# Islet Transplantation

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For those interested in islet transplantation, this meeting was an excellent opportunity to exchange new ideas, techniques, and experiences with investigators from all over the world. Although most presentations dealt with the use of adult islet tissue, significant papers describing the use of fetal islet tissue were also presented. The sessions were divided into the following topics.

## ISLET TISSUE: ISOLATION AND FUNCTION

**Documentation of islet tissue.** In terms of documentation studies, one presentation described that islets isolated from dogs from different sites in the pancreas may have a different susceptibility to the effects of hyperglycemia posttransplantation. A study on rat islets also demonstrated direct reduction of islet function in which islets were isolated from donors with prolonged hyperglycemia. Another study in dogs presented confirmation of the need to quantitate the yields of islet tissue after islet isolation. Two papers also dealt with fetal tissue documentation. The first confirmed the low quantity of insulin in the human fetal pancreas ≤12 wk gestational age and proposed a reason why 1st-trimester human fetal tissue may not be practical for human clinical trials. The second described development of a monoclonal antibody that may be focused to a direct cell type that could be important in β-cell replication.

**Islet processing.** Most reports dealt with technical aspects of islet isolation by collagenase digestion and various means of islet purification. In terms of adult islet tissue, there were many studies examining the collagenase digestion step. One was an analysis of the different component enzymes of collagenase to determine whether such an analysis could relate to collagenase's effectiveness in islet isolation. This is an important area that needs much additional effort. There were preliminary reports on new components to add to the digestive steps, including the addition of glycerol or methylcellulose to prevent gelatinous material and islet clumping. Another suggestion was to use Tyrode's solution to isolate islets to reduce their damage from the isolation process. The use of a selective exocrine toxin, selenomethionine, was described in an improved protocol that may be more practical for potential clinical application. Although comparisons between ductal distension and ductal perfusion of the human pancreas were made, a new automated digestion-filtration system was described that seems to offer the largest yield of human islet tissue so far. Islet isolation in different animal species was also reported for neonatal pig and beef. A new rapid supravital stain was also suggested for islets that may correlate with their viability.

As for purification techniques, the most dramatic reports involving new equipment described a modified cell sorter with a special islet chamber for human islet tissue and a second automatic gradient rotor. The apparent high cost of each of these devices will probably limit their applicability to islet processing to a few centers until additional feasibility is demonstrated. A thorough comparison of all the density gradients was reported as well as additional data on the use of dextran and bovine serum albumin as gradients. Additional data confirmed the potential use of exocrine cell–specific lectins modified for magnetic purification. Another report suggested development of antiexocrine cell monoclonal antibodies (MoAbs) for islet purification.

**Fetal islet tissue.** In terms of fetal reports, the most promising demonstrated an increase in growth potential of fetal islets by the use of nicotinamide in vitro. Additional reports on use of fetal tissue included the effect of culture conditions on fetal islet growth and the ability of fetal tissue to mature. The effect of the type of abortion done in 180 human abortions on the recovery of fetal islet tissue was also reported.

**Preservation of islet tissue.** Documentation of the deleterious effects of pancreatic warm ischemia and cold ischemia

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on islet isolation and function was presented. Although not specifically addressing islet isolation after pancreas preservation, the data of improved pancreas preservation with the University of Wisconsin (UW) solution holds promise for its potential use before islet isolation.

In terms of cryopreservation, there were many reports of successful protocols demonstrating the feasibility of islet banking with mouse, rat, dog, and pig islets, as well as adult human islets and fetal human islet tissue. A unique report was the vitrification of mouse islets. Two studies suggested that cryopreservation may selectively destroy the passenger immune cells and not the islet cells, adding this as another method for potential islet immunoalteration.

**Revascularization of islets.** Several studies opened a new area of important research in islet transplantation: the examination of the revascularization of islet tissue after transplantation. The use of microsphere technology suggested that early hyperglycemia posttransplant reduces islet vascularization. Microsphere studies also document that islet vascularization of intraportally transplanted islets seems to originate from the hepatic artery. Two new technologies offer the potential of observing revascularization of islet tissue in vivo. The first uses a chamber for vascular visualization monitored in the back muscle. The second uses in vivo fluorescence of the renal subcapsular site. Both are exciting new studies that enrich our understanding of this important area of research.

**Islet transplants in large animals.** The results of long-term splenic islet autografts in dogs document normal fasting glucose and insulin values and insulin sensitivity. However, glucose effectiveness is impaired, as demonstrated by glucose tolerance testing. Two studies confirmed the advantage of the splenic transplant site over other sites in dogs for partially purified islet tissue, and a third study suggested the importance of the type of islet preparation being transplanted into this site. Additional canine autotransplantation studies examined the portal site. Two studies examined the effect of immunosuppression on islet transplants in dogs. The first suggested that cyclosporin A is an effective reagent for allografts for dogs even with mild hyperglycemia. The second suggested failure of triple immunosuppression to support canine islet allografts. Another interesting report in spontaneously diabetic dogs suggests islet function is limited in the intraportal site.

A preliminary report on pig islet transplantation was presented, as well as reports on fetal pig autografts and allografts, showing partial function of intraportal islet transplants when sufficient quantities of islets are used.

## PREVENTION OF REJECTION

**Immunosuppression.** There were three presentations on the problem of islet toxicity to immunosuppressive agents. The first suggested that rat islets are deleteriously affected by the release of interleukin 1 as exposed in culture by loss of insulin secretion; this is a reversible effect. Another suggested a similar reduction in secretory capacity by azathioprine exposure in vitro. A third suggested that the experimental agent FK-506 is not harmful to human fetal islet tissue but that cyclosporin at high concentrations could reduce insulin secretion.

Four presentations addressed the effectiveness of immunosuppression on islet acceptance. The first demonstrated mild prolongation of canine islet allografts with antilymphocyte serum, cyclosporin, and azathioprine. The second confirmed earlier studies demonstrating that high doses of cyclosporin effectively prolong islet allografts; however, altered glucose tolerance tests were noted during its use. A third study examined cyclosporin for fetal rat pancreas and suggested that the immunosuppressant altered major histocompatibility complex (MHC) expression and prolonged rat survival. The fourth presentation evaluated the use of a new immunosuppressive agent, Ciamexon, in rats, finding its use as a single agent had a mild effect in minor processes but no effect in major allograft processes.

**Immunoalteration.** Several studies presented data examining the phenomenon of immunoalteration in terms of examining alterations taking place during the study. The first presentation demonstrated the potential of dispersed β-cells apparently devoid of class II antigen–presenting cells to be able to stimulate a cytotoxic T-lymphocyte response in a mixed lymphocyte cell-culture condition, which could be blocked by the addition of a class I modified antibody. The second study gave similar results. Another study presented data suggesting the destruction of pig proislet tissue in a mouse T-lymphocyte–dependent process. A detailed study of rat islets demonstrated the potential usefulness of immunoelectron-microscopic identification of MHC structures located on the surface of and within the cells. Another study suggested that human exocrine cells express HLA-DR/DP but not HLA-DQ, in contrast to previous studies in rats, and that islet cells lack all HLA-dependent markers.

Other studies examined the effect of differently expressed cell surface antigens on their immune response. One demonstrated that an increase in the expression of class I antigens in rat islets produced by γ-interferon or tumor necrosis factor increased in vitro islet damage when exposed to alloreactive cytotoxic T-lymphocytes. A second suggested that exposure of neonatal rat islets devoid of antigen-presenting cells to γ-interferon and supernatant from lymphocyte cultures resulted in increased class II expression with increased cell death. Another paper suggested new class II expression on islets undergoing rejection.

Several presentations addressed different treatment modalities to induce immunoalteration and acceptance of islet grafts. A series of studies examined the use of irradiation. The first suggested a marked prolongation of γ-irradiated mouse islets; the dosage required was strain dependent, and islet toxicity was also a problem. Three studies demonstrated similar effects of irradiation treatment. The use of MoAbs infused into the pancreas before transplantation of islets showed a decreased response in vitro and failed to prevent rejection. In studies in mice the repeated use of Lyt2 or L3T4 MoAbs led to prolonged allograft survival. Another study in mice confirmed the use of antilymphocyte serum and complement to the islets plus cyclosporin to the recipient to be an effective mode of treatment. One study found treatment of fetal islet tissue by immunotoxin to prolong allograft survival.

Four studies were presented in which the BB rat model was used to examine recurrent autoimmune diabetes. In one study the injection of OX8 or anti-AGM1 MoAbs prevented the recurrence of diabetes by engrafting Wistar-Furth islets.

The second study used MoAbs to interleukin 2 receptors to treat islets in combination with cyclosporin to prolong recurrence of diabetes in the BB rat after islet allografting. One study suggested that the use of multiple donors providing small quantities of islets to the same recipient in a sequential manner could be a potent method of providing graft acceptance.

**Immunoisolation.** Several participants presented various ways of preventing islet rejection by protecting the islets from cell-mediated destruction. Three presentations described the use of hollow fibers to prevent rejection. One examined rat islets with Cuprophan fibers; this study showed transient responses. The second study used Brockmann bodies from the fish species *Osphronemus gourami* enclosed in Amicon fibers or Thomaphor fibers in the diabetic rat. The transplantation of hollow fibers into the nonobese diabetic mouse also showed transient success with the use of induced islet tumor cells.

Several other studies examined the use of microencapsulation techniques. Transient reversal of diabetes in mice with the use of hamster islets was demonstrated by enclosing the islets in agarose gel capsules modified with oil. A different encapsulation technique used polyornithine alginate or polylysine alginate capsules to study the inflammatory and fibroblastic responses of the islets. Another new polymer, polyurethane-silicone, showed in vitro responsiveness with pig islets. One study reported on polylysine alginate capsules for transplantation of several islets.

## CLINICAL ISLET TRANSPLANTATION

Although most of the presentations were on animal and experimental studies, there were 10 poster summaries on the use of human fetal tissue. Four were from China, four from the USSR, one from Hungary, and one from Yugoslavia. The summaries reported 174, 414, 3, and 14 transplants respectively, for a total of 605 transplantations. These reports of transplantation with human fetal tissue are difficult to understand because so few details were given in a scientific manner. There is a great discrepancy between the claims of successful islet grafts from these countries and the lack of success from other countries. One major problem seems to be the lack of standardized reporting as demonstrated by these 10 poster summaries. An accepted method of reporting is imperative for future reports so the results can be accepted by investigators. Therefore, I offer a full approach to providing a standardized method of reporting these results (Table 1). The use of such criteria in reporting islet-transplantation studies to the scientific community will eliminate the confusion that now exists. Rational presentation of factual information and the sharing of meaningful results must be the objective of all who participate in future meetings.

TABLE 1
Criteria for documentation of successful islet transplantation

- A. Pretransplantation documentation
  - C-peptide level
    - Fasting
    - Poststimulation
  - Average insulin requirement
  - Glycosylated hemoglobin level
  - Average glucose level
  - 24-h profile
  - Degree of complications
- B. Peritransplantation documentation
  - Islet preparation
    - Yield
    - Pretreatment
    - Viability
    - Sterility
  - Pancreas donor
  - Islet treatment
    - Immunoalteration methods
  - Transplantation method
    - Transplant site
    - Transplantation technique
- C. Posttransplantation documentation
  - Prevention of rejection
  - Immunosuppression regimen
  - Immunoalteration regimen
  - Immunoisolation regimen
  - Rejection incidents and treatment
  - Islet function
    - C-peptide level
      - Fasting
      - Poststimulation
    - Insulin requirement
    - Glycosylated hemoglobin level
    - 24-h profile
  - Complications
- D. Experimental animals
  - Reliance on graft by its removal