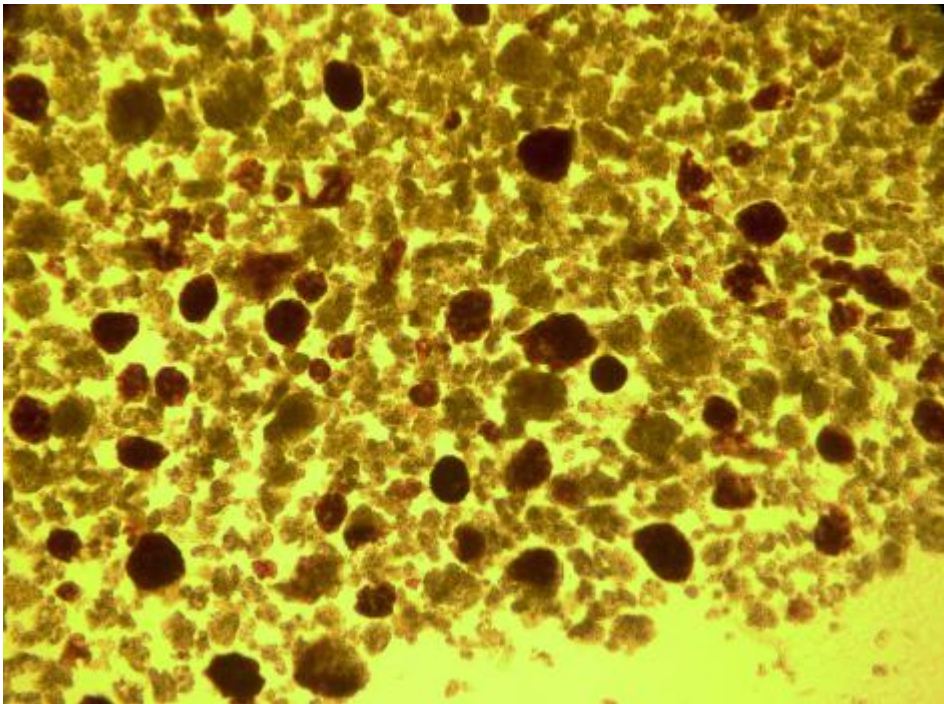
Pancreas is placed inside the Ricordi chamber



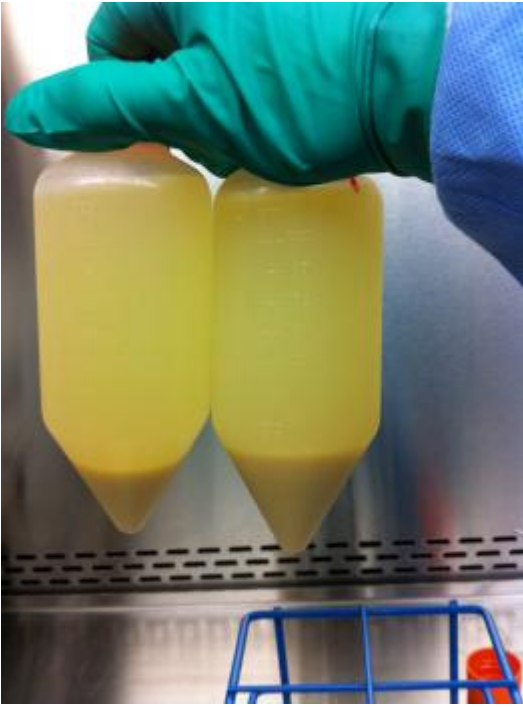
Media is heated up and circulated through the Ricordi chamber in this open loop system



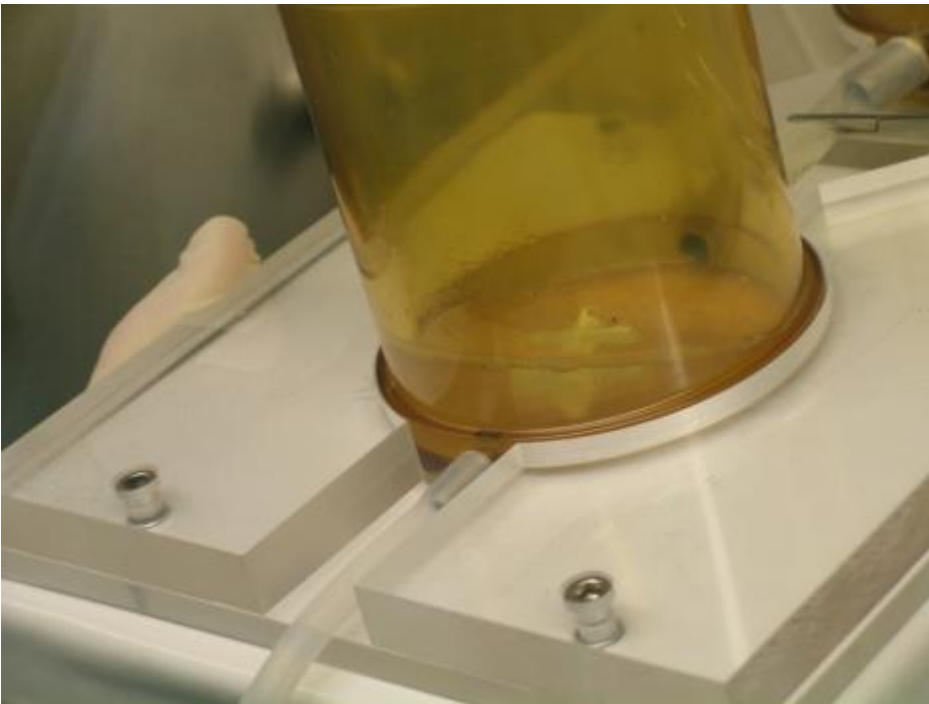
Ricordi chamber is shaken to facilitate mechanical and enzymatic digestion of the pancreas



Pancreas is digested into acinar and islets. Islets can be visualized (red) by staining with dithizone



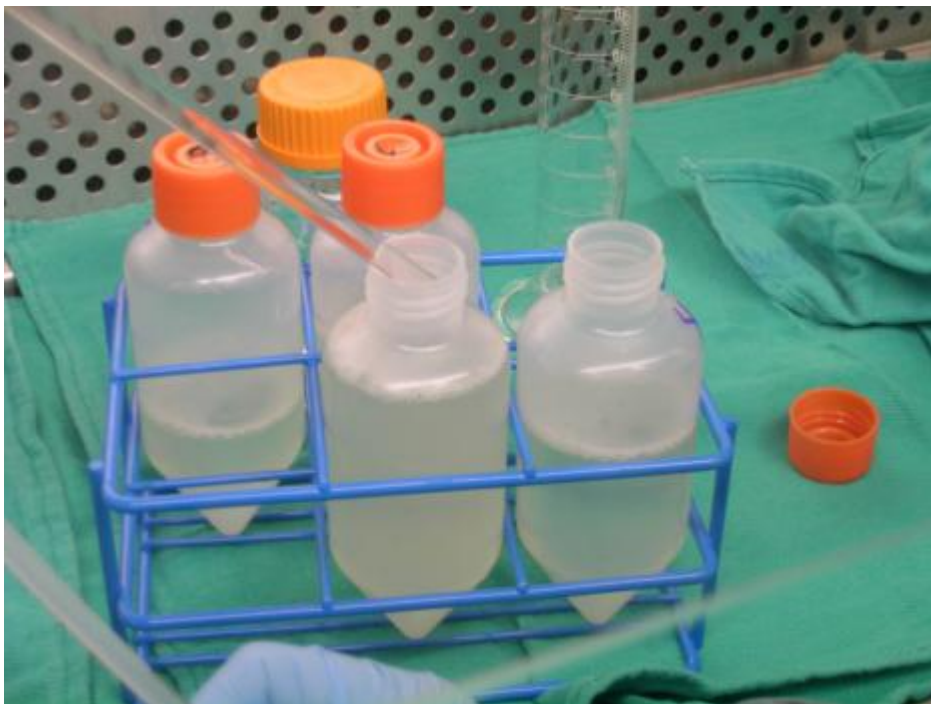
Once enzymatic digestion is completed, the pancreas is reduced to a volume of 50g comprising of acinar and islets



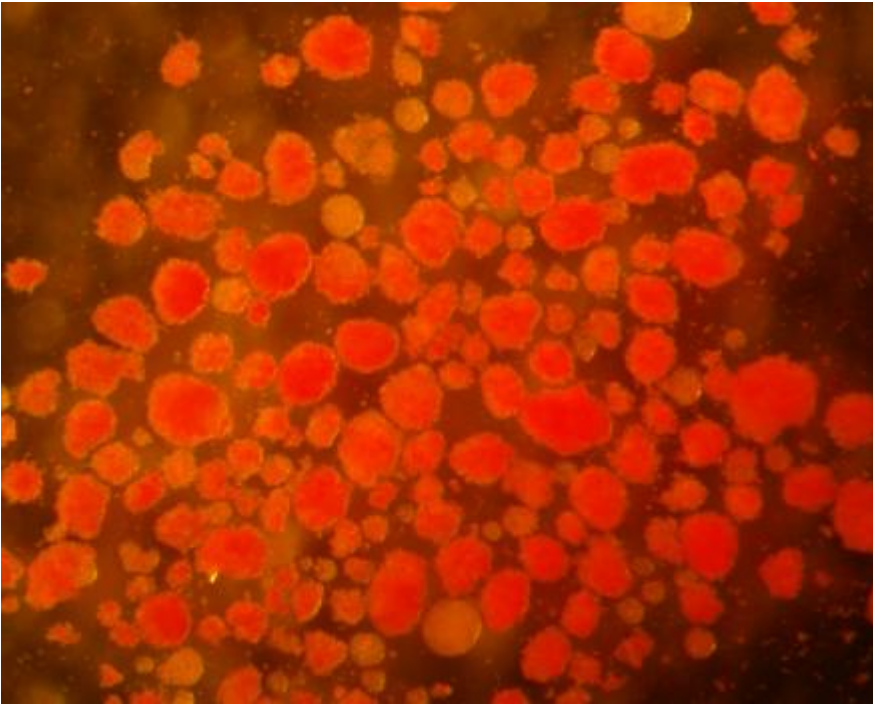
The digested pancreatic tissue is purified using gradients of various densities



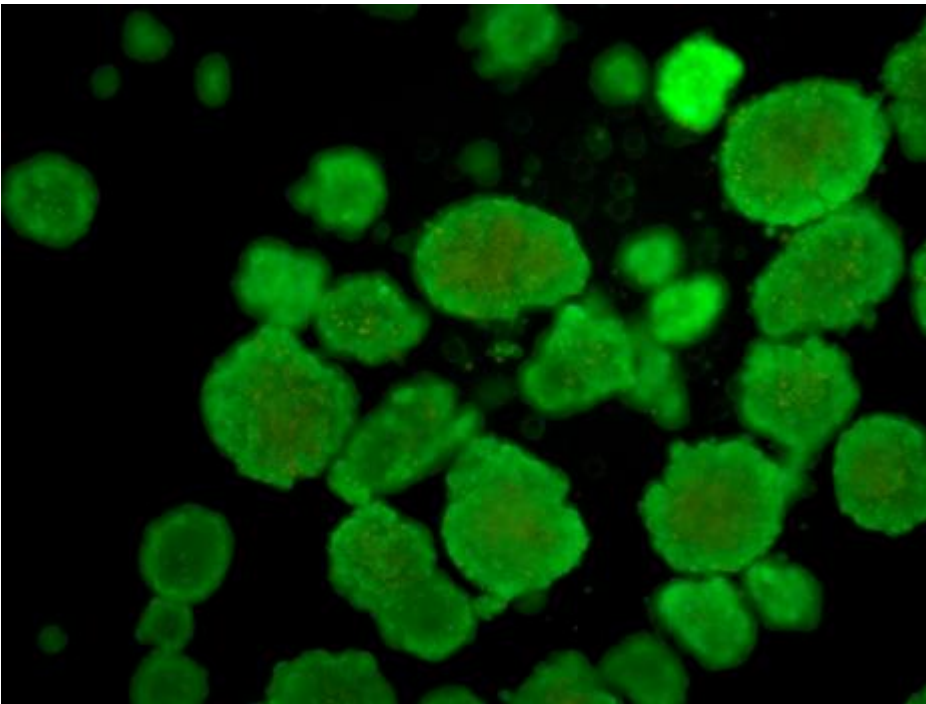
Gradient purification process using the COBE 2991 Cell Processor, separates the islets from the acinar cells



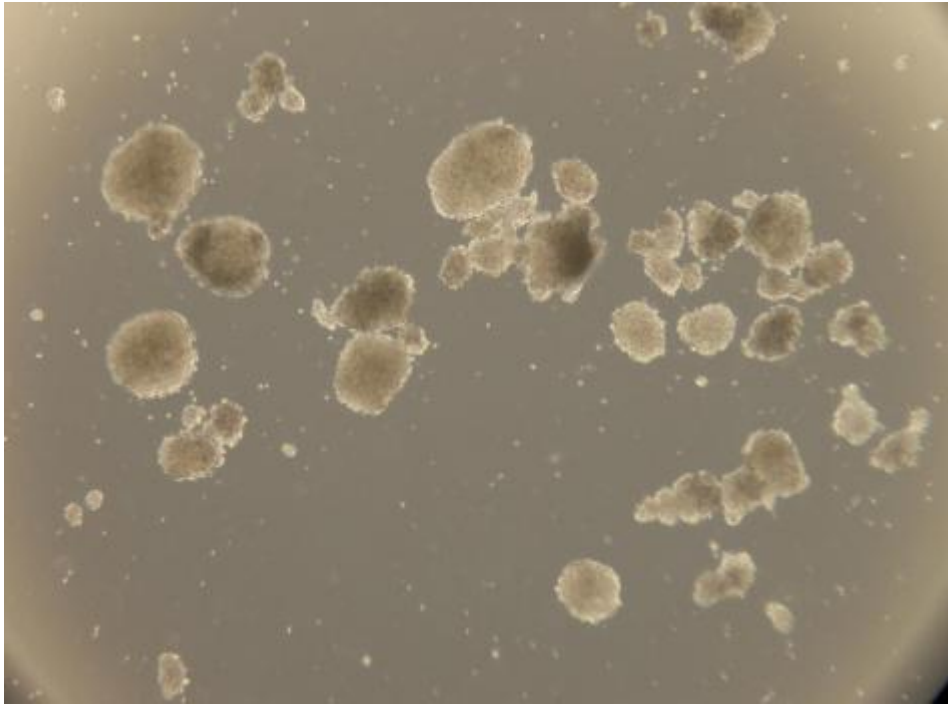
The pure islets are then collected and cultured



Dithizone staining helps visualize the islets by staining them red



Viability fluorescent staining identifies dead (red) versus alive (green) islets



Purified islets are cultured in flasks and incubated at constant temperature