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Laparoscopic Biopsies in Pancreas Transplantation

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As there is no precise laboratory test or imaging study for detection of pancreas allograft rejection, there is increasing interest in obtaining pancreas tissue for diagnosis. Pancreas allograft biopsies are most commonly performed percutaneously, transcystoscopically, or endoscopically, yet pancreas transplant surgeons often lack the skills to perform these types of biopsies. We have performed 160 laparoscopic pancreas biopsies in 95 patients. There were 146 simultaneous kidney–pancreas biopsies and 14 pancreas-only biopsies due to pancreas alone, kidney loss, or extraperitoneal kidney. Biopsies were performed for graft dysfunction (89) or per protocol (71). In 13 cases, an additional laparoscopic procedure was performed at the same operation. The pancreas diagnostic tissue yield was 91.2%; however, the pancreas could not be visualized in eight cases (5%) and in 6 cases the tissue sample was nondiagnostic (3.8%). The kidney tissue yield was 98.6%. There were four patients with intraoperative complications requiring laparotomy (2.5%) with two additional postoperative complications. Half of all these complications were kidney related. There were no episodes of pancreatic enzyme leak and there were no graft losses related to the procedure. We conclude that laparoscopic kidney and pancreas allograft biopsies can be safely performed with very high tissue yields.

Abbreviations: PTA, pancreas transplant alone; SPK, simultaneous pancreas kidney

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Introduction

Over the last 2 decades, pancreas transplantation has experienced marked improvements in graft survival in all three categories. Current immunosuppression protocols are associated with significantly lower 1-year rejection rates compared to the pre–mycophenolate mofetil and tacrolimus era (1). However, immunological causes continue to contribute to pancreas graft loss posttransplant (2–4). In pancreas transplantation, serial creatinine and pancreatic enzyme determinations are typically used to monitor for pancreatic rejection and dysfunction. However, serum creatinine is only useful in simultaneous pancreas–kidney (SPK) recipients, and even in these patients isolated pancreas rejection has been described, thereby limiting changes in serum creatinine as a reliable marker alone (5). Pancreatic enzyme elevations are associated with pancreatic rejection; however, numerous other nonrejection causes have been identified such as native pancreatitis, bowel obstruction, allograft enzyme leak, and cytomegalovirus graft pancreatitis (5,6), in which the empiric treatment of rejection without a biopsy could be counterproductive. Moreover, rejection grade and type and therefore therapy cannot be predicted by the degree of enzyme elevations. Thus, despite decades of research to identify sensitive and specific noninvasive laboratory tests or imaging studies for detection of graft rejection (7), tissue biopsies remain the "gold standard."

Pancreas biopsies have been performed percutaneously, transcystoscopically, endoscopically, laparoscopically, or by open laparotomy with differing results and complication rates. Percutaneous, transcystoscopic, and endoscopic biopsies often rely on the availability and skills of personnel in other ancillary departments such radiology, urology, and gastroenterology. On the other hand, laparoscopic techniques are familiar to, and frequently performed, by transplant surgeons and could be used to perform organ allograft biopsies. Whether these can safely and consistently be performed with adequate tissue yields and without imaging guidance has not been demonstrated.

Over the past 4 years, we have routinely performed laparoscopic kidney and pancreas biopsies in pancreas transplant recipients without radiological guidance. The objective of the present study is to describe the surgical technique, tissue yields, and complications associated with a large series of wedge and needle laparoscopic

pancreas and kidney biopsies performed at a single center.

Study Design and Biopsy Techniques

This is a retrospective review of a prospectively collected database. We evaluated indications for biopsy, yield of tissue samples, as well as intraoperative and postoperative complications.

Surgical Technique: After general anesthesia and intubation, a Foley catheter was placed. Carbon dioxide was insufflated through an Optiview® port placed in the left upper quadrant to achieve abdominal distension. Two 5-mm trocars were placed in the right upper quadrant and in the midline incision below the umbilicus. When necessary, a limited lysis of adhesions was performed in order to visualize the grafts. The kidney biopsy was performed percutaneously by a core biopsy needle (16G) under direct visualization, and hemostasis was achieved by compression with the aspiration cannula and cautery (Figure 1). Usually two samples were obtained for histopathological assessment. Then the tail of the pancreas was visualized after dissecting overlying bowel or omentum and a wedge biopsy was performed using scissors and cautery (Figure 2). The wedge biopsy was typically 3–5 mm in length. In cases in which the surface of the pancreas appeared fibrotic, we performed an additional needle biopsy to have a deeper sample. No drains were used. After surgery, a 1-kg weight was positioned over the kidney to provide compression for a few hours.

Results

From October 2011 to December 2015, we have attempted 160 pancreas biopsies in 95 patients after either SPK or pancreas transplantation alone (PTA) (85 and 10 cases, respectively). Biopsies were performed because of suspected rejection (89) or per protocol (71). Protocol biopsies were performed in the case of high immunological risk patients, solitary pancreas transplants, postrejection treatment surveillance, or monitoring after changes in immunosuppression, BK viremia, and appearance of donor-specific antibodies. Thirteen cases had an additional procedure performed at time of biopsy (seven incisional hernia repairs, five laparoscopic cholecystectomies, and one ovarian cyst removal). In 14 cases, only a pancreas biopsy was performed because of PTA (10 cases), kidney loss in SPK recipient (3 cases), and an extraperitoneal kidney in a pancreas after kidney recipient (Table 1).

The median time for the procedure was 48 min (26–94 min) for cases without simultaneous procedures. Ninety-four percent of the patients were in condition to be discharged home the day after the procedure (many of them stayed to receive medical treatment for rejection, BK nephropathy, etc.).

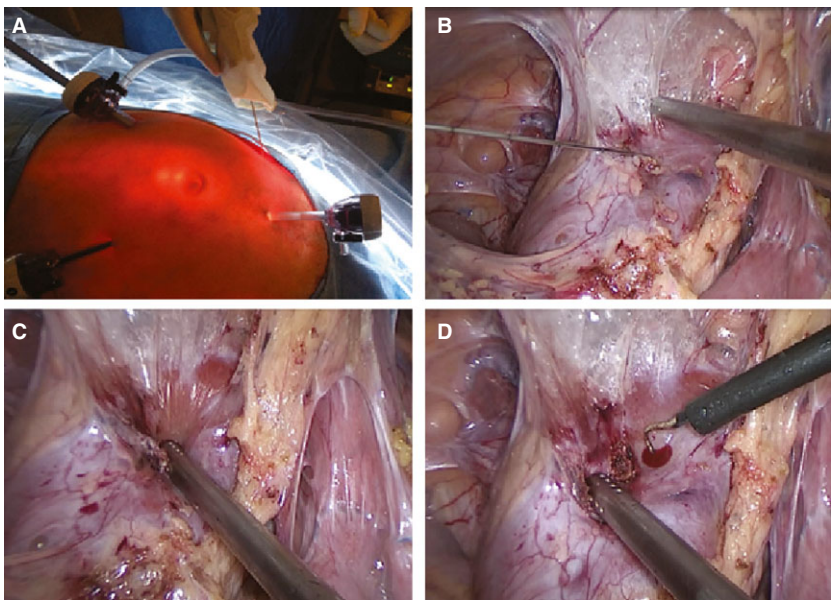

Figure 1: Laparoscopic kidney biopsy. (A) Trocar placement. Note camera port in left upper quadrant to visualize kidney in left lower quadrant. Direct needle insertion through the skin. Other ports include lower midline and right flank ports. This port arrangement is effective for biopsying both kidney in left lower quadrant and pancreas in right lower quadrant. (B) Tru-cut needle insertion into kidney. (C) Direct pressure compression of biopsy site. (D) Direct cautery of biopsy site.

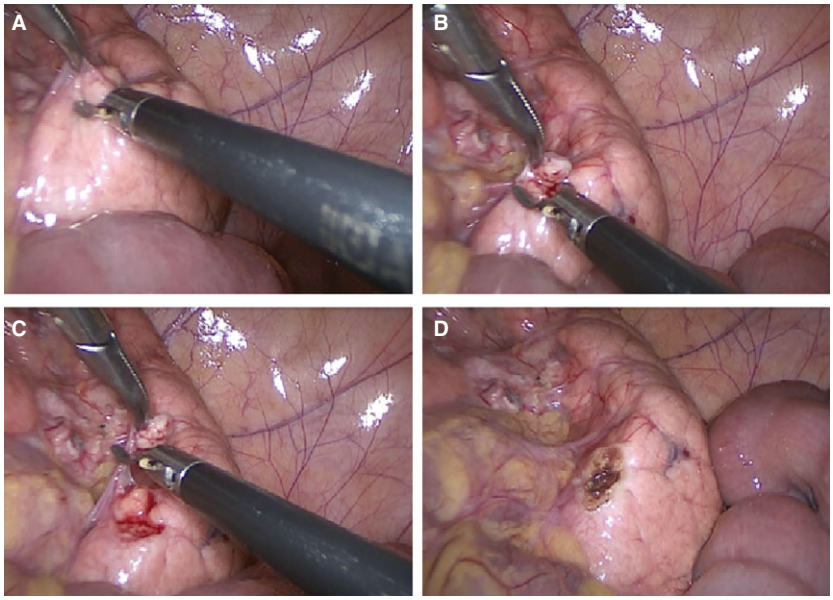

Figure 2: Laparoscopic pancreas wedge biopsy. (A–C) Wedge biopsy of pancreatic allograft parenchyma with scissors. (D) Cautery sealing of biopsy site.

Table 1: Results

	Kidney	Pancreas
Biopsy attempts	146	160
Attempts excluded	1	1
Attempts analyzed	145	159
Failed attempts	2 (1.4%)	8 (5%)
Nondiagnostic samples	0 (0%)	6 (3.8%)
Laparoscopic (Lap) tissue yield	143 (98.6%)	145 (91.2%)
Open biopsies	1 (0.7%)	4 (2.5%)
Lap + open tissue yield	144 (99.3%)	149 (93.7%)
Intraoperative complications	2 (1.2%)	2 (1.2%)
Postoperative complications	1 (0.6%)	1 (0.6%)
Graft losses	0 (0%)	0 (0%)

We performed 160 pancreas biopsy attempts. In one case, laparoscopy diagnosed an intestinal obstruction that was the cause of the pancreatic enzyme elevation and a laparotomy was performed for treatment, leaving 159 attempts for analysis. Of these 159 cases, we were not able to perform the pancreas biopsy laparoscopically in 8 patients because of extensive adhesions (in 6 cases), the presence of an *en-bloc* pediatric kidney–pancreas transplant that did not allow us to reach the pancreas (in 1 case), and in the other case the pancreas biopsy was not done because of a laceration of the graft duodenum. In four of these eight patients, increased pancreatic enzymes and suspected pancreas rejection were the indications for biopsy, and therefore we proceeded to open biopsy during the same anesthesia through a small laparotomy over the pancreas graft. Thus, in the remaining 151 cases, the pancreatic allograft was visualized and tissue was obtained laparoscopically without imaging support. Of these, six samples did not contain sufficient pancreatic tissue for adequate histopathological diagnosis of rejection; five were classified as adipose tissue and one as lymphoid tissue. Adequate pancreatic tissue that allowed a diagnosis to be made was achieved in 91.2% of biopsies (145 of 159 cases). Pancreas pathology reports included normal grafts in 97 cases, Banff Borderline rejection in 5 cases, Banff grade 1 rejection in 22 cases, Banff grade 2 rejection in 2 cases, acute humoral rejection in 1 case, and chronic changes without acute rejection in 18 cases.

We attempted 146 simultaneous kidney biopsies during the same surgical procedure as the laparoscopic pancreas biopsy. One case was excluded (same case discussed earlier in pancreas biopsy results), leaving 145 attempts for analysis. In two cases we were not able to reach the kidney laparoscopically due to extensive adhesions. In one case a percutaneous kidney biopsy was performed at a later time after the laparoscopic procedure. In the other case, kidney dysfunction was the reason for biopsy but a percutaneous biopsy was not possible because of overlying viscera around the kidney, and therefore an open biopsy though a small laparotomy overlying the kidney graft was performed during the same procedure. In the remainder of the kidney biopsy attempts, tissue was obtained for proper diagnosis, representing a yield of 98.6% (143 of 145 cases).

We experienced four intraoperative complications (2.5%). There were two cases of laceration of the graft duodenum.

In both cases, a small laparotomy was performed over the site to oversew the duodenum, and both healed without further complications. The other two intraoperative complications were kidney bleeding at the biopsy site that could not be stopped laparoscopically and required a small laparotomy over the kidney for hemostasis. Both of the cases of laparotomy for control of post–kidney biopsy hemorrhage were in the same patient in the presence of severe humoral rejection.

The postoperative complication rate was 1.2% with two cases. One patient had gross hematuria requiring a three-way Foley catheter and saline irrigation for 48 h, and one patient required relaparoscopy to drain a hematoma due to bleeding from a trocar site. No graft was lost due to a biopsy procedure complication. There were no episodes of pancreatic enzyme leak, pancreatic fistula, or pancreatic ascites postoperatively. Follow-up was performed with abdominal ultrasound at discharge, at first visit, and every 6 months, which failed to reveal any new peripancreatic fluid collections.

Discussion

As the pancreas transplant field continues to evolve, pancreas transplantation has now achieved very high graft and patient survival rates, and now compares with other solid organ transplants. However, close graft monitoring continues to be difficult to accomplish as there are few sensitive and specific laboratory tests or imaging modalities for pancreas rejection.

Although widely used, serum amylase and lipase levels are not 100% reliable. Troxell et al showed that abnormal pancreatic enzymes were present in only 8 out of 16 biopsy-proven pancreas rejection patients (50%). Furthermore, pancreatic enzymes paralleled the subsequent control biopsy in only 43% of the cases, making control biopsy extremely important in graft assessment after rejection therapy (8).

Surrogate duodenal biopsies have been performed in both bladder- and enteric-drained patients (9). Some centers are now using special surgical techniques with duodenoduodenostomies or proximal duodenojejunostomies to allow enteroscopic duodenal biopsies. However, the published experience is limited and the concordance of findings in the duodenum and pancreas were found in only 36% of the cases (10).

Surrogate kidney biopsies are widely used in simultaneous pancreas–kidney transplantation patients. In a series of simultaneous kidney and pancreas biopsies, Troxell et al (8) reported 16 patients with pancreas rejection, with only 10 of them with concurrent kidney rejection (62.5%). Likewise, the experience from the University of Wisconsin corroborates this finding (5).

In a recent report by de Kort et al, unrecognized antibody-mediated rejection has been suggested to play a role in early pancreas graft loss, generally attributed to technical failure (2). Moreover, grade and type of rejection, which merit differing therapies, cannot be distinguished on clinical grounds alone.

Consequently, obtaining pancreas tissue for diagnosis is essential and represents the "gold standard" in pancreas graft rejection diagnosis. Pancreas graft biopsies have been obtained transcystoscopically, enteroscopically (transduodenal), percutaneously, laparoscopically, and by open laparotomy. Transcystoscopic biopsies were performed in bladder-drained transplant patients with a yield of 56% for pancreas tissue and 87% when duodenal samples were included. Studies report gross hematuria rates in the range of 4% (11). In enteric-drained patients, an endoscopic ultrasound-guided transduodenal pancreas biopsy has been reported in a patient with a duodenoduodenostomy (12). However, these two techniques cannot be used in the majority of the pancreas transplants with a standard enteric drainage. Percutaneous biopsy of the pancreas graft was first described by Allen et al (13), and is currently the most used technique for obtaining pancreas tissue with some large series reported. The University of Maryland group (14) has reported 426 biopsies in 183 patients. Their tissue yield was 88%. Although the procedure was very safe in their hands, they reported complications including 1.2% of patients requiring laparotomy, three biopsies of other organs (including small bowel), and one pancreas graft loss. The University of Wisconsin group (15) reported 406 percutaneous biopsies with a tissue yield of 94% with only two patients requiring laparotomy and no graft losses. In a previously discussed report, the University of Minnesota group (11) reported a tissue yield of 73% in 93 percutaneous attempts with an incidence of pancreatitis and hemorrhage in 7% and 3% of patients, respectively. Laparoscopic biopsy was first described by the University of Minnesota group in 1996 on one case (16), followed by Silver et al with another case the next year (17). In 2002, Kayler et al reported a series of 12 biopsies in 11 patients with a yield of 91.6% and 1 patient requiring pancreatectomy due to a duodenal perforation (18). The present report is, to the best of our knowledge, the largest series of laparoscopic pancreas graft biopsies. Our pancreatic tissue yield was comparable to previously reported laparoscopic and percutaneous biopsy series (91.2%). The yield increased up to 93.7% when the four open biopsies are added. These were performed under the same anesthesia via a small incision of 1–2 inches just above the graft in a similar fashion as described by Kitada et al (19) with the advantage of having done the kidney biopsy laparoscopically.

Our intraoperative complication rate was also comparable to other percutaneous series (2.5%) and only half was pancreas related (1.25%). The two cases of graft duodenal laceration did require laparotomy for repair but did

not result in adverse sequelae or graft loss. We experienced only two cases of kidney bleeding, which could not be managed laparoscopically with compression and coagulation and required a small laparotomy. There were no pancreas or kidney graft losses due to these procedures. This is especially important when considering doing protocol biopsies. Furthermore, despite performing numerous pancreatic parenchymal wedge biopsies, we did not experience any cases of pancreatic enzyme leak, pancreatic fistula, or pancreatic ascites, which not only demonstrates safety of the wedge biopsy technique but also allays fears of developing these devastating complications. The demonstrated safety for doing wedge pancreatic parenchymal biopsies as described in the present study could provide an incentive for considering intraoperative (e.g. procurement, back table, or postreperfusion) biopsies, but added risks may be associated with biopsies in this situation and further studies should be conducted. While additional studies confirming the safety of pancreatic graft biopsies in the intraoperative setting are warranted, the prospect of safe intraoperative biopsies presents the important opportunity for studying aspects of pancreas graft outcomes relative to graft histology such as is often done in the setting of liver and kidney transplantation.

Laparoscopic biopsy also affords the surgeon the opportunity to simultaneously tackle other intraabdominal surgical disease posttransplantation. In our series, laparoscopy led us to an unexpected diagnosis of bowel obstruction because of a distal intestinal obstruction of a Roux-en-Y limb leading to surgical treatment. In another case, it warned us not to perform a percutaneous kidney biopsy because of overlying small bowel adhesions.

In conclusion, laparoscopic biopsies of both pancreas and kidney transplant grafts can be safely performed in patients after pancreas transplantation with very high yield and acceptable morbidity. In the case of pancreas graft biopsies, this was accomplished safely as a wedge biopsy without radiological imaging support under direct vision and without pancreatic enzyme leaks. In centers performing percutaneous pancreas allograft biopsies, this laparoscopic approach could be an excellent option when the former is unsuccessful.

Disclosure

The authors of this manuscript have no conflicts of interest to disclose as described by the *American Journal of Transplantation*.

References

1. Gruessner AC. 2011 Update of pancreas transplantation: Comprehensive trend analysis of 25,000 cases followed up over the course of twenty-four years at the International Pancreas Transplant Registry. Rev Diabet Stud 2011; 8: 6–16.
2. de Kort H, Mallat MJK, van Kooten C, et al. Diagnosis of early pancreas graft failure via antibody-mediated rejection: Single center experience with 256 pancreas transplantations. Am J Transplant 2014; 14: 936–942.
3. Gruessner AC, Gruessner RW. Long-term outcome after pancreas transplantation: A registry analysis. Curr Opin Organ Transplant 2016; 21: 377–385.
4. Kandaswamy R, Skeans MA, Gustafson SK, et al. OPTN/SRTR 2014 annual data report: Pancreas. Am J Transplant 2016; 16 (Suppl 2): 47–68.
5. Redfield RR, Kaufman DB, Odorico JS. Diagnosis and treatment of pancreas rejection. Curr Transplant Rep 2015; 2: 169–175.
6. Klassen DK, Drachengerg CB, Papadimitriou JC, et al. CMV allograft pancreatitis: Diagnosis, treatment and histological features. Transplantation 2000; 69: 1968–1971.
7. Margreiter C, Pratschke J, Margreiter R. Immunological monitoring after pancreas transplantation. Curr Opin Organ Transplant 2013; 18: 71–75.
8. Troxell ML, Koslin DB, Norman D, Rayhill S, Mittalhenkle A. Pancreas allograft rejection: Analysis of concurrent renal allograft biopsies and posttherapy follow-up biopsies. Transplantation 2010; 90: 75–84.
9. Margreiter C, Aigner F, Resch T, et al. Enteroscopic biopsies in the management of pancreas transplants: A proof of concept study for a novel monitoring tool. Transplantation 2012; 93: 207–213.
10. Nakhleh RE, Sutherland DE, Benedetti E, Goswitz JJ, Gruessner RW. Diagnostic utility and correlation of duodenal and pancreas biopsy tissue in pancreaticoduodenal transplants with emphasis in therapeutic use. Transplant Proc 1995; 27: 1327–1328.
11. Laftavi M, Gruessner AC, Bland B, et al. Diagnosis of pancreas rejection: Cystoscopic transduodenal versus percutaneous computed tomography scan-guided biopsy. Clin Transplant 1998; 65: 528–532.
12. Serwanska-Swietek M, Degowska M, Patena K, Perkowska A, Rydzewska G, Rydzewski A. Endoscopic ultrasound-guided biopsy of the pancreas in simultaneous pancreas kidney transplant recipient: Case report. Transplantation 2014; 98: e42–e43.
13. Allen RD, Wilson TG, Grierson JM, et al. Percutaneous biopsy of bladder-drained pancreas transplants. Transplantation 1991; 138: 303.
14. Klassen DK, Weir MR, Cangro CB, Bartlett ST, Papadimitriou JC, Drachenberg CB. Pancreas allograft biopsy: Safety of percutaneous biopsy – Results of a large experience. Clin Transplant 2002; 73: 553–555.
15. Niederhaus SV, Leverson GE, Lorentzen DF, , et al. Acute cellular and antibody-mediated rejection of the pancreas allograft: Incidence, risk factors and outcomes. Am J Transplant 2013; 13: 2945–2955.
16. West M, Gruessner RWG. Laparoscopic biopsy after pancreaticoduodenal transplantation: A first report. Transplantation 1996; 62: 1684–1687.
17. Silver JM, Vitello JM, Benedetti E. Laparoscopic-guided biopsy of pancreatic transplant allograft. J Laparoendosc Adv Surg Tech A 1997; 7: 319–322.
18. Kayler LK, Merion RM, Rudich SM, , et al. Evaluation of pancreatic allograft dysfunction by laparoscopic biopsy. Transplantation 2002; 74: 1287–1289.
19. Kitada H, Sugitani A, Okabe Y, et al. Availability of pancreas allograft biopsies via a laparotomy. Transplant Proc 2009; 41: 4274–4276.