Strategic Opportunities in Clinical Islet Transplantation

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More than 471 patients with type 1 diabetes have received islet transplants at 43 institutions worldwide in the past 5 years. High rates of insulin independence have been observed at 1 year in the leading islet transplant centers, and an international multicenter trial has demonstrated reproducible success of the approach. Loss of insulin independence by 5 years in the majority of recipients remains of concern, and immunosuppressant drug side effects necessitate stringent inclusion criteria for islet-alone candidates that have the most severe, unstable glycemic control despite optimal insulin therapy. The advent of new immunosuppressive drugs with superior side-effect profiles (e.g., LEA29Y and FTY720) may open up opportunities for more "islet-friendly" approaches. Future opportunities to expand the donor pool using living donor islet transplantation are within reach, and will be enhanced considerably with both donor and recipient adjunctive treatment using islet-specific growth-factors.

Keywords: Clinical transplantation, Islets, Immunosuppression (clinical and experimental), Immunosuppressive drugs, Experimental transplantation.

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Olle Korsgren and colleagues provide us with an astute view of the current state of clinical islet transplantation and the remarkable opportunities for improvement that lie ahead (*1*). We share Korsgren's enthusiasm, and provide our own perspective both on the potential challenges and exciting road ahead.

In our initial report of the Edmonton Protocol in 2000, we demonstrated 100% insulin independence in our first seven patients treated with a steroid-free, sirolimus-based immunosuppressant protocol (*2*). An unprecedented, exponential increase in clinical islet transplant activity followed, with an estimated 471 patients with type 1 diabetes treated at 43 institutions worldwide (Fig. 1). This represents a significant milestone, as more patients with type 1 diabetes have now received islet implants in the past 5 years than in the entire preceding 30-year history of islet transplantation.

Recent improvements have included: the routine use of the 'two-layer' oxygenated perfluorodecalin system for pancreas transportation (*3*, *4*); the development of component-based collagenase constituents for final blending at the time of pancreas digestion to improve enzyme stability and enhance the reliability of islet isolation; the routine use of insulin-transferrin-selenium CMRL-based islet culture while preparing the recipient for transplant (*3*, *5*); and the use of alternative immunosuppressive therapies in an attempt to enhance single donor islet transplant success (*3*, *6*). The safety of the percutaneous transhepatic intraportal approach to non-surgical islet delivery has been further enhanced by effective mechanical and physical methods to seal the catheter tract, thereby preventing risk of postprocedural bleeding (*7*, *8*).

The rates of insulin independence during the first year continue to be impressive at the three most active North American institutions, and in a recent combined analysis, 82% of a total of 118 recipients of completion transplants in Edmonton, Miami and Minnesota were insulin free at one year (Fig. 2) (*6*). The 'Immune Tolerance Network' recently supported the first multicenter trial in islet transplantation to study a cohort of 36 patients treated with the Edmonton Protocol at nine international sites. The preliminary data indicates that the protocol was successfully replicated, with >80% of recipients at the three most experienced sites achieving sustained insulin independence (*9*). Success was more variable (0%–63%) at the remaining sites, reflecting not only the unique challenges involved with the setting up of new islet isolation facilities but also experience with sirolimus-based immunosuppression (*9*, *10*).

We have treated a total of 70 islet-alone recipients at the University of Alberta since 1999. Sixty-nine patients are alive. One 43-year-old patient died unexpectedly at home at 21 months following transplant from a presumed sudden cardiac arrhythmia, in the absence of sepsis or transplant-related complications. Of our first four patients to reach 5 years with the Edmonton Protocol, one remains insulin free. Kaplan-Meier statistical projections demonstrate progressive loss of insulin independence over time leaving only 50% of patients still insulin free at 3 years. While insulin independence appears to wane in the majority of patients over time, 83% continue to demonstrate persistent islet function at 5 years when measured by C-peptide secretion (Kaplan-Meier). The benefits of persistent islet transplant function in the absence of

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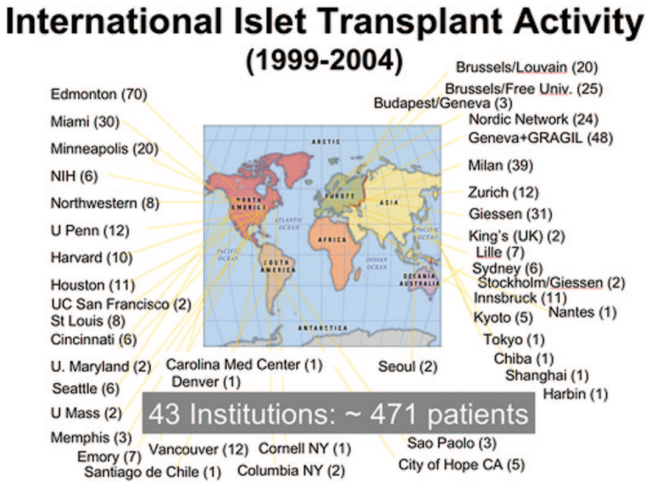

FIGURE 1. Exponential increase in clinical islet transplant activity within the most recent five years, with 471 patients transplanted at 43 international institutions.

insulin independence should not be entirely discounted; effective prevention of recurrent hypoglycemia or severe lability combined with correction in glycated HbA1c to a level far superior to that readily achievable with intensive insulin therapy, is seen as a substantial benefit (Fig. 3) (*11*). It remains to be seen whether stable improvement in glycemic control from a partially functional islet transplant can be justified against the real and potential risks of current lifelong immunosuppression (*12–14*).

The decay in rates of insulin independence observed in more long-term follow-up with the Edmonton Protocol provide not only a challenge but a unique opportunity to further define the biology for a better understanding of how to promote and sustain islet survival. We have currently embarked on an in-depth study of factors that are likely influencing decay in islet mass in our patients, and anticipate that the results of serial islet graft biopsies, and serological analysis of

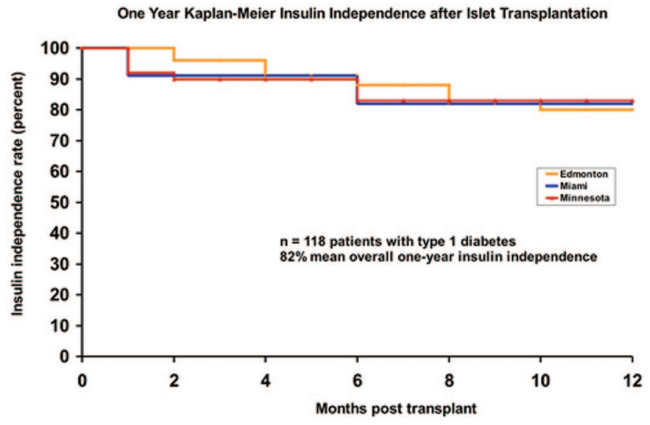

FIGURE 2. Kaplan-Meier survival curves showing a mean of 82% insulin independence in 118 consecutive islet-alone patients receiving 'completed' islet infusions at the Universities of Alberta, Miami and Minnesota (data generously provided by Dr. Camillo Ricordi and Dr Bernhard Hering for this composite analysis).

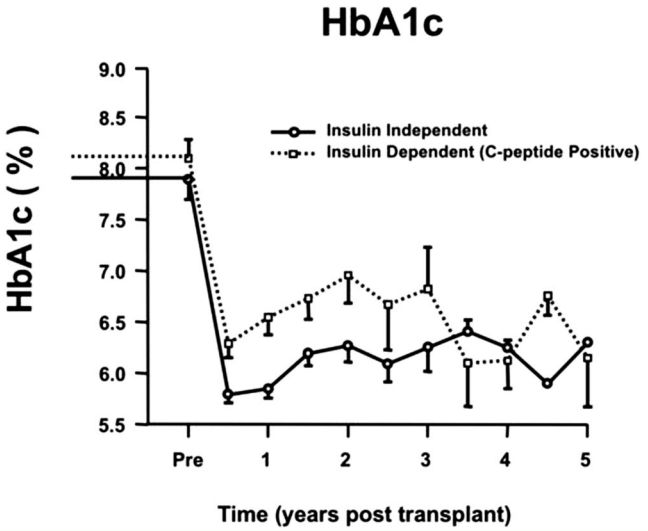

FIGURE 3. Glycated HbA1C is corrected in insulin independent islet-alone recipients, and improved in insulin-requiring, C-peptide positive islet recipients at the University of Alberta, 5-year follow-up.

donor sensitization, cytokine gene activity (granzyme B) and changes in autoantibody status will collectively provide instructive information over time. Possibilities for deterioration in islet mass over time include chronic allograft rejection, undiagnosed acute rejection, local islet toxicity from the drugs, recurrent autoimmunity and/or failure of islet regeneration over time—resulting from the antiproliferative properties of sirolimus (*15*).

While the combined use of sirolimus and low-dose tacrolimus has helped move islet transplantation forward from clinical curiosity to effective therapy for many more patients, it is recognized that these drugs are still far from ideal. Sirolimus and tacrolimus have near-ubiquitous targets of distribution, and as a result lead to a number of side effects in islet recipients, including mouth ulceration, peripheral edema, a high rate of ovarian cysts in female recipients, increase in proteinuria in some patients with underlying preexisting diabetic renal damage; hypertension and hypercholesterolemia (*14*, *16*).

A number of exciting, emerging compounds with distinct mechanisms of action will shortly be entering pilot clinical islet transplant trials. These agents provide an opportunity to develop more "islet-friendly" approaches with fewer non-immune related side effects. Emerging opportunities include: 1) A potent, costimulatory signal blocker LEA29Y found to be highly effective in promoting islet survival in primate trials will be evaluated in Emory and Edmonton (*17*); 2) The lymphocytehoming agent FTY720 has proven to be highly effective in controlling autoimmunity in NOD mice and in promoting marginal mass islet transplants in primates and will be evaluated shortly in Miami, Minnesota, and Edmonton (*18*, *19*); 3) The combination of anti-thymocyte globulin and rituximab (anti-CD20) has been shown by Naji et al. to induce tolerance in primates, and will be explored shortly at the University of Pennsylvania; 4) The non Fc-binding hOKT3- gamma1-ala-ala antibody developed by Bluestone et al. has been effective in abrogating autoimmunity in new-onset diabetes, and have facilitated single-donor

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islet transplant success in ongoing trials at the University of Minnesota (*3*, *20*); 5) The T-cell depleting antibody alemtuzumab (Campath-1H) has shown promise in clinical solid organ transplantation, and is currently being evaluated in Edmonton; and 6) A potent, diphtheria-conjugated anti-CD3 immunotoxin combined with deoxyspergualin has provided remarkable results with robust tolerance induced and sustained for over 5 years in a series of monkeys treated by Thomas and colleagues at the University of Alabama (*21*). If these agents can provide equal or greater protection against both allo- and autoimmunity, and if the safety profiles prove to be superior to current therapies, the face of islet transplantation will likely be further transformed in the coming few years.

The disconnect between the number of potential organ donors and the potential need for islet replacement therapy in the 2 million people in North America with type 1 diabetes will only be addressed by more radical approaches. Data from UNOS currently indicate that only 23.8% of the potential 6,182 available US multiorgan donors were procured or used for pancreas or islet transplantation. This could be more effectively addressed by improved legislation and by education of the multiorgan retrieval teams.

Opportunities to pretreat the donor with anti-inflammatory and anti-apoptotic compounds such as 17β-estradiol or atorvastatin could potentially mitigate the negative impact of islet damage induced by brain-injury derived pro-inflammatory cytokines (Fig. 4) (*22*, *23*). Immune depletion of donor passenger lymphocytes by donor pretreatment with agents such as alemtuzumab may also enhance islet survival after transplantation by reducing immune sensitization.

Living donor islet transplantation will provide 'near-perfect' partial grafts for islet transplantation and trials are likely to move forward imminently. Avoidance of exposure to pro-inflammatory cytokines, immediate graft processing without cold ischemia, and the low anticipated tissue digest volume from a distal third pancreatectomy will likely eliminate the need for islet purification—all of which will substantially enhance the potency of the final islet preparation. It will be interesting to see if the increased potency will more than offset the reduced islet mass from the partial pancreas in terms of insulin independence rates. The use of laparoscopic surgery in the donor will enhance palatability for the approach (*24*). The potential risk of diabetes in the donor could be substantially reduced by avoidance of obese donors, by confirming a normal intravenous glucose tolerance test in the donor prior to acceptance, and by only accepting donors with negative autoantibody profiles (*25*). The surgical risk of pancreatic fistula in the donor is small but manageable. The opportunity to augment the islet mass both the donor in the months before and in the recovery phase after surgery, during islet culture and subsequently in the islet recipient using combination growth factors (including GLP-1, exendin-4, EGF, gastrin, INGAP, or hepatocyte growth factor), could further minimize the potential risk of diabetes in the donor, and could substantially enhance the rate of single donor islet transplant success in the recipient (*26–30*). Integration with the anti-thrombotic (IBMIR) strategies developed by Korsgren using nicotinamide, inactivated factor VIIa or low-molecular weight dextran sulphate during islet culture or in the recipient posttransplant to inhibit islet tissue factor expression, will further considerably enhance the success of the living donor approach (*1*, *31*, *32*).

In summary, phenomenal progress has occurred in the field of clinical islet transplantation in the most recent 4 years, with high one-year rates of insulin independence, and high 5-year rates of persistent C-peptide secretion. Loss of insulin independence over time still remains a concern with current protocols.

While the marketed antirejection drugs available today have had an acceptable safety profile in islet transplantation, the drug-related and dose-limiting side effects have proved to be a challenge in some patients. Remarkable opportunities lie ahead for development of successful living donor islet transplantation, improved engraftment, islet proliferation in vitro and in the recipient, and newer, more "islet-friendly" immunosuppressants with minimal nonimmune side effects. Given these opportunities, islet transplantation will be within reach for many more patients with type 1 diabetes including children, and will not be restricted to the most unstable patients as it is today.

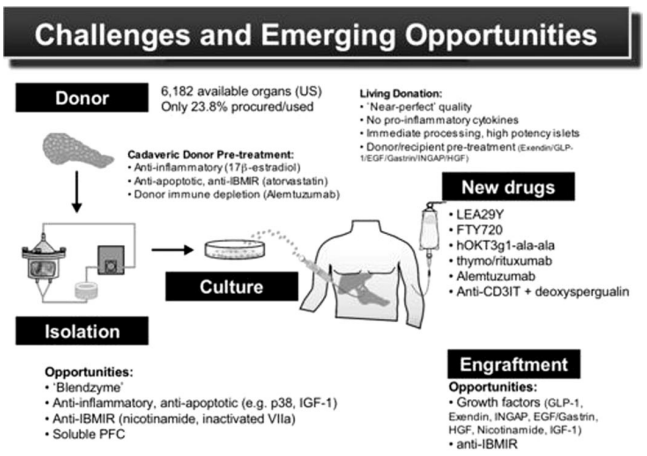

FIGURE 4. Challenges and strategic opportunities ahead in clinical islet transplantation.

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