

# Human Pancreatic Cell Autotransplantation Following Total Pancreatectomy

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During total pancreaticoduodenectomy for chronic pancreatitis, four patients received an intraportal pancreatic mixed-cell autograft prepared by collagenase digestion. The technique of this autotransplantation procedure was successfully developed using a normal canine pancreas, but has proved difficult to apply in the human chronic pancreatitis model. Our four patients became insulin-dependent, with proof of intrahepatic insulin production in only one patient. Three factors have contributed to the lack of graft success: 1) the preoperative endocrine status, 2) systemic hypotension and portal hypertension secondary to graft infusion, and 3) difficulty applying the successful technique in a normal dog pancreas to an extensively scarred human pancreas. The preoperative insulin response during a glucose tolerance test was blunted or delayed in the three patients tested. An immediate decrease in blood pressure and rise in portal pressure occurred in every patient and prevented infusion of the entire graft (30–50%) in three patients. Unfortunately, the patient with the most compromised insulin status was the only patient able to receive the entire graft. Our experience would indicate that further refinements in technique are necessary to prevent the vascular reaction and allow infusion of the entire graft. Furthermore, normal islet cell function is necessary before a successful graft can be expected.

**T**RANSPLANTATION OF THE ENDOCRINE pancreas from cadaver donors to diabetic recipients remains an attractive goal. In the instance of surgically-induced diabetes secondary to total pancreatectomy for chronic pancreatitis, preservation of endocrine function by autotransplantation would also prove useful. We have investigated autotransplantation techniques not hampered with immune rejection and, therefore, have only one major problem: preservation of sufficient viable endocrine tissue. An intrasplenic autotransplantation canine model, which was routinely successful in our laboratory,<sup>1</sup> was developed after the method of Mirkovitch.<sup>2</sup> An intraportal transplant in the dog, however, was unacceptable because of fatal portal hypertension and systemic hypotension. We were encouraged by our successful canine pancreatic

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autotransplants and the first human intraportal autotransplant after 95% pancreatectomy, performed in Minnesota<sup>3</sup> on February 14, 1977, and proceeded to the first human pancreas autotransplant following a total pancreatectomy on November 15, 1977. Since that time, three factors have directly affected the success of our human autografts: 1) the status of preoperative endocrine function, 2) the inability to transplant the entire graft because of marked portal hypertension, and 3) difficulty applying the successful techniques for a normal dog pancreas to the scarred gland of human chronic pancreatitis.

## Case Reports

**Case 1.** On November 15, 1977, a 45-year-old man with calcified chronic pancreatitis and chronic pain underwent total pancreatectomy with preservation of the pylorus.<sup>4</sup> A preoperative intravenous glucose tolerance test was normal, but simultaneous insulin levels were not obtained. The pancreas to the left of the portal vein was initially removed for processing with collagenase digestion, exactly as in the intrasplenic dog model.<sup>1</sup> The remainder of the pancreatectomy was performed while the autotransplant was prepared. The 60 cc graft was suspended in Ringer's lactate solution. A slow portal vein infusion was accompanied after five minutes with a systemic drop in blood pressure from 110 mmHg to 60 mmHg. Portal pressure was not measured. Infusion of the autotransplant was immediately terminated and the blood pressure quickly returned to preinfusion levels after five minutes with volume expansion. Approximately 30% of the processed autograft had been infused. Postoperatively, the patient was diabetic immediately but there were no complications, and the patient was pain free. A subsequent follow-up examination two years after operation indicated that the patient required 25 units of isophane insulin daily with good glucose control and has regained his normal weight on adequate exocrine replacement.

**Case 2.** A 50-year-old man with chronic calcific pancreatitis secondary to alcohol ingestion, experienced abdominal pain over a seven year period. A preoperative intravenous glucose tolerance test was normal but the insulin response was blunted (Fig. 1). Because of a 20% loss of body weight over the previous year, three weeks of total parenteral nutrition was administered three weeks before the operation. Intraoperative pancreatography was unsuccessful because

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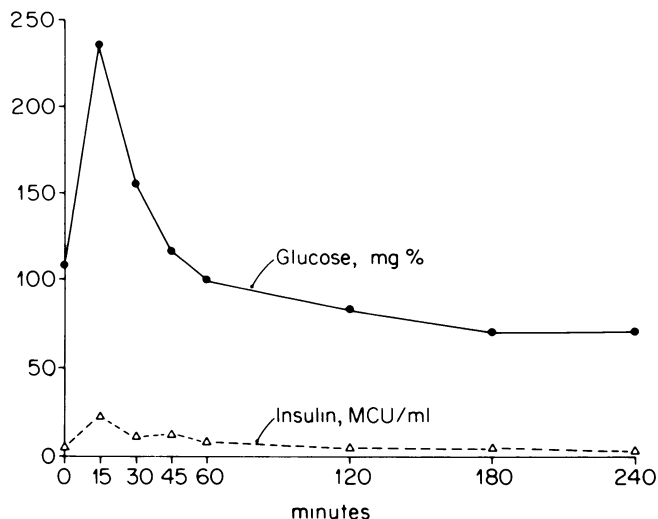


FIG. 1. A preoperative intravenous glucose tolerance test (0.5 g/kg of glucose) with insulin levels in Patient 2. A blunted insulin response in spite of a normal glucose curve indicates compromised endocrine function (see Fig. 5).

the pancreatic duct was completely obliterated by calcium deposits and fibrosis. A total pancreatectomy was performed with preservation of the pylorus on July 24, 1978. The entire shrunken pancreas was processed with collagenase digestion. The 50 cc graft was suspended in 500 cc of fresh frozen plasma with 3,000 units of heparin, and slow infusion into the portal vein was initiated. A systemic hypotension occurred from 110 to 70 mmHg, which reversed with volume replacement and ephedrine. Portal pressure rose from 12 cm H<sub>2</sub>O to 30 cm H<sub>2</sub>O within ten minutes, but subsided to less than 20 cm H<sub>2</sub>O after ten more minutes. Over the next hour, the reaction recurred after each infusion until portal pressure finally increased above 50 cm H<sub>2</sub>O and the infusion was abandoned. At that point, we had placed approximately 50% of the graft. The portal pressure slowly declined to normal during closure of the abdomen.

Postoperative liver enzymes (SGOT and SGPT but not alkaline phosphatase) were twenty times that of normal values. These levels declined to normal by the seventh day after operation. The patient required insulin on the second day after operation, while receiving 5% glucose intravenous fluids. There were no complications, and on the sixteenth day after operation, fasting levels of glucose and insulin were obtained from the inferior vena cava (IVC), via femoral vein catheterization, above and below the hepatic veins and also selectively from the left and right hepatic veins (Fig. 2). Orange juice was then administered, and 30 minutes later the study was repeated. Suprahepatic IVC insulin levels increased after the orange juice was administered. The hepatic vein wedge pressure was not elevated. The patient was free of abdominal pain and was discharged from the hospital. After discharge from the hospital, the patient was administered 20 units of isophane insulin a day. One year postoperatively, he was still free of pain, but died after an insulin overdose and a protracted hospital course.

**Case 3.** A 16-year-old girl complained of abdominal pain intermittently since age 2. She had undergone two previous pancreaticojejunostomy procedures for a diffusely dilated ductal system without calcifications, thought secondary to congenital absence of the duct of Wirsung. After each operation, the pain returned within four to six months. The patient presented with chronic abdominal pain necessitating oral meperidine HCl several times a day. A preoperative oral glucose tolerance test demonstrated a diabetic curve and a delayed rise of insulin (Fig. 3). A total pancreatectomy with pylorus preservation was performed on September 6, 1978. Once again, the gland was processed and suspended in heparinized (3000 units) fresh frozen plasma and slowly infused into the portal vein. Portal pressure transiently rose from 8 to 40 mmHg, but over the next hour, the entire transplant was able to be infused. Systemic hypotension once again occurred, but was also transient and was controlled with volume expansion and a slower infusion of cellular suspension. Postoperatively, the patient was free of pain and required insulin immediately. There were no complications. The IVC-hepatic vein study was obtained on the eighth day after operation, but showed no endocrine function (Fig. 4). Sixteen months after operation she was free of pain and required 32 units of isophane insulin a day.

**Case 4.** A 40-year-old man presented with a nine-year history

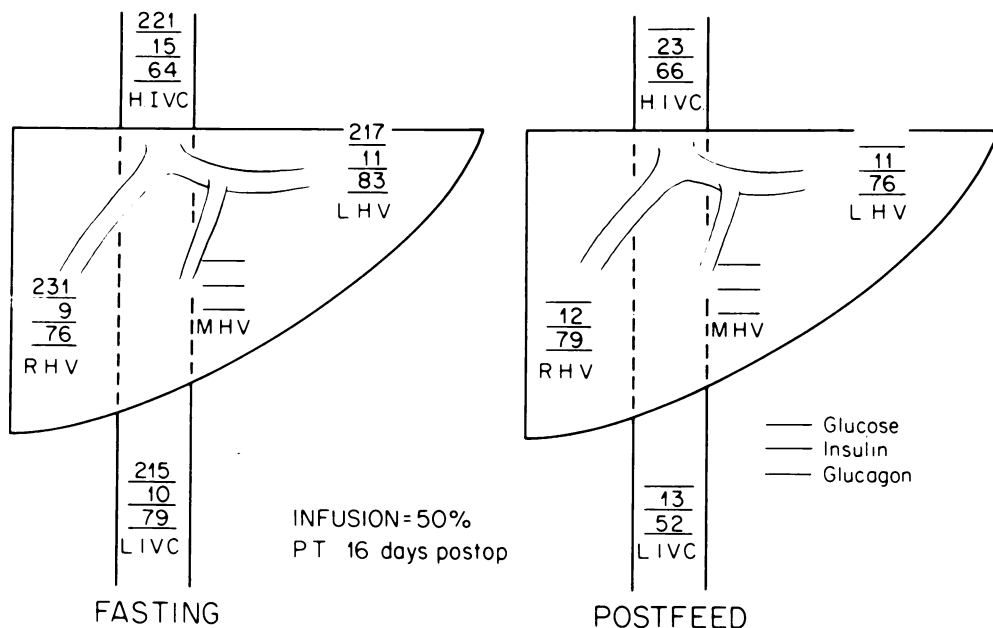


FIG. 2. In Patient 2, insulin levels ( $\mu$ U/ml, normal: 6–26) were measured in blood on the sixteenth day after operation from the lower inferior vena cava below the liver (LIVC), high inferior vena cava above the liver (HIVC), right hepatic vein (RHV), and left hepatic vein (LHV). Infusion of 50% of the autograft was possible before termination secondary to elevated portal pressure above 50 cm H<sub>2</sub>O. Glucagon levels (pg/ml, normal: 50–200) were not significantly changed. Post-feed means after orange juice, see text. Glucose in milligrams per decaliter, normal: 70–100.

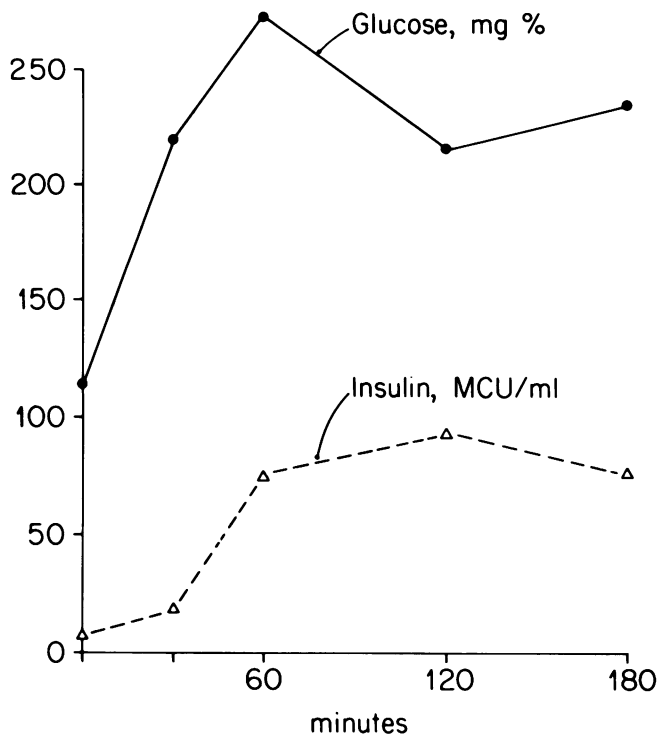


FIG. 3. A preoperative oral glucose tolerance test with 0.5 g/kg of glucose in Patient 3 showed a chemical diabetes curve as well as a delayed insulin response.

tension to 70 mmHg and an elevated portal pressure from 8 to 40 cm H<sub>2</sub>O occurred. Before operation, a Swan-Ganz catheter had been inserted to monitor cardiac output, which initially was stable but decreased from 7.35 to 5.14 L/minute toward the end of the graft infusion. Further discussion of the hemodynamic parameters and anesthesia management of this patient are presented elsewhere.<sup>5</sup> Insulin was required immediately after operation, but there were no postoperative complications. The IVC-hepatic veins study showed no production of insulin from the liver. When the patient was discharged from the hospital, he was receiving 15 units of isophane insulin a day and was pain free.

**Discussion**

This experience confirms the theory that endocrine function is already compromised when a patient finally undergoes operation to relieve the pain of chronic pancreatitis. Preoperatively, patients with this disease have a 24% incidence of diabetes, while 11% are administered insulin as we had previously observed.<sup>6</sup> However, the incidence of a prolonged glucose tolerance test or an inadequate insulin response during a glucose tolerance test (patients 2 and 4) is probably much higher. It is unknown if our present autograft technique can preserve enough endocrine function to prevent insulin use under these compromised conditions, because the entire graft cannot be placed into the portal vein secondary to portal hypertension. A similar vascular response has occurred in our canine model, but it is much more prominent and leads to death. If the entire graft could have been placed in patients 1, 2, and 4, a successful graft might have occurred. Unfortunately, the only autotransplant to be totally infused (Case 3) was associated with a preoperative prolonged glucose tolerance test and failure. The reactive systemic hypotension and portal hyper-

of chronic abdominal pain from alcoholic calcific chronic pancreatitis. An oral glucose tolerance test was slightly prolonged with a blunted insulin response. On April 19, 1979, the patient underwent a total pancreatectomy, the pancreas transplant was prepared with collagenase digestion, and the graft suspended in heparinized (3000 units) saline. Infusion of less than 50% of the graft was possible over the next hour, again, because of a transient systemic hypo-

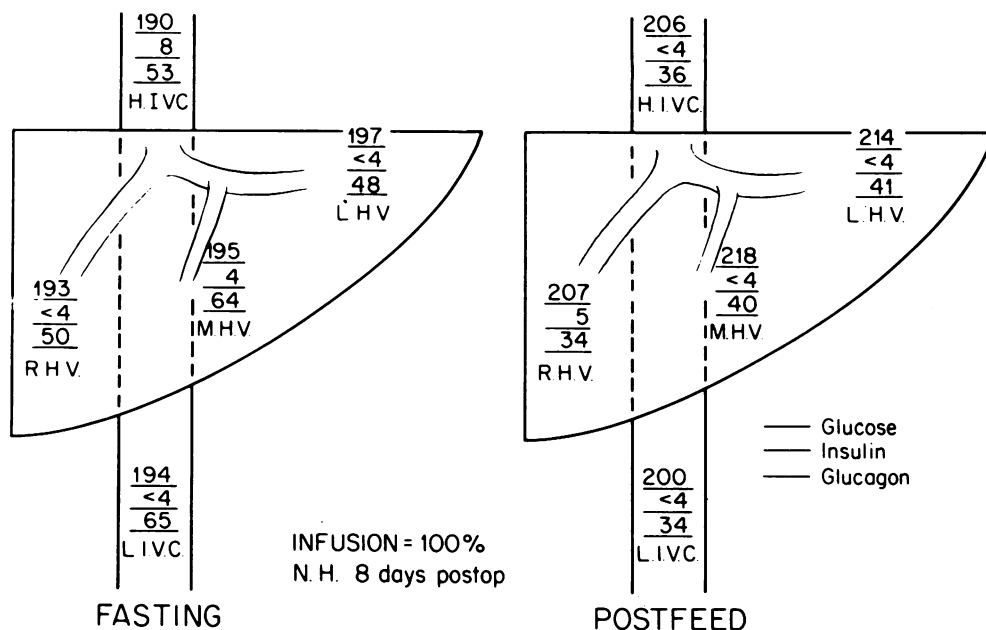


FIG. 4. On the eighth day after operation, in Patient 3, an inferior vena cava blood study was obtained showing no production of insulin or glucagon from the liver. Abbreviations and hormone level units are the same as in Figure 2.

tension have been recently investigated in our laboratory.<sup>7</sup> Initially pancreatic cell fragments were thought to act as emboli in the portal vessels, resulting in portal hypertension. Centrifuged and millipore filtered supernatant from the collagenase digestion, however, would still produce the vascular response, but not undigested minced pancreas or collagenase alone. In the dog pancreas, there appears to be a cold and heat stable substance elaborated during digestion with collagenase which, in very small doses, probably activates the kinin system. Commercially available bradykinin was found to mimic the vascular response. Using similar techniques, Mehigan and co-workers<sup>8</sup> have also reported this vascular response associated with disseminated intravascular coagulation (DIC) in canine and human autotransplants.<sup>8</sup> These authors found that the use of heparin (30 units/ml of graft) and aprotinin (200 units/ml of graft) in the canine autograft would decrease the portal hypertension elevation and also prevent the coagulation abnormalities of DIC. We routinely sterilize our collagenase solutions by filtering, and heparinize the autograft before placement in the portal vein, and have had no coagulation or bleeding problems. We agree with the use of heparin. Aprotinin will block kinin activation in humans, but it does not prevent the vascular response in dogs, even in high doses. Activation of the canine kinin system is by a direct splitting of kininogen, and is not adequately blocked by aprotinin. Indirectly activated human kininogen is mediated by activation of Hageman factor, prokininogenase, or plasminogen and is blocked by aprotinin.<sup>9</sup> Perhaps the routine use of systemic heparin (by blocking Hageman factor) by the Minnesota group<sup>10</sup> has accounted for the absence of this hypotensive syndrome in their series, even though their techniques are otherwise similar to this study as well as the Hopkins group.<sup>8</sup>

The postoperative evaluation of a pancreatic autograft after total pancreatectomy would indicate graft success if an increased level of insulin was found in the suprahepatic vena cava. Indeed, this was found in Case 2 but the amount of insulin produced was not sufficient to prevent the use of exogenous insulin in the postoperative period. The serum level of endogenous insulin determined by radioimmune assay will become falsely lowered once a patient receives exogenous insulin, due to the production of insulin antibodies. C-peptide insulin assays then need to be obtained. The IVC-hepatic vein study used in this study did not measure C-peptide insulin because exogenous insulin had been administered, at most, for 15 days. This time period is well before the six weeks required before insulin antibodies appear.<sup>11</sup>

Other investigators have attempted autotransplants

into the portal vein following a 95% pancreatectomy for chronic pancreatitis.<sup>8,12</sup> Without autotransplantation, the majority of patients after this subtotal pancreatectomy will be diabetic. The evaluation of graft success after subtotal pancreatectomy cannot use the IVC insulin method unless the graft was infused exclusively into the right or left branch of the portal vein. Elevated insulin levels in selective hepatic vein samples would then provide evidence for graft success. Cameron and co-workers have suggested the use of simultaneously obtained hepatic vein (via the IVC) and portal vein (transhepatic catheter) samples following 95% pancreatectomy.<sup>13</sup> With this method, elevated hepatic vein insulin levels above those in the portal vein would indicate a functioning graft in the liver.

The number of pancreatic autotransplants increased as North American and European surgeons began using and perfecting the technique. As of this writing, 45 pancreatic cell autotransplants have been reported to the Human Pancreas Islet Transplant Registry.<sup>14</sup> The difficulty with these data is determining how much pancreas was removed. Thirty-seven patients have received at least a 90% pancreatectomy and 17 patients are reported to be insulin-dependent. Of these 37 patients, nine underwent total pancreatectomies (four at UCLA, four in East Berlin, and one in Minnesota), and only one patient is insulin-independent (East Berlin). The preoperative endocrine status and stage of chronic pancreatitis is not known for all of these cases; however, the results with insulin independence would suggest that some islet cell function in the remaining pancreas is contributing to a better success rate. Our results were compromised by both inadequate preoperative endocrine function and the inability to infuse the entire autograft due to portal hypertension. The former may prove to be more important. Others<sup>10</sup> have not routinely experienced the vascular response as we have, even though similar techniques were used, but have not met with improved graft success either. Mehigan and colleagues<sup>15</sup> investigated this problem using pancreatic duct ligation to produce a canine chronic pancreatitis model. Six weeks after operation, the resulting fibrosed glands were autotransplanted, using the same techniques as the successful canine model with a normal pancreas. The chronic pancreatitis glands could not be successfully transplanted.

Further studies by these authors<sup>16</sup> have shown that graft success also varied with the commercial collagenase sources and minced particle size. Kretschmer and colleagues<sup>17</sup> showed that minced normal pancreas without collagenase digestion is not successfully transplanted, but requires an optimum digestion of 20 minutes before the tissue is successfully transplanted.

able. We have shown that commercial collagenases activate pancreatic proteolytic activity,<sup>18</sup> and have felt that the function of collagenase was to promote an endogenous tryptic digestion. An additional benefit would be the depletion of exocrine enzymes in the graft decreasing local autodigestion in the transplant site. Perhaps this is verified by the poor success rate in the human and dog chronic pancreatitis autografts, as acinar atrophy will decrease the presence of sufficient endogenous trypsin digestion. Already compromised endocrine tissue buried in fibrosis cannot acquire adequate nutrition by diffusion in their new transplant site, and might die before revascularization can occur. Calcifications were present in three of our four patients, indicating long-standing disease and acinar atrophy. The three successful University of Minnesota cases did not have calcified glands.<sup>19</sup>

### References

1. Traverso LW, Abou-Zamzam AM, Tompkins RK. The effect of a preoperative elemental diet on canine pancreatic autografts. *Surg Forum* 1978; 29:378.
2. Mirkovitch V, Campiche M. Successful intrasplenic autotransplantation 1976; 21:265.
3. Sutherland, DER, Matas AJ, Najarian JS. Pancreatic islet cell transplantation. *Surg Clin North Am* 1978; 58:365-382.
4. Traverso LW, Longmire WP. Preservation of the pylorus during pancreaticoduodenectomy. *Surg Gynecol Obstet* 1978; 146:959.
5. Torres LE, Traverso LW, Sohn YZ. Intraoperative hemodynamic changes in patients undergoing pancreatic islet autotransplantation. *Anesthesiology* 1980; 53:67-69.
6. Traverso LW, Tompkins RK, Urrea PT, Longmire WP. Surgical treatment of chronic pancreatitis: 22 years' experience. *Ann Surg* 1979; 190:312-319.
7. Traverso LW, Gomez RR, Wise WR, Dixon RS. Investigation of a shock factor obtained from canine pancreatic autografts. Unpublished data.
8. Mehigan DG, Bell WR, Zuidema GD, et al. Disseminated intravascular coagulation and portal hypertension following pancreatic islet autotransplantation. *Ann Surg* 1980; 191:287-293.
9. Vargaftig BB, Giroux EL. Mechanism of clostripain-induced kinin release from human, rat, and canine plasma. *Adv Exp Biol* 1975; 70:157-175.
10. Najarian JS, Sutherland DER, Rynasiewicz JJ, et al. Total or near total pancreatectomy and islet autotransplantation for treatment of chronic pancreatitis. *Ann Surg* 1980; 192:526-542.
11. Berson SA, Yalow RS, Bauman A, et al. Insulin-I<sup>131</sup> metabolism in human subjects: Demonstration of insulin binding globulin in the circulation of insulin treated subjects. *J Clin Invest* 1956; 35:170-190.
12. Najarian JS, Sutherland DER, Matas AJ, Goetz FC. Human islet autotransplantation following pancreatectomy. *Transplant Proc* 1979; 11:336-340.
13. Cameron JL, Mehigan DG, Harrington DP, Zuidema GD. Metabolic studies following intrahepatic autotransplantation of pancreatic islet grafts. *Surgery* 1980; 87:347-350.
14. Sutherland DER. International human pancreas and islet transplant registry. *Transplant Proc.* 1980; 12(no. 4, suppl. 2) 229-236.
15. Mehigan DG, Zuidema GD, Eggleston JC, Cameron JL. Pancreatic islet autotransplantation: results in dogs with chronic duct ligation. *Am J Surg* 1980; 139:170-174.
16. Mehigan DG, Zuidema GO, Cameron JL. Pancreatic islet autotransplantation in dogs: critical factors in technique. *Am J Surg* In press.
17. Kretschmer GJ, Sutherland DER, Matas AJ, et al. Autotransplantation of pancreatic islets without separation of exocrine and endocrine tissue in totally pancreatectomized dogs. *Surgery* 1977; 82:74-81.
18. Traverso LW, Abou-Zamzam AM. Activation of pancreatic proteolytic enzymes by commercial collagenases. *Transplantation* 1978; 25:226-227.
19. Sutherland, DER. Personal communication.