Transplantation of Microencapsulated Neonatal Porcine Islets in Patients with Type 1 Diabetes: Safety and Efficacy

R.B Elliott, O Garkavenko, P Tan, N.N Skaletsky, A Guliev, B Draznin

Auckland, New Zealand; Moscow, Russia; Denver, CO, USA
Presenter Disclosure

Boris Draznin, MD, PhD

Board Member/Advisory Panel: none
Consultant: none
Employee: none
Research Support: Amylin; Sanofi-Aventis; Novo-Nordisk
Speaker’s Bureau: none
Stock/Shareholder: none
Other: none
• Destruction or severe malfunction of pancreatic islets is the core pathogenetic feature of diabetes

• Cure of diabetes is impossible without restoration of pancreatic islet – β-cell – function

• Pancreatic β – cells are the only cells capable of sensing ambient blood glucose and converting this information into regulated insulin secretion
Strategies to Restore β-cell Function

- Islet transplantation
- Stem cell derived insulin producing cells
- Non-islet cells genetically modified
- Islet cell regeneration
• T1DM - > 2,000,000 patients
• T2DM - >20,000,000 patients in the U.S. alone
• Only around 1,000 cases of allogenic islet transplantation
Islet Transplantation

Intrahepatic allotransplantation with immune suppression is not the answer to treatment of Type 1 diabetes
Porcine Islets

Pro:
• Porcine insulin
• Essentially unlimited supply of cells for transplantation
• Islets respond to the same physiological range of glucose as human islets
• Potential to do implants without immunosuppression
• Ethical considerations

Con:
• Humans express high titers of antibodies against Galactose (1,3) α-galactose residue present in most pig cells
• Retroviruses
Pig Viruses

- Pig Hep E
- Pig circovirus type 2
- Cytomegalovirus
- Pig lymphotrophic herpesvirus
- Others
Pig Endogenous Retrovirus (PERV)

- Part of the pig genome (with more than 100 of per-viruses)
- PERV- A and PERV- B are infections to human cells in vitro
Prerequisites for Xenotransplantation

- Cells must be free from any xenotic agent
- Islets must be uncontaminated, undamaged, free of exocrine tissue
- Anti-rejection strategy should not be based on immunosuppression
Cells Free From any Xenotic Agent

- Source herd must be free of infections capable of being transmitted to man
- Specific Pathogen Free animals (SPF)
- Must be housed in the Designated Pathogen Free (DPF) facilities
- Must be checked for infection status frequently
Discovered in 1806; Area 220 sq miles; Temp 35-65° F; humid, cloudy and very windy.
<table>
<thead>
<tr>
<th>Virus</th>
<th>Prevalence in general pig Population (PCR)</th>
<th>Prevalence in BioCert herd (PCR)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&gt; 20 weeks</td>
<td>&gt;20 weeks</td>
</tr>
<tr>
<td></td>
<td>7 days</td>
<td>7 days</td>
</tr>
<tr>
<td>PCMV</td>
<td>70%</td>
<td>Not detected</td>
</tr>
<tr>
<td>PLHV</td>
<td>95%</td>
<td>Not detected</td>
</tr>
<tr>
<td>PCV1</td>
<td>Not detected</td>
<td>Not detected</td>
</tr>
<tr>
<td>PCV2</td>
<td>96%</td>
<td>Not detected</td>
</tr>
<tr>
<td>HepEV</td>
<td>90% (14 weeks)</td>
<td>Not detected</td>
</tr>
<tr>
<td>EMCV</td>
<td>Not detected</td>
<td>Not detected</td>
</tr>
<tr>
<td>Conventional pathogen</td>
<td></td>
<td>Not detected</td>
</tr>
<tr>
<td>AuJD</td>
<td>Not detected</td>
<td>Not detected</td>
</tr>
<tr>
<td>BVD</td>
<td>Not detected</td>
<td>Not detected</td>
</tr>
<tr>
<td>PPV</td>
<td>Present</td>
<td>Not detected</td>
</tr>
<tr>
<td><em>Toxoplasma gondii</em></td>
<td>Not detected</td>
<td>Not detected</td>
</tr>
<tr>
<td>Leptospiroses</td>
<td>Present</td>
<td>Not detected</td>
</tr>
<tr>
<td><em>Mycoplasma hyopneumoniae</em></td>
<td>Not detected</td>
<td>Not detected</td>
</tr>
</tbody>
</table>

- Not detected
- Present
Islet Isolation

- Stress to the tissue
- Harsh enzymatic digestion
- Encapsulation
- Storage
- Transport
- Viability at the time of transplantation
Uncontaminated
Undamaged

Free floating islet day 15 culture. Insulin producing cells red Zinc staining DTZ.
Viable

Porcine Free Islets AOPI staining

Viability > 95%
Shelf Life Stability (BR103-109)

Islet Viability

Data are mean + SD
Differentiation in Neonatal Islets

- **Insulin**
  - **day 1** (free islets)
  - **day 4** (free islets)
  - **day 35** (encapsulated islets)

- **Glucagon**
  - **day 1** (free islets)
  - **day 4** (free islets)
  - **day 35** (encapsulated islets)
Exocrine Free (amylase)
Microencapsulation

• Surrounding of islet cells with a highly biocompatible biopolymer called alginate which reduces the host’s immune response to the implanted islets

• Alginate coat allows insulin, glucose, oxygen and other nutrients to diffuse freely, while blocking antibodies and T-cells
NanoBioCapsule Attributes

Strong, Elastic Physical Barrier

Unrestricted Cell Viability

Allow Inward Nutrient Diffusion

Outward Protein and Metabolite Release

Immunoisolation

Control Internal Cell Attachment

Biocompatible
DIABECELL® Encapsulated Neonatal Porcine Islets
LCT’s Encapsulation

Other alginate capsule [90days]

LCT capsule [215 days]
Patients and Methods

- 8 Patients with T1 DM
- Age 23 – 63
- Duration of diabetes 5 to 15 years
- Dose of neonatal islets between 5,000 and 10,000 Islet Equivalents per Kg body weight
- Delivered laparoscopically into the lesser sac of omentum
Clinical Data
Phase I/IIa 2007 – 2010
Sklifasovsky Institute, Moscow, Russia

Subjects
• 8 adult Type 1 diabetes patients
• Insulin dependent > 5 years

Dose
• 5,000 – 10,000 islet equivalents/kg
• Up to 3 repeat implants
## Results of the First Human Trial

<table>
<thead>
<tr>
<th>Patient No.</th>
<th>No. of Implants</th>
<th>Follow-up (Weeks)</th>
<th>Insulin Dose (Units/day)</th>
<th>% Dose Reduction</th>
<th>HbA1c (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Pre-Implant</td>
<td>3-6mo after</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Pre-Implant</td>
<td>3-6mo after</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>3</td>
<td>96</td>
<td>113</td>
<td>76</td>
<td>33</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>3</td>
<td>84</td>
<td>22</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>3</td>
<td>72</td>
<td>66</td>
<td>53</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>3</td>
<td>60</td>
<td>30</td>
<td>27</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>2</td>
<td>30</td>
<td>68</td>
<td>58</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>1</td>
<td>20</td>
<td>41</td>
<td>51</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>2</td>
<td>18</td>
<td>37</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>1</td>
<td>18</td>
<td>83</td>
<td>83</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Results

- Two out of 8 patients became insulin independent
- Insulin dose was reduced by 34%
- HbA1c was reduced from 8.86% to 6.91%
- Viable islets were recovered from 5 patients
- Porcine insulin was detected
- No side effects and no xenosis
Insulin Detection in Post HPLC Eluates
Patient# 1 before and after glucagon stimulation

- Human Insulin
- Porcine Insulin
Efficacy – Reduction in HbA1c (mean)

- Mean reduction in HbA1c:
  - 6 mo: 1.9%
  - 12 mo: 1.8%
  - 18 mo: 1.4%
  - 24 mo: 1.3%

Number of Patients:
- 6 mo: 8
- 12 mo: 5
- 18 mo: 3
- 24 mo: 1
Patient Implant – Recovered Cells
Safety Within a Week of Transplant

Significant adverse events - NONE

2 pts – abdominal discomfort – resolved in 5 days

2 pts – creatinine elevations to 1.5 for 3 days

2 pts – low grade fever for 3 days
Safety at 24 Months

- No significant adverse events to date
- PERV RNA - negative
- PERV DNA - negative

Preliminary Efficacy

- Improved blood glucose control with reduced HbA1c
- Reduced daily dose of insulin injections
- Two patients off insulin up to 32 weeks
- Intact capsules retrieved after 6 months
- Pig insulin detected in patient blood
Candidates for Xenotransplantation

- “Brittle diabetes” with or without other complications
- Severe hypoglycemia with attempts to optimize care
- Hypoglycemic unawareness

7 patients in New Zealand have received transplants in the past 6 months.
Summary

• Safety objectives have been met

• Proof of concept in humans – possible to achieve therapeutic success without immunosuppression

• Future questions – the effect of the appropriate dose of islets on magnitude and duration of response
Conclusions

• Careful manufacturing of encapsulated islets from neonatal pigs yields a product that shows significant promise as a treatment of T1DM without immunosuppression

• Regulatory concerns around PERV appear to be a non-issue. Other xenoses can be avoided
Thanks for your kind attention...