# Pancreatic Islet-like Cells Derived from Fibroblasts using Non-viral Direct Cell Reprogramming


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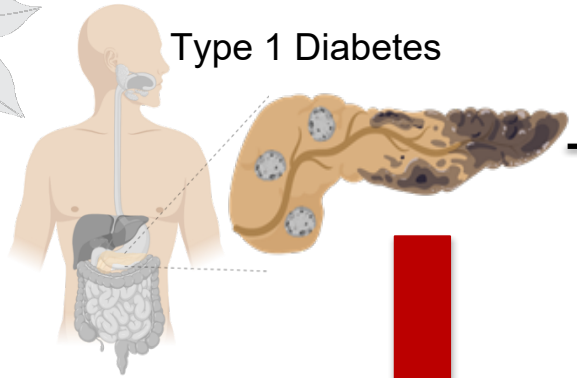
Disclosures: None

THE OHIO STATE UNIVERSITY

# What is type 1 diabetes?



Type 1 Diabetes

Low insulin production

Cell cannot uptake glucose

↑ blood glucose levels

Allogenic beta cell transplantation

normal insulin production

Cell can uptake glucose

normal blood glucose levels

Limited due to a lack of cell source

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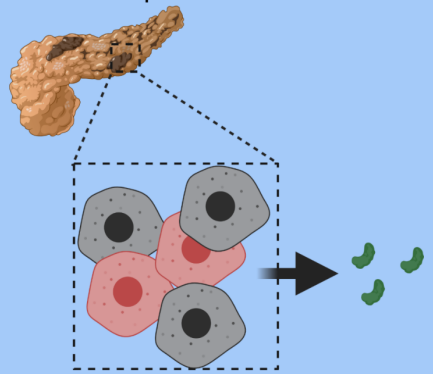
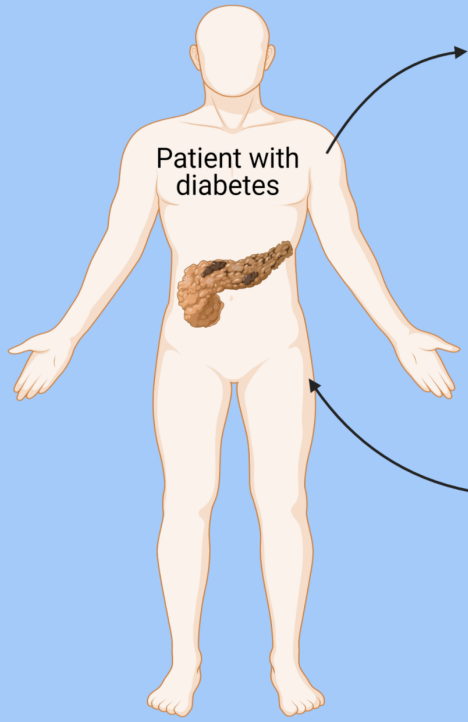
# Alternative method to treat type I diabetes

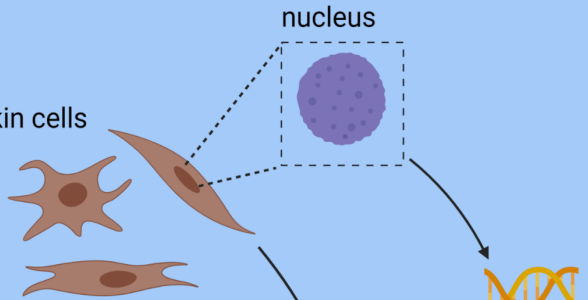
Insulin protein

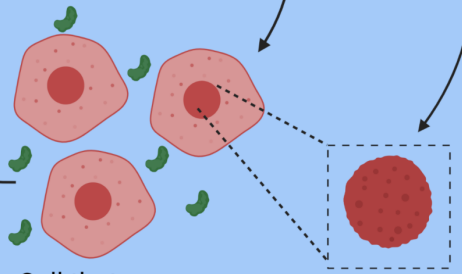
Current standard of care

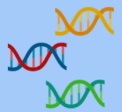
Root of the problem



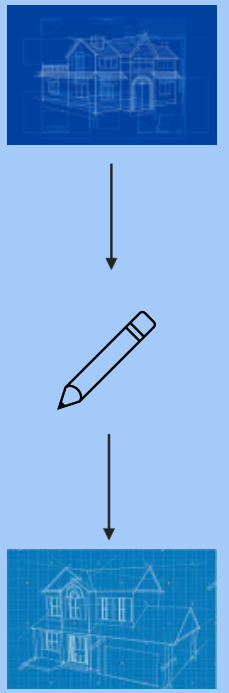


What is the optimal DNA "recipe"?



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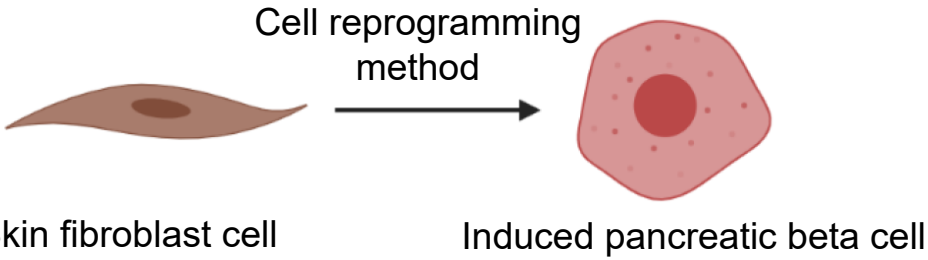
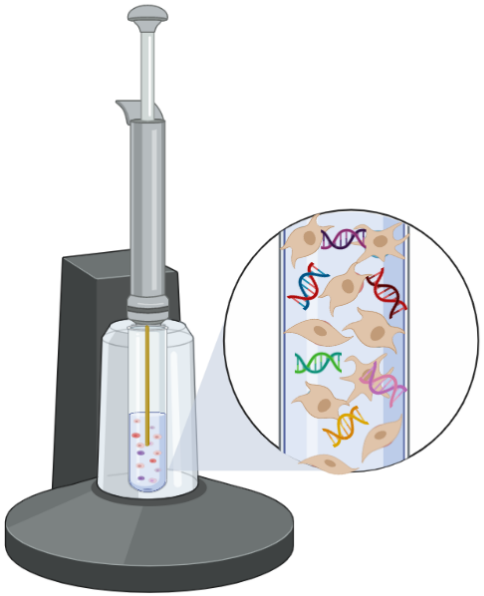
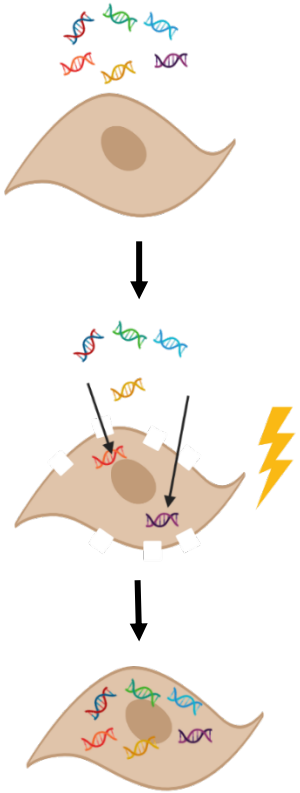
The purpose of this study is to develop a cell reprogramming method to induce pancreatic beta cells from fibroblast




Illustrations created with BioRender.com

# Bulk electroporation (BEP) is used to deliver instructional DNA to fibroblasts to drive beta cell reprogramming





Cells and DNA in suspension

Electric field applied across suspension

DNA delivered into cell

## Cell source:

Ins1-EGFP Mouse Embryonic Fibroblasts (MEFs)

Express insulin 1

GFP expression

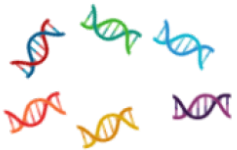
## Instructional DNA used:

(7) Pancreatic Beta Cell Development

- *Pdx1, Ngn3, Mafa, NeuroD1, Pax4, Nkx6.1* and *FoxA2*

(3) Skin Plasticity

- *Tcf3, Trp63,* and *Sox9*



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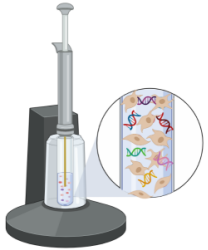
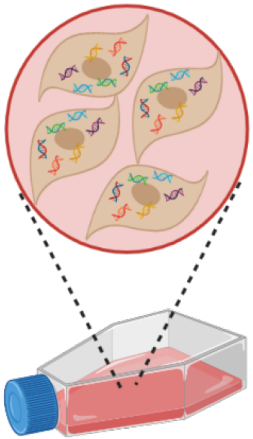
# Examined GFP and insulin expression 1 and 4 days after BEP

BEP

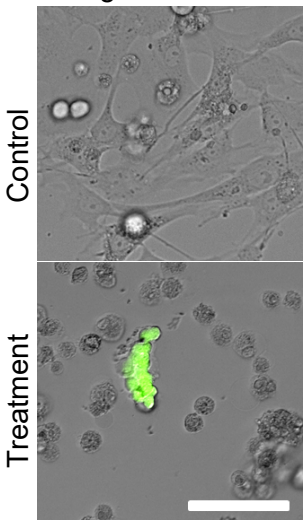


Cultured cells for 1 and 4 days after BEP



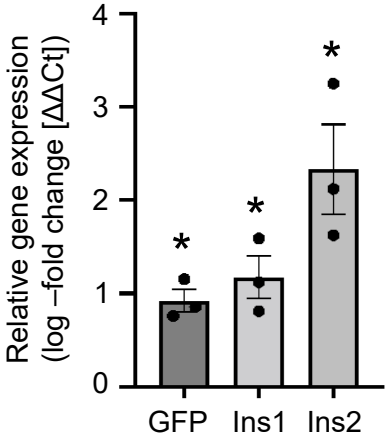
**Fluorescence imaging: 1 day after BEP**

Brightfield/GFP




Scale bar = 100 µm
Objective: 20X

**Gene expression: 4 days after BEP**



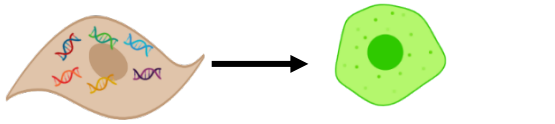

* P < 0.05 compared to control

Delivery of these 10 DNA genes can increase insulin gene expression in fibroblasts as early as 4 days after BEP

GFP signal represents Ins1 promotor activation

# Implications of results include

- BEP with all 10 DNA encoding genes led to ↑ GFP, Ins1 and Ins2 gene expression in MEFs by day 4



MEF loaded with 10 DNA encoding genes

↑ GFP, Ins1 and Ins2 gene expression

# Future Directions

- Determine of each DNA encoding gene individually
- Culture BEPed MEFs to longer timepoints (e.g., 7, 14 and 21 days)
- Examine protein expression profile  (e.g., ICC, ELISA)
- Functional assays (e.g., glucose stimulated insulin response assay)

Unpublished data

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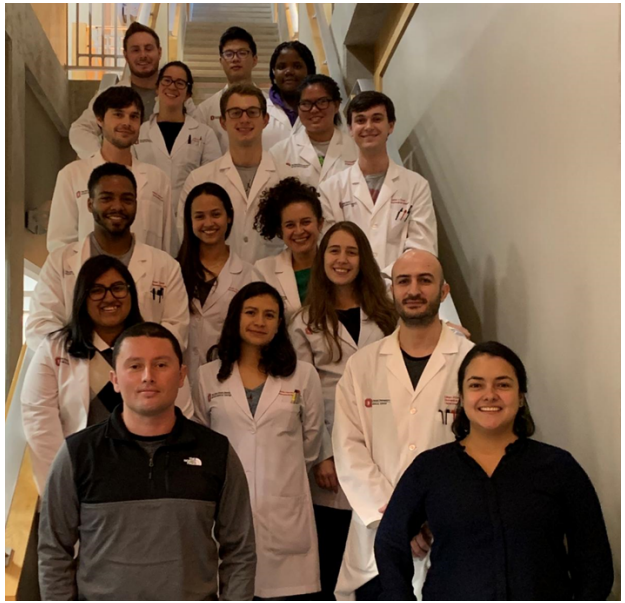

# Questions?