RETROSPECTIVE ASSESSMENT OF ISLET CELL AUTOANTIBODIES IN PANCREAS ORGAN DONORS

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**Objective:** 3-4% of deceased pancreas donors may have autoantibodies (AAb) to pancreatic islet cell antigens, which are well-established markers of type 1 diabetes (T1D). We investigated whether donor AAb positivity could affect the outcome of pancreas transplantation.

**Research Design and Methods:** We retrospectively tested AAb in 135 donors whose pancreata and kidneys were transplanted in T1D patients. We measured AAb to glutamic acid decarboxylase (GAD-AAb), the tyrosine-phosphatase-like protein IA2 (IA2-AAb) and insulin (insulin-AAb). We then evaluated pancreas transplant outcome data.

**Results:** 4/135 (2.96%) donors were AAb positive: 3 donors had GAD-AAb and 1 donor had insulin-AAb. Their respective recipients became insulin-independent on follow-up. Three of the 4 recipients have normal, insulin-producing grafts 3-5.8 years after transplant. The recipient of the insulin-AAb positive donor developed chronic rejection following discontinuation of immunosuppression 3.3 years after transplant.

**Conclusions:** Single AAb positivity did not affect the outcome of pancreas transplantation in our study.
**Type 1 diabetes (T1D)** is an autoimmune disease resulting in β-cell loss and insulin-dependence (1). Autoantibodies (AAb) to several islet antigens are predictive and diagnostic for T1D (2). Presence of multiple AAb correlates with higher disease risk in first-degree relatives (3). Simultaneous kidney-pancreas (SPK) transplantation is therapeutic for T1D patients with end-stage renal disease (4). Studies suggest that 3-4% of organ donors have at least one AAb to islet cell antigens (5,6). It is not known whether AAb-positivity could be a donor-related factor affecting the outcome of pancreas transplants. We performed retrospective AAb testing in 135 deceased donors whose pancreata and kidneys had been transplanted in T1D patients and then verified clinical outcome.

**RESEARCH DESIGN AND METHODS**

We have performed 350 SPK transplants over the past 18 years, in T1D patients with end-stage renal disease. T1D diagnosis is routinely verified by lack of detectable C-peptide after a sustacal challenge. All pancreas transplants are bladder-drained (7). We retrospectively tested AAb in 135 donors (90 males and 45 females). The mean age was 25.8 years, age range was 1.9-51 years. Pancreata and kidneys from the tested donors were transplanted into T1D patients between 1998 and 2005. We measured AAb to glutamic acid decarboxylase (GAD-AAb), the tyrosine-phosphatase-like protein IA2 (IA2-AAb) and insulin (insulin-AAb) using standard radioimmunoassays. AAb levels are expressed as index levels calculated from the counts per minute of the test sample and the positive and negative control samples. Receiver operating curves identified assay cut-offs of 11.44, 3.72 and 6.85 for the GAD, IA-2 and insulin AAb assays, respectively. Our laboratory participated in the Diabetes Autoantibody Standardization Programs of the Immunology of Diabetes Society and Center for Disease Control in 2000, 2002, 2003 and 2005 (8). Donors and recipients were HLA-typed using standard serology. The Institutional Review Board approved the study.

**RESULTS**

Four of 135 (2.96%) donors were AAb-positive: 3 donors had GAD-AAb, 1 donor had insulin-AAb. Donors with GAD-AAb had low AAb levels. Donor# 4 had markedly elevated insulin-AAb levels. No donor had IA-2-AAb or multiple AAb. Table I shows the characteristics of the AAb-positive donors and corresponding recipients. Two donors with GAD-AAb were homozygous for the HLA-DR4 or -DR3 susceptibility alleles; the remaining GAD-AAB-positive donor carried a presumably protective HLA-DR2. The donor with insulin-AAb had neutral HLA types.

We then evaluated outcome data from the respective recipients. Our SPK recipients have a mean follow-up of 5 ± SD 2.1 years. All patients transplanted with a pancreas from a single AAb-positive donor became insulin-independent; 3/4 patients transplanted with a pancreas from an AAb-positive donor have normal, insulin-producing grafts 3-5.8 years after transplant (Table I). The recipient of GAD-AAb-positive donor#1 had a pancreas transplant biopsy 3.2 years after transplantation showing no β-cell loss, insulitis or other abnormalities. This recipient had elevated GAD-AAb levels preceding the transplant which persisted essentially unchanged during follow-up. The recipient of the insulin-AAb-positive donor (#4) developed chronic rejection following discontinuation of immunosuppression 3.3 years after transplant. At that time GAD-AAb were transiently positive. The patient returned
to insulin dependency despite maintaining residual C-peptide secretion for up to 2.2 years after developing chronic rejection. The patient’s last C-peptide level was 2.3 ng/ml. Loss of graft function did not differ among recipients of AAb-positive and AAb-negative donors (1/4 vs 12/131, p=0.33).

CONCLUSIONS
There is interest in screening pancreas donors for autoantibodies to identify prediabetic pancreata which may not be suitable for transplantation and could be made available for research (5). The Juvenile Diabetes Research Foundation is supporting large scale screening to identify AAb-positive pancreas donors for research (www.jdrfnpod.org). A recent analysis of 25-60 year-old pancreas donors from the general population showed that single AAb positivity is not commonly associated with insulitis and β-cell loss analyzing ~0.5 cm³ biopic fragments of pancreata that were used for islet cell isolation (6). Insulitis was found only in 2 donors who were positive for 3 AAb and not in 59 donors positive for 1-2 AAb.

We identified 4 donors with a single AAb, consistent with the reported frequency in organ donors (5). Our data include younger subjects compared to previous studies (5,6): 55% of our donors were <25 years-old, an age group with higher T1D incidence. Indeed, this group yielded 3 of the 4 AAb-positive donors. Our analysis is unique in providing transplant outcome data from patients who received a pancreas from a single AAb-positive donor. All patients became insulin-independent on follow-up. In a patient who continues to be euglycemic, a biopsy performed 3 years after transplantation did not evidence islet damage. The recipient of the insulin-AAb-positive pancreas lost transplant function due to chronic rejection related to noncompliance. Overall, our outcome data are consistent with biopsy data from previous studies showing that single AAb positivity may not always be associated with clinically significant autoimmunity and β-cell damage in organ donors (5,6). The findings are consistent with the low diabetes risk associated with single AAb positivity in the general population (9,10). Relevant to clinical pancreas transplantation, our data suggest that single autoantibody positivity is unlikely to affect pancreas transplant outcome, and may help refining strategies for ongoing pancreas donor AAb screening initiatives, of which we remain strong supporters. Limited access to human pancreata with ongoing autoimmunity remains a major obstacle to advance our understanding of human T1D.

ACKNOWLEDGEMENTS
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REFERENCES

Table I. Age and HLA types of all donors screened, AAb status, age, HLA types and outcome data for AAb positive donor/recipient pairs

**Age distribution and HLA-DR types of the pancreas donors**

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<th>Age Groups (years)</th>
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<th>Non-DR3/4</th>
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<th>DR4/X</th>
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<td>3</td>
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<td>11</td>
<td>7</td>
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<tr>
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<td>4</td>
<td>12</td>
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<td>14</td>
<td>7</td>
<td>3</td>
<td>3</td>
<td>1</td>
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<tr>
<td>Total</td>
<td>135</td>
<td>75</td>
<td>26</td>
<td>31</td>
<td>3</td>
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</table>

**AAb status, age, HLA types and clinical outcome data for AAb positive donors and respective recipients**

<table>
<thead>
<tr>
<th>Donors</th>
<th>Positive AAb</th>
<th>Index Levels</th>
<th>Age</th>
<th>HLA</th>
<th>Clinical Outcome</th>
<th>Recipients</th>
<th>Age</th>
<th>HLA</th>
<th>GAD AAb</th>
<th>IA-2 AAb</th>
<th>Follow-up (years)</th>
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<td>41</td>
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<td>#1 NGT</td>
<td>4.4 48 DR3/4</td>
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<td>+</td>
<td></td>
<td></td>
<td>4.4 48 DR3/4</td>
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<tr>
<td>#2 GAD</td>
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<td>22</td>
<td>DR1/2</td>
<td>#2 NGT</td>
<td>3 38 DR3/4</td>
<td>#2 NGT 38 DR3/4</td>
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<td>-</td>
<td></td>
<td></td>
<td>3 38 DR3/4</td>
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<tr>
<td>#3 GAD</td>
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<td>5</td>
<td>DR3/3</td>
<td>#3 NGT</td>
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<td>5.8 39 DR3/7</td>
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<tr>
<td>#4 Insulin</td>
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<td>19</td>
<td>DR6/7</td>
<td>#4 PCR</td>
<td>6.8 44 DR1</td>
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<td>6.8 44 DR1</td>
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1SPK Recipient # 4 expressed GAD AAb transiently following chronic rejection.
NGT: Normal Glucose Tolerance
PCR: Pancreatic Chronic Rejection