‘Islet Transplantation: From Research to Reality’

Paul R V Johnson

Director of Oxford Islet Transplant Programme
UK ANNOUNCES NEW 50P COIN TO COMMEMORATE BREXIT
• Rationale for Islet Transplantation
• Methods
• Current Results
• Ongoing Challenges / Future Opportunities
Rationale
Ideal Treatment for T1DM (once it has developed)

1) Restore true normal glucose homeostasis by coordinated islet hormone release (rather than just manage glycaemic excursions)

2) ‘Switch off’ ongoing autoimmune destruction

3) Replace islet-cells that have been destroyed

4) Prevent any secondary complications or reverse them if they have already developed

5) Treatment well-tolerated + appropriate for children
Tight blood sugar control reduces the risk of developing microvascular diabetes complications. The evidence of benefit is mainly from studies in younger patients at early stages of the disease. Benefits need to be weighed against risks including severe hypoglycaemia, and patient training is an important aspect in practice. The effects of tight blood sugar control seem to become weaker once complications have been manifested. However, further research is needed on this issue. Furthermore, there is a lack of evidence from RCTs on the effects of tight blood sugar control in older patient populations or patients with macrovascular disease. There is no firm evidence for specific blood glucose targets and treatment goals need to be individualised taking into account age, disease progression, macrovascular risk, as well as the patient’s lifestyle and disease management capabilities.
Advancing Technology

1922
First commercial insulin.

2. Control System
3. Insulin and glucagon administration
1. Glucose Measurement

Subcutaneous delivery
Insulin
Glucagon

HYPODERM
ILETIN
31.99 UNITS IN 5 C.C.

Iletin is obtained from pancreas and is the active principle in this solution.
This solution should be kept in a refrigerator.

ELI, LILLY & COMPANY, INDIANAPOLIS.
<table>
<thead>
<tr>
<th>Treatment Aim</th>
<th>Technology</th>
<th>Transplantation</th>
</tr>
</thead>
<tbody>
<tr>
<td>1) Potential to restore normal glucose homeostasis</td>
<td>(✔)</td>
<td>✔</td>
</tr>
<tr>
<td>2) Prevent /Reverse secondary complications</td>
<td>✔</td>
<td>✔</td>
</tr>
<tr>
<td>3) Replace lost beta-cells</td>
<td>X</td>
<td>✔</td>
</tr>
<tr>
<td>4) Switch off autoimmunity</td>
<td>X</td>
<td>✔</td>
</tr>
</tbody>
</table>
Whole Pancreas Transplantation
UK SPK Results

Pancreas graft survival
Log-rank (1 year) p=0.3

Patient survival
Log-rank (1 year) p=0.5

<table>
<thead>
<tr>
<th>Year Range</th>
<th>N</th>
<th>1 Year</th>
<th>3 Year</th>
<th>5 Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>2007-2008</td>
<td>319</td>
<td>88 (84-91)</td>
<td>81 (76-84)</td>
<td>77 (72-81)</td>
</tr>
<tr>
<td>2009-2010</td>
<td>252</td>
<td>84 (79-88)</td>
<td>76 (70-81)</td>
<td></td>
</tr>
<tr>
<td>2011-2012</td>
<td>268</td>
<td>83 (77-87)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Patient Survival

!"#$%&'$( )*+%,. *%&$/)'1 * , &2-. ) *+% #/). " , 2'2+0
### SPK vs PTA

#### % Graft survival

<table>
<thead>
<tr>
<th></th>
<th>1 year</th>
<th>3 year</th>
<th>5 year</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>SPK</strong></td>
<td>88%</td>
<td>86%</td>
<td>84%</td>
</tr>
<tr>
<td><strong>PTA</strong></td>
<td>79%</td>
<td>70%</td>
<td>70%</td>
</tr>
<tr>
<td><strong>PAK</strong></td>
<td>76%</td>
<td>74%</td>
<td>74%</td>
</tr>
<tr>
<td><strong>2nd Tx</strong></td>
<td>57%</td>
<td>50%</td>
<td>50%</td>
</tr>
</tbody>
</table>

#### Months post-transplant

*Cold Spring Harb Perspect Med 2014;4:a015610*
Whole Pancreas Tx

- Very good results when combined with renal graft with high levels of insulin independence and stabilisation / reversal of secondary complications

- Still major procedure, with significant morbidity and mortality

- Unlikely to ever be applicable to children without $2^\circ$ complications

- Is Risk / Benefit of PTA justifiable for treating hypoglycaemic unawareness alone?
Whole Pancreas vs Islets

< 5 ml
Islet Transplantation

- Endocrine pancreas only
- Minimally invasive with minimal morbidity or mortality
- Can restore normal glucose homeostasis with coordinated secretion of all islet hormones
- Potential to immunomodulate islet graft or promote strategies for immune tolerance
- Techniques all potentially applicable to children
Islet Transplant Options

• **Diabetes with hypoglycaemia-unawareness**
  – Islet transplant alone (ITA)

• **Diabetes with renal failure**
  – Simultaneous islet + kidney (SIK)
  – Islet after kidney (IAK)

• **Surgically-induced diabetes**
  – Islet Auto-transplantation
• Diabetes with hypoglycaemia-unawareness
  – Islet transplant alone (ITA)

• Diabetes with renal failure
  – Simultaneous islet + kidney (SIK)
  – Islet after kidney (IAK)

• Surgically-induced diabetes
  – Islet Auto-transplantation
Whole Pancreas vs Islets!
‘Beta Cell Replacement’

Whole Pancreas Transplantation, Nephrology
Islet Transplantation, Diabetology,
Stem-Cell Biology, Bioengineering, Nanotechnology
Methods
Human Islet Isolation

2 stages:

- Collagenase Digestion (releasing islets)
- Density-Gradient Purification (separating islets)
Islet Isolation and Islet Transplantation
Dissection and Perfusion
Pancreas Digestion
Density-Gradient Purification
Why purify?

20 – 50 ml

< 5 ml
Culture

• Logistics
• Quality testing
• Patient pre-treatment
• Percutaneous transhepatic approach via portal vein (or laparoscopic / mini-lap.)
• Antibiotic and Heparin cover
• Infuse over 20-30 minutes
• Monitor portal pressure throughout
Alternative Transplant Sites

- Spleen
- Omentum
- Forearm
- Kidney subcapsule
- Testis / Ovary / Eye
History of Islet Tx

1980s
• Routine reversal of diabetes in animal models

1990s
• Translation to human studies with 493 transplants in 40 institutions

2000 - 2008
• Edmonton Protocol enables good outcomes of clinical islet transplantation for at least 1 year post transplantation

2008 - present
• More consistent outcomes with improved graft longevity
ISLET TRANSPLANTATION IN SEVEN PATIENTS WITH TYPE 1 DIABETES MELLITUS USING A GLUCOCORTICOID-FREE IMMUNOSUPPRESSIVE REGIMEN

A.M. James Shapiro, M.B., B.S., Jonathan R.T. Lakey, Ph.D., Edmond A. Ryan, M.D., Gregory S. Korbutt, Ph.D., Ellen Toth, M.D., Garth L. Warnock, M.D., Norman M. Kneteman, M.D., and Ray V. Rajotte, Ph.D.
Geneva Results


Graft survival (C-peptide>0.5 ng/ml)

Insulin independence

p=0.04

p=0.001
Edmonton Results 2015

Insulin Independence

Time in Years

Edmonton Protocol
Alemtuzumab

58% 7-yr insulin independence Alemtuzumab

p<0.0001

11% EP

<table>
<thead>
<tr>
<th></th>
<th>3y II (%)</th>
<th>5yr II (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PTA</td>
<td>60%</td>
<td>52%</td>
</tr>
<tr>
<td>UMN-TCD</td>
<td>53%</td>
<td>50%</td>
</tr>
<tr>
<td>CITR-TCD</td>
<td>38%</td>
<td>41%</td>
</tr>
<tr>
<td>CITR-NTCD</td>
<td>33%</td>
<td>16%</td>
</tr>
</tbody>
</table>

No statistical difference
Selection Criteria

- Established Type 1 diabetes for more than 5 years
  - >18 years old
  - normal renal function or microalbuminuria
  - NOT if insulin dose more than 0.7U/kg

1) Recurrent severe hypoglycaemia >1 year
   - documented evidence of glucose <2mmol/L
   - despite compliance with intensified insulin regimen
   - normal renal function or microalbuminuria

2) Sub-optimal control despite functioning renal graft
   - severe hypoglycaemia; unstable diabetes
   - HbA1c >7%
Proposed treatment algorithm for patients with T1D and problematic hypoglycemia.

Pratik Choudhary et al. Dia Care 2015;38:1016-1029
Primary Outcome Measures for Islet Tx

- Resolution of life-threatening hypoglycaemic unawareness
- Stabilisation of glycaemic control (HbA1C)
- NOT primarily insulin independence
Outcomes in 24 UK patients in first 3 years of NHS funded programme (April 2008 to March 2011)

Brooks et al., Diabetic Medicine

**Severe hypoglycaemic event / patient / year**

- Pre-transplant: 23
- Post-transplant: 0.56

* p < 0.01 versus pre-transplant

**Glycated haemoglobin (HbA1c %)**

- Pre-transplant: 8.23
- Post-transplant: 6.83

* p < 0.01 versus pre-transplant
CITC Phase 3 Trial

Primary end-points were freedom from severe hypoglycaemic events Day 28-365 and an HbA1C of <7% at Day 365

Achieved in 87.5% at 1 year; 71% at 2 years
Patient RML after 12 months

HbA1c 7.0%

HbA1c 5.1%

100% Insulin Reduction
Patient CH after 11 months

HbA1c 7.7%

HbA1c 5.3%

Insulin Reduction 85%
HbA1c 9.6%

HbA1c 6.3%

Insulin Reduction 100%
TRIMECO Trial

Benefits of Graft Function (C-peptide +ve)

**TABLE 2.** Annual rate of change in GFR by $^{99m}$Tc-DTPA and MDRD in the medical and post-ICT groups

<table>
<thead>
<tr>
<th>$\Delta$ GFR (mL/min/1.73 m$^2$/yr)</th>
<th>Medical (95% CI)</th>
<th>ICT (95% CI)</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$^{99m}$Tc-DTPA all subjects</td>
<td>$-2.98 (-1.81$ to $-4.15)$</td>
<td>$-1.27 (-0.50$ to $-2.04)$</td>
<td>$&lt;0.0001$</td>
</tr>
<tr>
<td>$\geq 2$-yr follow-up</td>
<td>$-4.79 (-2.44$ to $-7.14)$</td>
<td>$-1.42 (-0.44$ to $-2.40)$</td>
<td>$&lt;0.0001$</td>
</tr>
<tr>
<td>$\geq 3$-yr follow-up</td>
<td>$-3.55 (-1.53$ to $-5.57)$</td>
<td>$-1.40 (-0.32$ to $-2.48)$</td>
<td>$&lt;0.0001$</td>
</tr>
<tr>
<td>MDRD all subjects</td>
<td>$-3.53 (-2.49$ to $-4.57)$</td>
<td>$-1.49 (-1.06$ to $-1.92)$</td>
<td>$&lt;0.0001$</td>
</tr>
</tbody>
</table>

GFR, glomerular filtration rate; ICT, islet cell transplantation; DTPA, $^{99m}$Tc-diethylene triamine pentaacetate; MDRD, modification of diet in renal disease; CI, confidence interval.

**TABLE 3.** Progression of diabetic retinopathy in the medical and post-ICT groups

<table>
<thead>
<tr>
<th></th>
<th>Medical</th>
<th></th>
<th>ICT</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. eyes</td>
<td>No. progressed</td>
<td>No. eyes</td>
</tr>
<tr>
<td>Mild NPDR</td>
<td>16</td>
<td>0</td>
<td>8</td>
</tr>
<tr>
<td>Moderate NPDR</td>
<td>19</td>
<td>1</td>
<td>12</td>
</tr>
<tr>
<td>Severe NPDR</td>
<td>6</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>PDR</td>
<td>41</td>
<td>7</td>
<td>29</td>
</tr>
<tr>
<td>Total</td>
<td>82</td>
<td>10*</td>
<td>51</td>
</tr>
</tbody>
</table>

* The progression is significantly more in the medical than the post-ICT group ($P<0.01$). ICT, islet cell transplantation; NPDR, nonproliferative diabetic retinopathy; PDR, proliferative diabetic retinopathy.

**Improved microangiopathy**

**Vancouver, ITA**

**Thompson et al. Transplantation 2011; 91: 373.**
Ongoing Challenges and Future Opportunities
Current Era
Next Era

- Retinopathy
- Renal failure
- Artherosclerosis

Graph shows the risk over years of diabetes.
1. **Optimise Islet Isolation**  
   (from pancreas procurement to targeted donor-specific pancreas digestion)

2. **Improve Islet Graft Survival**  
   (novel strategies of pre-transplant islet conditioning / islet modification)

3. **Availability of Non-Cadaveric Renewable Islet Source**  
   (xenogeneic or islet stem cells)

4. **Development of immunosuppressive-free immune strategies**  
   (tolerance or immuno-isolation)
Moving Islet Tx to Children

1. **Optimise Islet Isolation**
   (from pancreas procurement to targeted donor-specific pancreas digestion)

2. **Improve Islet Graft Survival**
   (novel strategies of pre-transplant islet conditioning / islet modification)

3. **Availability of Non-Cadaveric Renewable Islet Source**
   (xenogeneic or islet stem cells)

4. **Development of immunosuppressive-free immune strategies**
   (tolerance or immuno-isolation)
Moving Islet Tx to Children

1. **Optimise Islet Isolation**
   (from pancreas procurement to targeted donor-specific pancreas digestion)

2. **Improve Islet Graft Survival**
   (novel strategies of pre-transplant islet conditioning / islet modification)

3. **Availability of Non-Cadaveric Renewable Islet Source**
   (xenogeneic or islet stem cells)

4. **Development of immunosuppressive-free immune strategies**
   (tolerance or immuno-isolation)
Alternative Islet Sources

CRISPR/Cas9 gene editing leads to a strong revival of xenotransplantation

Endocrine progenitor

CRISPR/Cas9 gene editing leads to a strong revival of xenotransplantation

LUNG
- A factory farm is being designed to produce 1,000 pig lungs per year.

KIDNEY
- A kidney with six genetic modifications supported a baboon’s life for 4 months.

CORNEA
- Pig corneas were approved for marketing in China in April.

HEART
- A genetically modified pig heart implanted in a baboon’s abdomen survived for 2.5 years.

LIVER
- Livers could be engineered to produce their own antibodies against primate immune cells.

Reardon, Nature 2015
Yang et al Science 2015
1. **Optimise Islet Isolation**  
(from pancreas procurement to targeted donor-specific pancreas digestion)

2. **Improve Islet Graft Survival**  
(novel strategies of pre-transplant islet conditioning / islet modification)

3. **Availability of Non-Cadaveric Renewable Islet Source**  
(xenogeneic or islet stem cells)

4. **Development of immunosuppressive-free immune strategies**  
(tolerance or immuno-isolation)
Immune Strategies

• Imunoalteration (Immune Tolerance)

• Immunoisolation (Micro- and Macro-Encapsulation)
Immune Tolerance

Host T cell repertoire before tolerance to donor

Vβα
Vβb
Vβc
Vβx...

T cells recognizing donor Ags

Transplant tolerance induction

Host T cell repertoire after tolerance to donor

Immunity against pathogens mediated by these T cells will be lost

X X X

Host T cells recognizing donor antigens and superantigens will be clonally deleted and inactivated
Immune Strategies

• Imunoalteration (Immune Tolerance)

• Immunoisolation (Micro- and Macro-Encapsulation)
Micro-encapsulation
Macro-encapsulation
<table>
<thead>
<tr>
<th>1st intention</th>
<th>SERNOVA CORP</th>
<th>Beta-O2</th>
<th>VIACYTE</th>
<th>DEFYMED</th>
</tr>
</thead>
<tbody>
<tr>
<td>Implantation site</td>
<td>Sub-cutaneous</td>
<td>Extra-peritoneal</td>
<td>Sub-cutaneous</td>
<td>Extra-peritoneal</td>
</tr>
<tr>
<td>Filling/Empting of cells</td>
<td>Yes</td>
<td>No</td>
<td>Yes (Unique Entry/Exit)</td>
<td>Yes (Separate Entry/Exit)</td>
</tr>
<tr>
<td>Cell type for clinical trials</td>
<td>Human islets</td>
<td>Human islets</td>
<td>Stem cells</td>
<td>Human islets</td>
</tr>
<tr>
<td>Cell number</td>
<td>Sufficient</td>
<td>Sufficient</td>
<td>Non-sufficient</td>
<td>Sufficient</td>
</tr>
<tr>
<td>Stage of development</td>
<td>Phase I/II-a clinical</td>
<td>Phase I/II-a clinical</td>
<td>Phase I/II-a clinical</td>
<td>Preclinical</td>
</tr>
<tr>
<td>How does it look like?</td>
<td><img src="image1.png" alt="Image of interstitial med device" /></td>
<td><img src="image2.png" alt="Image of interstitial med device" /></td>
<td><img src="image3.png" alt="Image of interstitial med device" /></td>
<td><img src="image4.png" alt="Image of interstitial med device" /></td>
</tr>
</tbody>
</table>
MAILPAN Device

Islet Encapsulation:
- immunoprotection
- biocompatibility
- stability
- selective permeability
- capacity
Mimics the pancreas extracellular matrix (ECM)

- Composed of native ECM molecules pancreatic proteins (to support and support islets)
- Oxygen producing particles
- Provides structure and spacing to islets

β-Gel will protect the islets and support their growth outside the liver.

![Images of β-Gel and islets]

Protein 1

Control

Protein 2
β-Shell

A smart biocompatible implant for delivery of β-Gel to an extra-vascular site

- Drug eluting shell for enhanced bio-integration
- Engineered to selectively isolate immune system cells while allowing insulin and glucose transport.

via minimally invasive procedure
Biofunctionality (1)

Permeability (%)

-20 0 20 40 60 80

OXY_M2 non traitée
OXY_M2 traitée

% perméabilité de la membrane à l’IgG

temps de diffusion (h)

0 6 12 18 24

Insulin in the lower compartment (µg/g of protein)

0 4 8 12 16 20 24

Perméabilité IgG

Insulin
IgG
Biofunctionality (2)

C-peptide/glycemia (pmol/L) vs. Time after filling (days)

- 23 Diabetic w/ MAILPAN+2500 islets
- 25 Diabetic w/ MAILPAN+medium
- 22 Diabetic w/o MAILPAN
Islet Hypoxia

Dysfunction (Murphy, 2009)

Hypoxia

ROS high
“The future ain’t what it used to be!”

Yogi Bera 1998
Conclusions

- Islet transplantation works well in selected patients

- Number of ongoing challenges before it can be applied more widely

- Development of immune tolerance or encapsulation protocols required before implementing treatment in children

- Cell isolation / cell transplant techniques applicable for potentially treating many other surgical conditions in children e.g. liver, myocytes, engineering etc