BMT for RDEB and JEB
What have we learned, and what do we do next?

A Healthy skin

- Stratum corneum
- Stratum granulosum
- Stratum spinosum
- Stratum basale

Epidermis

- Basement membrane
- Dermis

B Skin in patient with recessive dystrophic epidermolysis bullosa

- Erosion of skin layers
- Blistering between the epidermis and dermis

Epidermal cell

- Hemidesmosome
- Lamina densa
- $\alpha_6\beta_4$ Integrin
- Laminin-332
- Anchoring plaque
- Anchoring fibrils (type VII collagen)

Dermis

- Damaged or absent anchoring fibrils

NEJM 2015

Jakub Tolar MD PhD
University of Minnesota
How does BMT work in EB?

Blood-producing stem cells taken from donor

Donated cells given to patient via bloodstream

Donated cells circulate through the recipient’s body and produce missing collagen

Donated cells engraft in bone marrow and produce healthy cells
Do bone marrow cells engraft in skin?

Kataoka *Am J Pathol* 2003

Körbling *NEJM* 2002

Fujita *PNAS* 2010
Does BMT work in EB?
Should we do BMT for EB?

Systemic impacts of EB

- Internal surfaces
- Pain
- Itch
- Fibrosis
- Anemia
- Malnutrition
- Autoimmunity
- Dentition
- Kidney
- Heart
Is BMT reasonably safe?

- UMN Institutional Review Board
- External review boards

Protocol and patient safety

Colleen Delaney, MD
Gay Crooks, MD

Patient review

John McGrath, MD
Alain Hovnanian, MD
Hiroshi Shimizu, MD, PhD
Katsuto Tamai, MD, PhD
How do we do BMT for EB?

Adaptive Clinical Trial – 2007 to present
Safety & effectiveness

• FDA Investigational New Drug IND 14166
• Clinical Trials (clinicaltrials.gov):
  • BMT for Severe EB (high dose): NCT00478244 - closed
  • MSCs for Severe EB: NCT02581775
  • Skin grafting – using BMT donor skin: NCT02670837
  • BMT for Severe EB (low dose/haplo donor): NCT0103355
How do we know if it works?

Multiple measurements made at set timepoints:

Engraftment: presence of donor cells in patient’s skin

Patient: XX Female
Donor: XY Male

Donor CD45-/CD31- cells

23% donor in skin
How do we know if it works?

Visual and antibody identification of anchoring fibrils (electron microscopy)
How do we know if it works?

Persistence: anchoring fibrils: 7 years after BMT

mAb185

LH24
How do we know if it works?

Skin strength (blistering machine)
Correlation of skin strength with skin engraftment

![Graph showing correlation between days after transplant and minutes to blister](image)
How do we know if it works?

Patient and parent report (iScorEB)

“How do we know if it works?"

“Today he ate Chex, ice cream, melon, yogurt, popcorn, nachos and cheese, soft shell taco, a chocolate chip cookie, chips and salsa, and cereal with milk.”

“He is standing, taking steps with support, and becoming active again.”
**How do we know if it works?**

*Parent report of estimated impact on time and money*

<table>
<thead>
<tr>
<th>Conditioning</th>
<th>Transfusions</th>
<th>Hospital days</th>
</tr>
</thead>
<tbody>
<tr>
<td>High dose</td>
<td>22.3</td>
<td>74.5</td>
</tr>
<tr>
<td>Low dose</td>
<td>6.3</td>
<td>38</td>
</tr>
</tbody>
</table>

- **P=0.003**
- **P=0.01**

- 5-10 fold reduction in bandages
- Reduced time bandaging
- Fewer antibiotics and surgical interventions
How do we know if it works?

<table>
<thead>
<tr>
<th>Before HCT</th>
<th>After: Short-term</th>
<th>After: Long-term</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="before_hct.png" alt="Image" /></td>
<td><img src="after_short_term.png" alt="Image" /></td>
<td><img src="after_long_term.png" alt="Image" /></td>
</tr>
</tbody>
</table>

![Before HCT, After Short-term, After Long-term images with arrows highlighting differences.](image.png)
How do we know if it works?
How do we know if it works?
Why does the trial change?

Overall 2-year survival = 75%

<table>
<thead>
<tr>
<th></th>
<th>Total Patients</th>
<th>Deaths</th>
<th>Transplant-related death</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Trial 1</strong></td>
<td>6</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>High dose &amp; MSCs</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Trial 2</strong></td>
<td></td>
<td>7</td>
<td>3</td>
</tr>
<tr>
<td>High dose &amp; MSCs</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Trial 3</strong></td>
<td>16</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>Low dose &amp; MSCs</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Should we do BMT for RDEB?

Clinical Improvement

<table>
<thead>
<tr>
<th>Trial 1</th>
<th>Trial 2</th>
<th>Trial 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>High dose</td>
<td>High dose &amp; MSCs</td>
<td>Low dose &amp; MSCs</td>
</tr>
<tr>
<td>Total Patients</td>
<td>Major Improvement</td>
<td>Minimal Improvement</td>
</tr>
<tr>
<td>6</td>
<td>7</td>
<td>16</td>
</tr>
<tr>
<td>1</td>
<td>3</td>
<td>8</td>
</tr>
<tr>
<td>2</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>3</td>
<td>4</td>
<td>3</td>
</tr>
</tbody>
</table>

Trial 1: High dose (1 patient with major improvement, 3 with moderate improvement, 2 with minimal improvement)

Trial 2: High dose & MSCs (3 patients with major improvement, 4 with moderate improvement)

Trial 3: Low dose & MSCs (8 patients with major improvement, 5 with moderate improvement, 3 with minimal improvement)
Should we do BMT for LAMA3 JEB?

1-year-old with LAMA3 JEB – high dose conditioning
Before transplant

1 year after transplant

23% donor in skin
BSA 90% before
< 5% after
Should we do BMT for LAMA3 JEB?

10-year-old with LAMA3 JEB – reduced intensity conditioning
Before transplant
2 years after transplant

50% donor in skin
BSA 75% before
< 50% after
Is HCT a cure for EB?

- No

- However, thus far...only clinical intervention shown to:
  - Correct the **systemic** disease pathology of RDEB
  - Have potential to improve **overall** quality of life
New BMT protocols for severe EB

1. Hematopoietic cells
   - Haploidentical grafts

2. Mesenchymal stromal cells
   - Three serial infusions

3. Epidermal cells
   - Skin grafting from HCT donor
   - Skin grafting from mosaic skin
New Hands New Life

By Zuzia
Biomedical Moment

**COL7A1**
- Mutations (founders)
- Genomic polymorphisms (modifiers)

**Burden of disease**
- Infections
- Malnutrition
- Chronic wounds

**Economic burden**
- Wound care
- Supportive care
- Transplant

**Ethical**
- External Advisory Panel
- Eligibility

**Societal**
- Personality of child
- Emotional support

**FAMILY**

**CLINICAL DECISION**
Damage vs Resilience

- Early
- Late

- Not the same disease...
- Not the same intervention

- Childhood
- Adulthood

- Birth
- Early
- Late
Cycle of learning

CLINICAL DECISION

Transplant

New Information

Outcome Measures

Learn from Every Patient
Combination therapy for RDEB

C  Combination therapy

- **Allogeneic cells**
- **Mosaic cells**
  - Autologous cells with natural reversion of COL7A1 mutation
- **Gene-corrected cells**
  - Autologous cells after gene editing or gene addition of COL7A1

**Cellular therapy**
- Skin
  - Keratinocytes
  - Fibroblasts
  - Induced pluripotent stem cells (iPSCs)
  - Mesenchymal (stem) cells
  - Bone marrow
  - Cord blood
  - Hematopoietic (stem) cells

**Type VII collagen protein therapy**

**Local therapy**
- Intradermal injection (e.g., with microneedles)
- Skin grafts

**Systemic therapy**
- Intravenous injection
- Intraarterial injection

**Skin-healing effects**

*NEJM 2015*
Cycle of learning

CLINICAL DECISION

Transplant

New Information

Learn from Every Patient

Outcome Measures
Follow-up and assessment over time

1. Biomarkers: C7, skin engraftment, anchoring fibrils
2. Skin strength
3. Wound healing, SCC
4. Time in hospital
5. Quality of life (iScorEB)
Cycle of learning

CLINICAL DECISION

Transplant

New Information

Outcome Measures

Learn from Every Patient
Learn from Every Patient

Good outcomes

Poor outcomes

New Information
Cycle of learning

CLINICAL DECISION

New Information

Learn from Every Patient

Outcome Measures

Transplant
What more should we do for EB?

Cell projects

2009
BMT in RDEB mice

2010
BMT in people with RDEB (N=7)

2011
iPSCs from human RDEB cells

2013
iPSCs from human JEB cells

2014
iPSCs from human mosaic RDEB cells

2017
BMT in people with RDEB (N=30) (in review)

Gene projects

2013
COL7A1 gene editing with TALENs

2016
C7 is regulated by mir-29

2016
COL7A1 gene editing with CRISPR/Cas9

2017
Col7a1 null mice for transfer of human cells
Transplant helps ease the symptoms of children with RDEB.

Patients given stromal cells from donor after hematopoietic cell transplant.

Patients have persistent wounds, even after transplant: use skin grafting.

Correct COL7A1 by CRISPR/Cas9 editing in iPSC and iPSC-HSC/MSC/KC.

NEXT?

First in EB patients

First in human
Collaborators and friends

John A. McGrath
Katsuto Tamai
Alain Hovnanian
Mark Osborn
Kristen Hook
Leena Bruckner-Tuderman
Jemma Mellerio
Anne Lucky
Elena Pope
Amy Paller
Michelle de Luca
Francis Palisson
Gabi Pohla-Gubo
Denis Roop
Peter Marinkovich
Jouni Uitto

tolar003@umn.edu