Engineering beta cells for resistance to immune attack and reversal of diabetes

Blavatnick Proposal
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Kevan C. Herold, MD
CNH Long Professor of Immunobiology and Internal Medicine
Yale University

Our team

Kevan Herold, MD

- Professor of Immunology and Endocrinology
- Chair of NIDDK/TrialNet
- Research interest: the development and progression of autoimmunity and the responses of target tissues to immune assault
- Responsible for the development of teplizumab, the first drug approved by FDA for delay/prevention of Type 1 diabetes recently

FDA NEWS RELEASE

FDA Approves First Drug That Can Delay Onset of Type 1 Diabetes

Today, the U.S. Food and Drug Administration approved Tzield (teplizumab-mzwv) injection to delay the onset of stage 3 type 1 diabetes in adults and pediatric patients 8 years and older who currently have stage 2 type 1 diabetes.

Matthias Hebrok, PhD

- UCSF/Hemholz Diabetes Center
- Help create embryonic stem cell-derived beta cells (ESCBC)
- eliminated TET2 from human cells by CRISPR/Cas9.)

Minutia (Startup@Berkley, CA)

- Help differentiate beta cells derived from human stem cells and gene knockout
- Expansion and differentiation of iPSC derived beta cells
- Tools for analyzing cellular stress responses

Management of diabetes (all forms) do not meet standards of care

- All forms of diabetes are caused by *insufficient insulin production* to meet metabolic demands
  - ***Type 1 diabetes:*** immune-mediated killing of insulin producing beta cells and patients require insulin for survival.
  - ***Type 2 diabetes:*** inadequate beta cell mass and also involves insensitivity to insulin.

- Unmet need:
  - Patients with Type 1 diabetes rarely meet prescribed goals
  - At risk for the endorgan complications of the disease (eye, kidney, nerve, vascular, etc).

Islet transplants (from organ donors)

- insufficient islets for transplantation.
- requires indefinite broad immune suppression
  – a non-starter for children with Type 1 diabetes

Immune therapy with beta cell replacement:

A cure for Type 1 diabetes

Teplizumab:
approved Nov 2022

↓

Replacement with human ES cell derived beta cells

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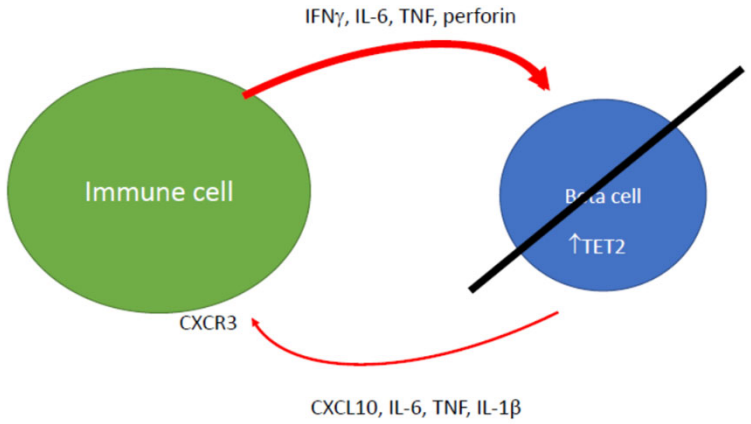
Engineered beta cells that resist immune killing

A key pathway of inflammation mediated beta cell killing that sets up a dynamic between the beta cells and immune cells and promotes their killing

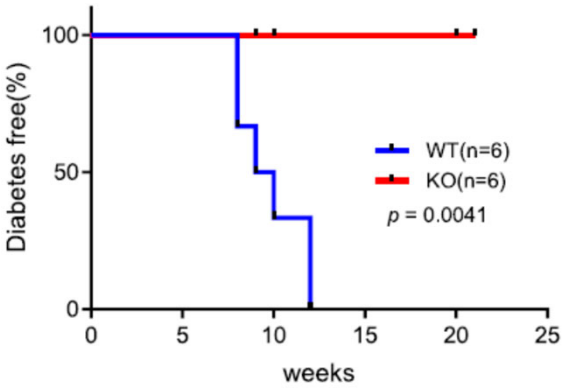
- Hypothesis
  - Eliminating of TET2 from human induced pluripotent stem cell-derived beta cells would cloak those cells from immune mediated killing
- Outcome
  - Establish proof of concept for developing personalized beta cell replacement from induced pluripotential stem cells (iPSCs).
  - Novelty: targeting a key regulator of inflammatory responses on the tissue rather than suppressing the immune system of the patient.



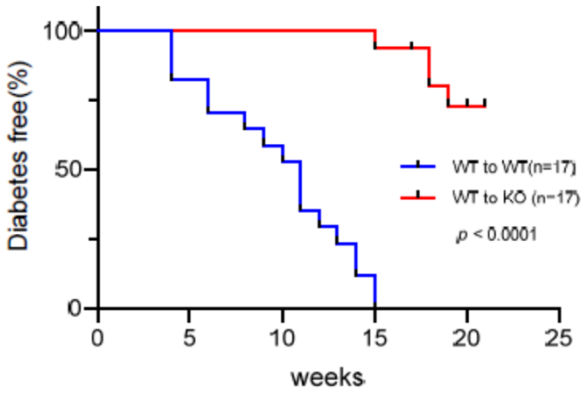
The enzyme TET2 is the regulator

Elimination of *Tet2* in islet cells can protect them from killing by immune cells in 3 experimental models

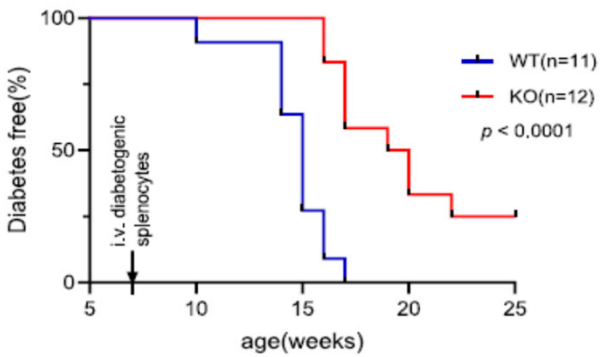
Adoptive transfer of diabetogenic spleen cells



Bone marrow transplant



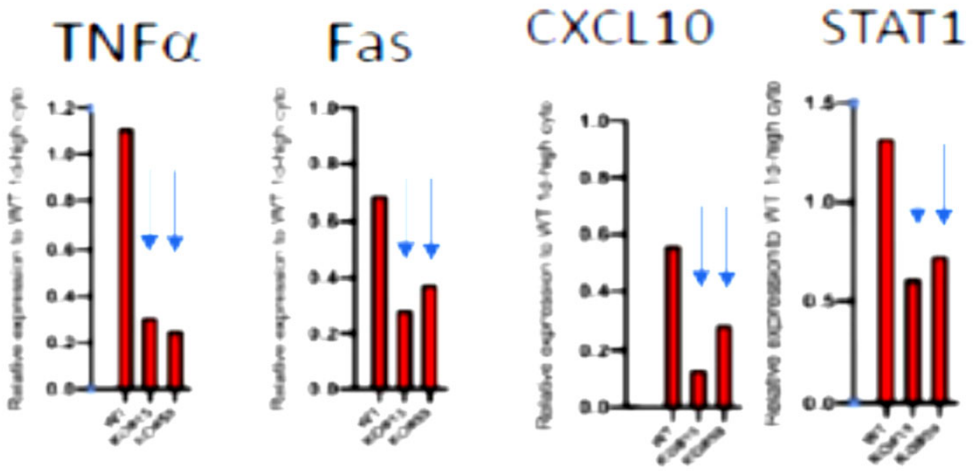
Islet transplant



Unlike directly blocking or depleting immune cells, the standard approach for immune suppression, eliminating Tet2 “cloaks” immune cells so that the immune cells and their products do not kill beta cells and beta cells do not provoke immune cells.

Credit to J Rui, J Lu, and K Herold at Yale

Similar effects of TET2 deletion occur with human embryonic stem cell derived beta cells.



Human embryonic stem cell derived beta cells were produced (by Dr. M Hebrok's lab) and TET2 was depleted by CRISPR/Cas9. These beta cells were cultured with a combination of inflammatory cytokines (TNFα, IL-1β, and IFNγ. The relative expression of the indicated mediators was determined by RT-PCR. The blue arrows indicate results with two TET2-deficient cell lines vs wild type (first bar). These studies show reduced gene expression of TNFα, Fas, CXCL10, and STAT1 in the TET2 deficient beta cells.

Proposal for pilot support (Costs: Appx $300,000)

1

Develop iPSC cell lines

- **Use an existing induced pluripotential stem cell (iPSC)**
- **Develop 3-4 iPSC cell lines from patients.**

can start immediately with existing line

Lab costs for differentiation of the line appx $5,000

2

The iPSC core lab at Yale will produce iPSC lines from 3 patients with Type 1 diabetes and differentiate into beta cells.

$13,500 + $20,000

3

Delete TET2 by CRISPR/Cas9 from the cell lines

- **test the metabolic function of the cells**
- **determine if the cells are resistant to immune killing**
- **test whether the human cells can reverse diabetes and evade killing in a humanized mouse model (i.e. mice reconstituted with human immune cells).**

TET2 deletion accomplished in one human ESC line

Lab cost including personnel, appx $85,000