Pancreatic Gene Therapy Durably Improves Glycemia and Delays Disease Progression in a Murine Model of Type 2 Diabetes

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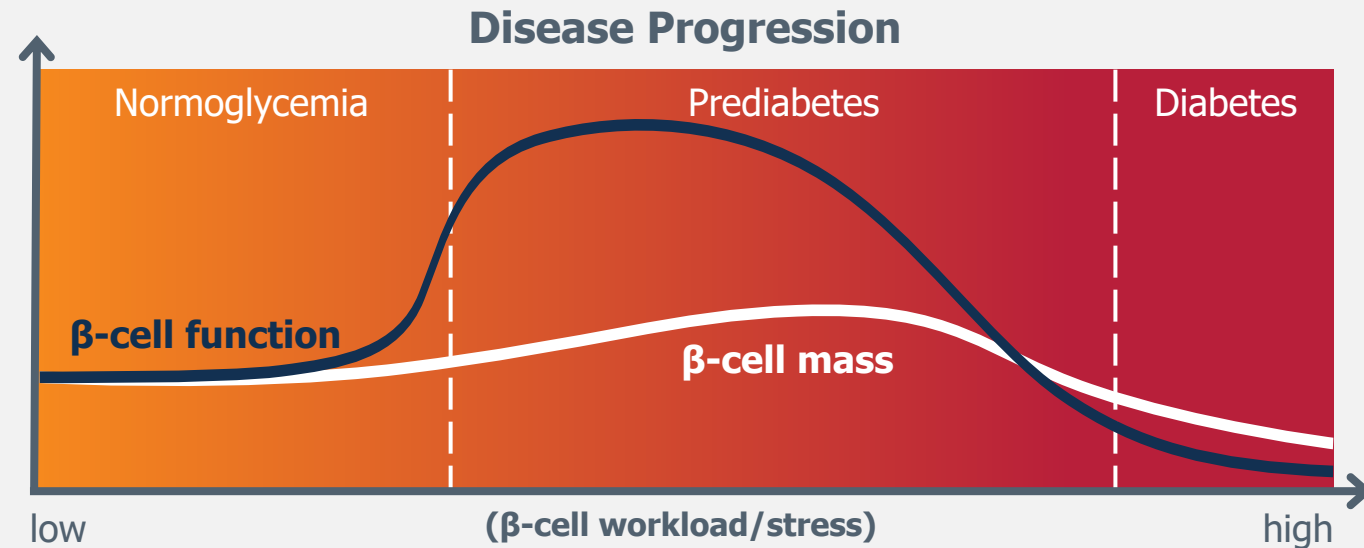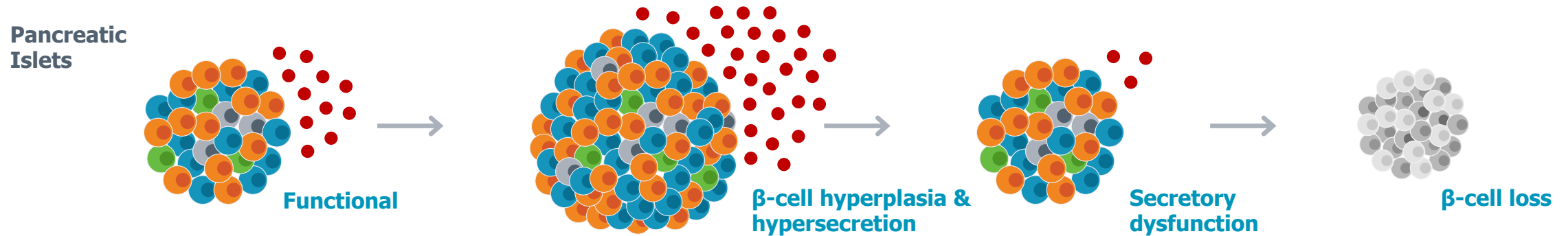
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Disclosure Statement

Authors

Harith Rajagopalan, Alice Liou, Emily Cozzi, Jacob Wainer, Rebecca Reese, and Jay Caplan are employees and shareholders of Fractyl Health, Inc. Nidhi Khanna and Jason A. West are former employees of Fractyl Health, Inc.

The Pancreatic Gene Therapy (PGTx) is in early development and not approved by any regulatory body for investigational or commercial use.

T2D Progression is Driven by Declining Islet Health
Loss of β-cell function is the sine qua non of T2D

Pancreatic Islets

Functional → β-cell hyperplasia & hypersecretion → Secretory dysfunction → β-cell loss

Disease Progression

Normoglycemia | Prediabetes | Diabetes

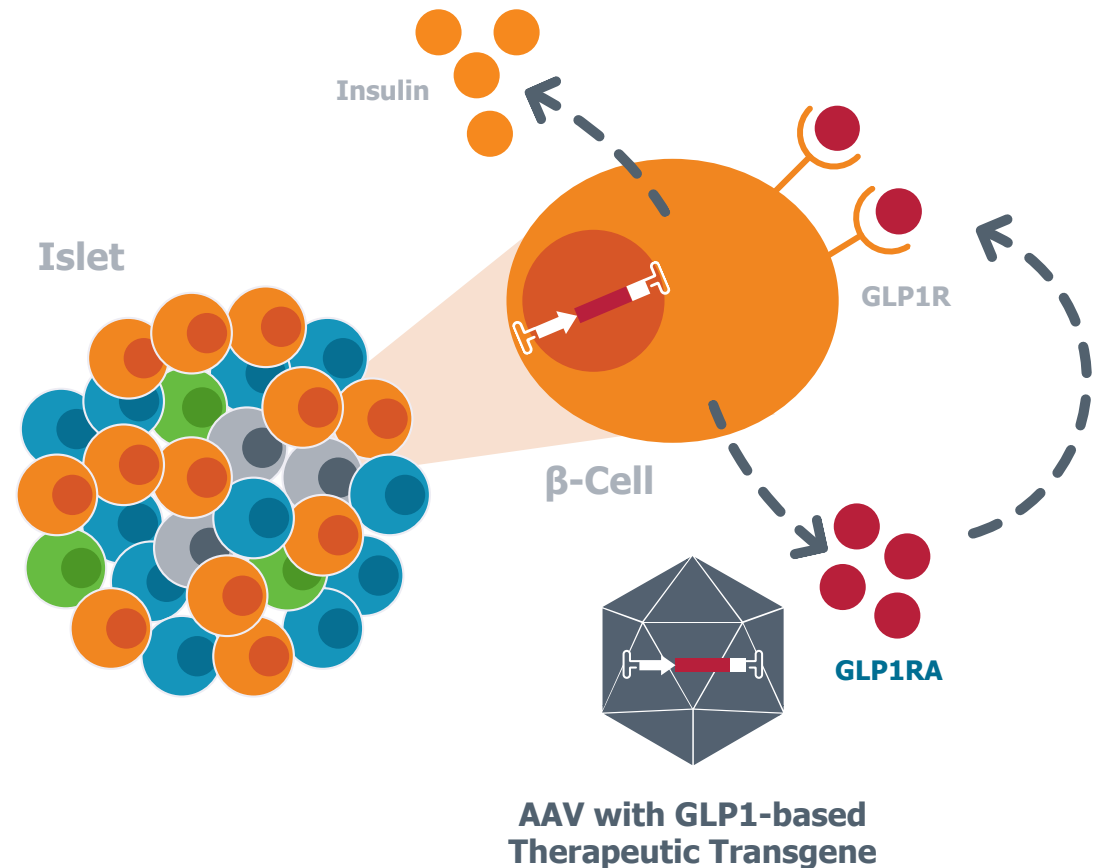β-cell function

β-cell mass

low → (β-cell workload/stress) → high

Pancreatic Gene Therapy (PGTx) to Improve Islet Function

Potential for durable improvement in β-cell function

Islet cells terminally differentiated, making adeno-associated virus (AAV) a suitable means of durable genetic modification[1,2]

Intra-islet GLP1 signaling is essential for β-cell function, health, and survival[3,4]

GLP1-based pancreatic gene therapy (GLP1 PGTx driven by the insulin promoter) may restore islet health in T2D via durable local production of GLP1RA
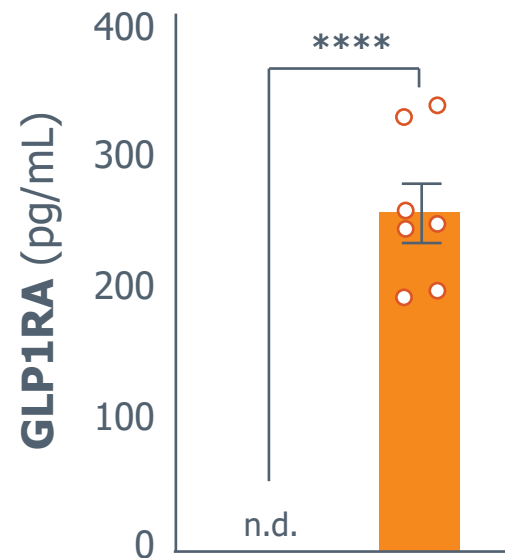

Insulin

Islet

β-Cell

GLP1R

GLP1RA

AAV with GLP1-based Therapeutic Transgene

Figure adapted from Saikia et al. JCI Insight. 2021 6:e1418511. 1. Ju et al. Diabetologia. 1998 41:736-739. 2. Kapturczak et al. Mol Ther. 2002 5:154-160. 3. Campbell and Drucker. Cell Metab. 2013 17:819-837. 4. Fava et al. J Diabetes Complications. 2016 30:1651–1658. AAV=adeno-associated virus, GLP1=glucagon-like peptide 1, GLP1R=GLP1 receptor, GLP1RA=GLP1R agonist, PGTx=pancreatic gene therapy

GLP1 PGTx Improves Insulin Production and GSIS in *db/db* Islets

Metabolic improvements in isolated islets 10 weeks after PGTx

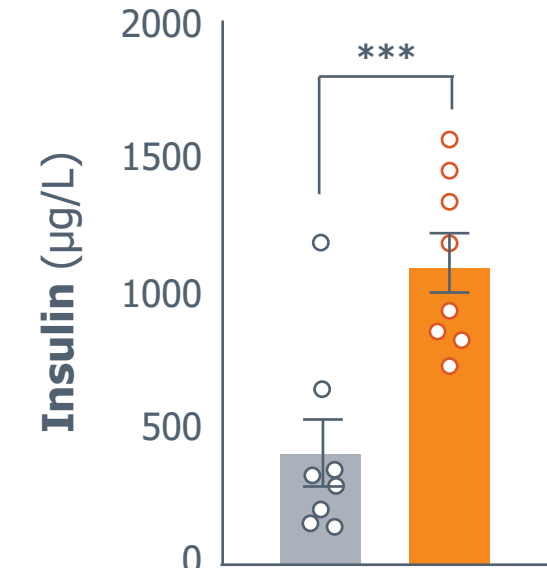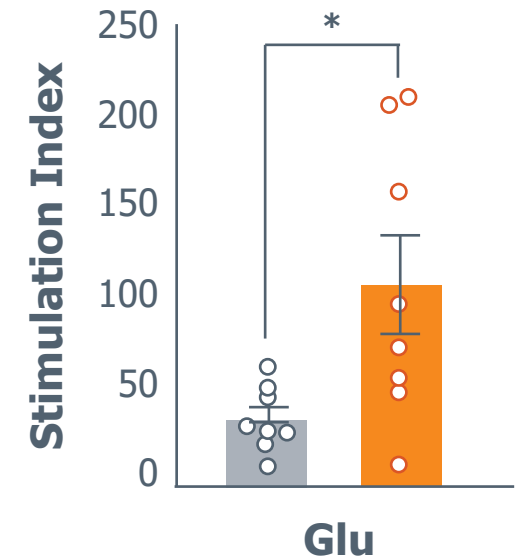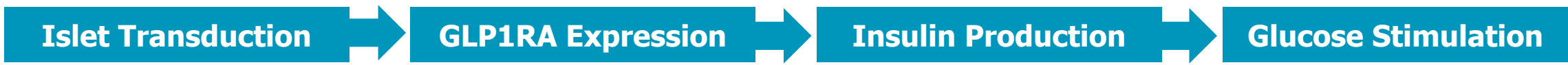**A)** Islet Transduction
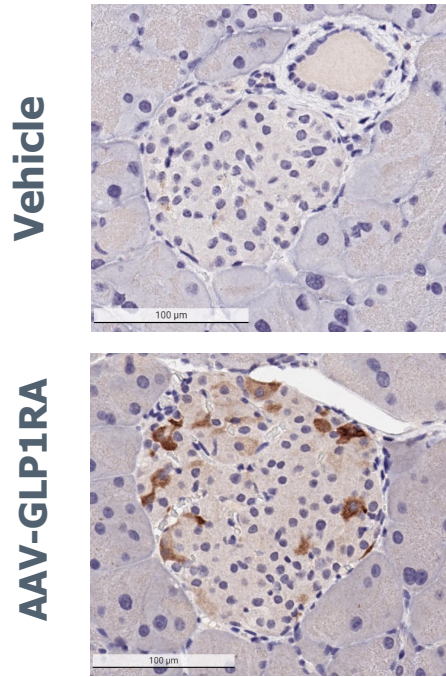
B) GLP1RA Protein Content

C) Insulin Content

D) Glucose-Stimulated Insulin Secretion (GSIS)

Vehicle   AAV-GLP1RA

Islet Transduction → GLP1RA Expression → Insulin Production → Glucose Stimulation

Mean ± SD shown; *p<0.05, ***p<0.001, ****p<0.0001; n=8 per group. D) Glucose stimulation of 16.7 mM +/- IBMX from 2.8 mM baseline. Rajagopalan et al. ASGCT 2023 oral presentation. Abstract no. 191. AAV=adeno-associated virus, GLP1=glucagon-like peptide 1, GLP1RA=GLP1 receptor agonist, Glu=glucose, GSIS=glucose-stimulated insulin secretion, n.d.=not detectable, PGTx=pancreatic gene therapy
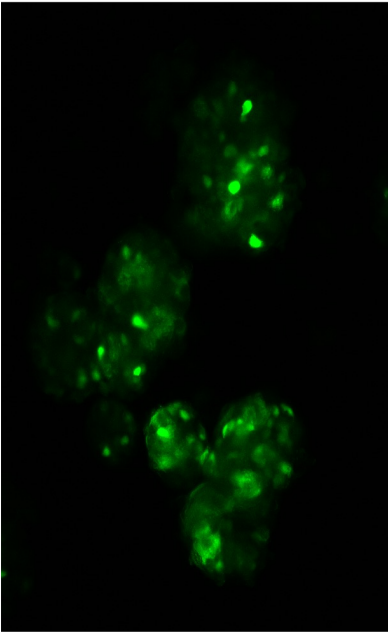
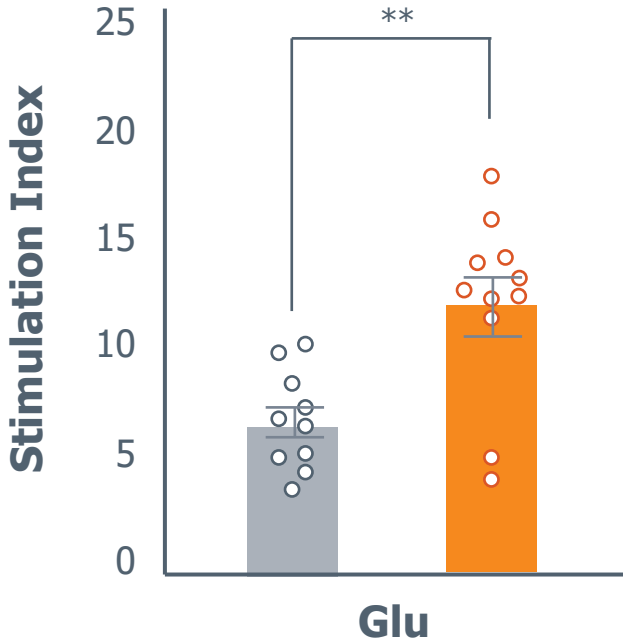# GLP1 PGTx Improves GSIS in Human Islets and Human β-cell Line
Improved GSIS mediated by GLP1R activation in human cells
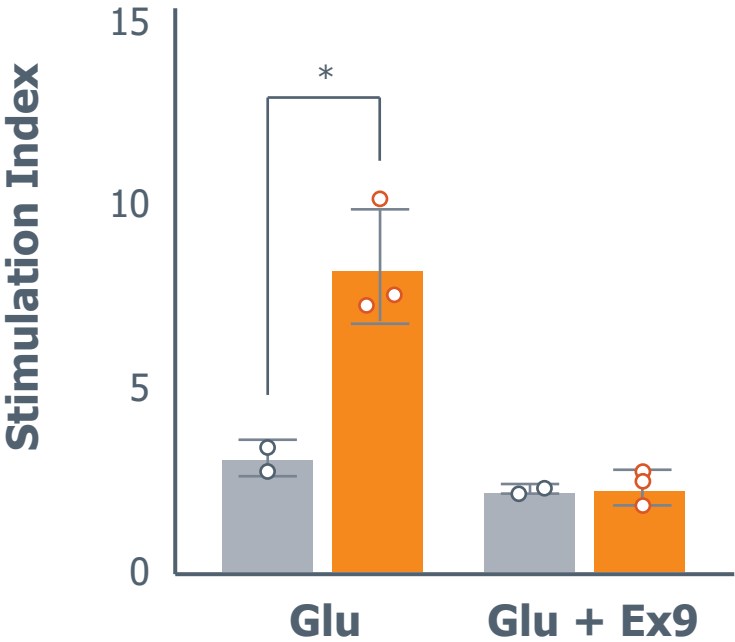
A) Human Islet Transduction

GFP Expression

B) Human Islet GSIS

Glu

C) Human β-cell Line GSIS ± Ex9 (GLP1R Antagonist)

Glu   Glu + Ex9

Untransduced   AAV-GLP1RA

Local Delivery of PGTx
Proprietary endoscopic ultrasound-guided infusion device
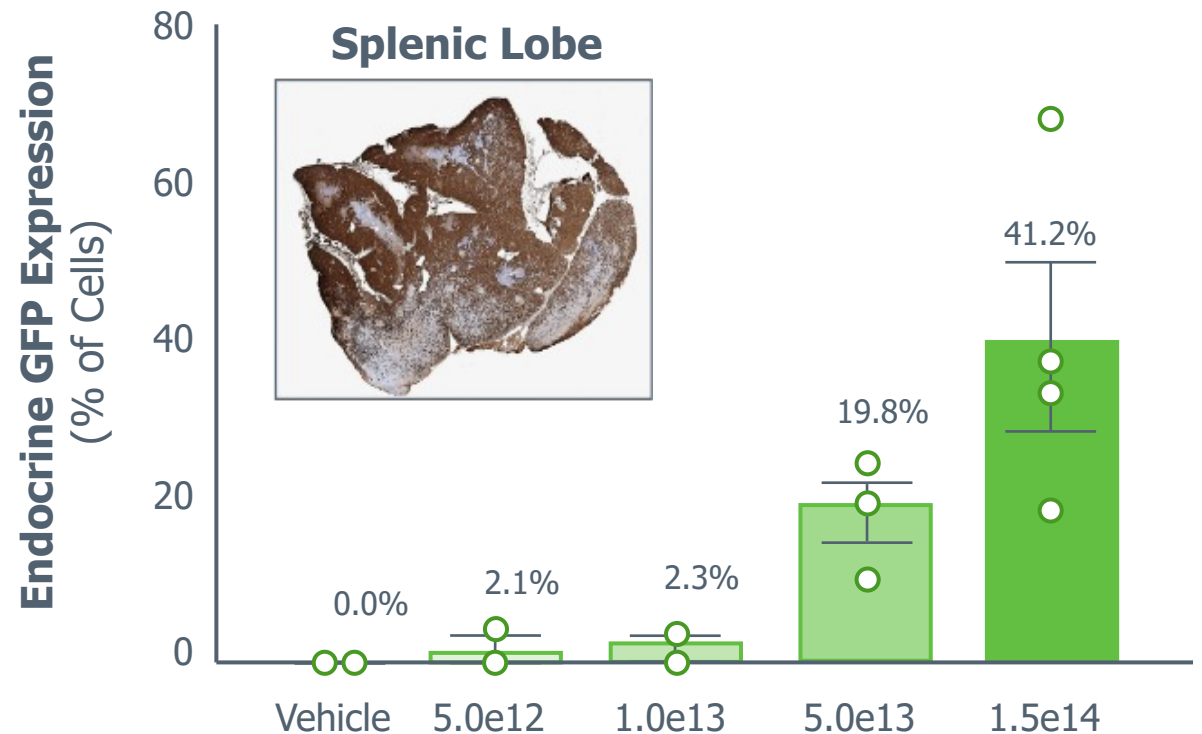
Yucatan pig model anatomy similar to humans

Proprietary device and endoscopic procedure previously described[1,2]

>50 animals treated with 100% technical success; no adverse safety signals to date

Dose-dependent AAV-GFP expression in targeted pancreatic lobe[1,2]

Low viral genome dose with limited systemic virus exposure – due to local delivery[2]

Yucatan Pig Islet Transduction

Splenic Lobe

Endocrine GFP Expression (% of Cells)

Vehicle: 0.0%
5.0e12: 2.1%
1.0e13: 2.3%
5.0e13: 19.8%
1.5e14: 41.2%

Compared to Chronic Semaglutide, Can One-Time GLP1 PGTx:
Improve Glycemia
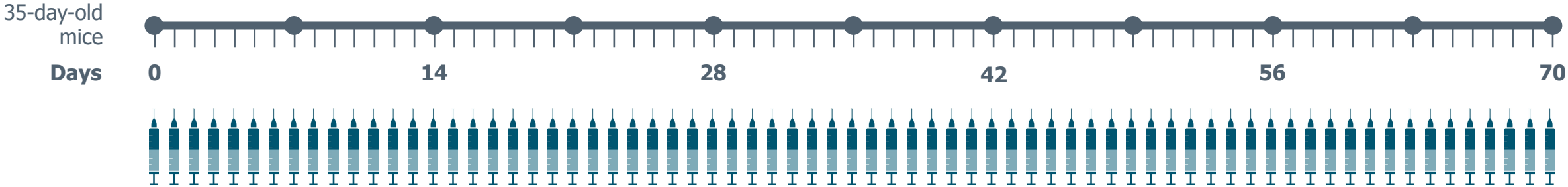Delay T2D Progression
and Prevent Weight Gain?

GLP1 PGTx Efficacy Proof of Concept

db/db murine model *de facto* standard for T2D development

Single I.P. Injection
(AAV-INS-GLP1RA or Vehicle)

35-day-old mice

Days   0   14   28   42   56   70

Daily S.C. Injection
Semaglutide (10 nmol/kg*) or
Vehicle

*Semaglutide dose selected for
glucose-lowering optimization[1]

Efficacy/MOA (day 0-70)
Weekly Fasting Blood Glucose
Biweekly Insulin
Weight

Sacrifice (days 58-70)
Organ Histology
Pancreatic GLP1RA Protein
Serum GLP1RA Protein

1. CDER (2017) Semaglutide NDA Application (209637Orig1s000), Section 4.4 Nonclinical Pharmacology/Toxicology. AAV=adeno-associated virus, GLP1=glucagon-like peptide 1, GLP1RA= GLP1 receptor agonist, INS=insulin promoter, I.P.=intraperitoneal, MOA=mechanism of action, PGTx=pancreatic gene therapy, S.C.=subcutaneous

GLP1 PGTx Expression Restricted to Pancreatic Islets
Safety and feasibility in *db/db* are reassuring thus far

High specificity for pancreas

Insulin promoter effectively restricts transgene expression to pancreatic islets

No detectable expression in off-target tissues (e.g., exocrine pancreas)

Favorable toxicity profile

No abnormal findings thus far in animal behavior or clinical chemistries

Histopathologic analysis showed no evidence of pancreatitis or pancreatic cancer

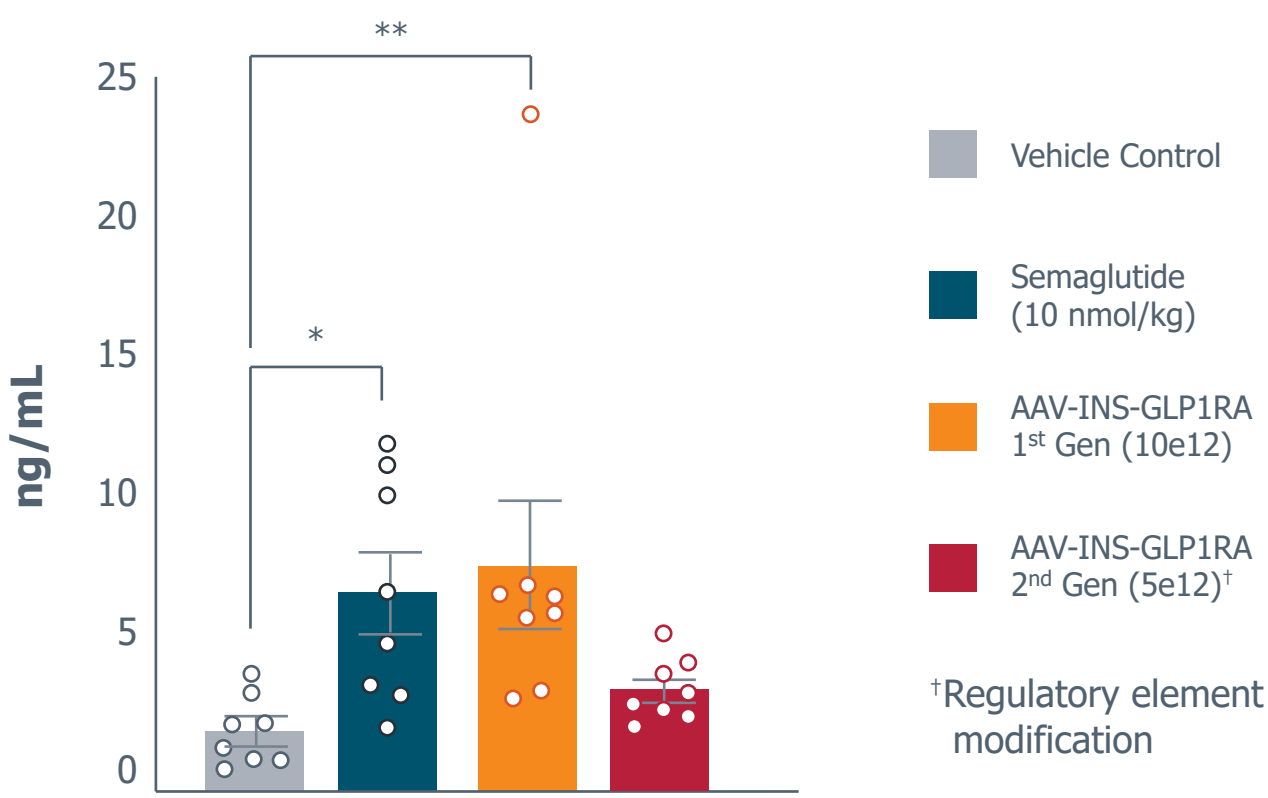GLP1=glucagon-like peptide 1, PGTx=pancreatic gene therapy

Glucose Lowering Efficacy in *db/db* Mouse
GLP1 PGTx improves fasting glucose vs. daily semaglutide

A) Fasting Blood Glucose
(Week 8, 4–6 hour fasted)
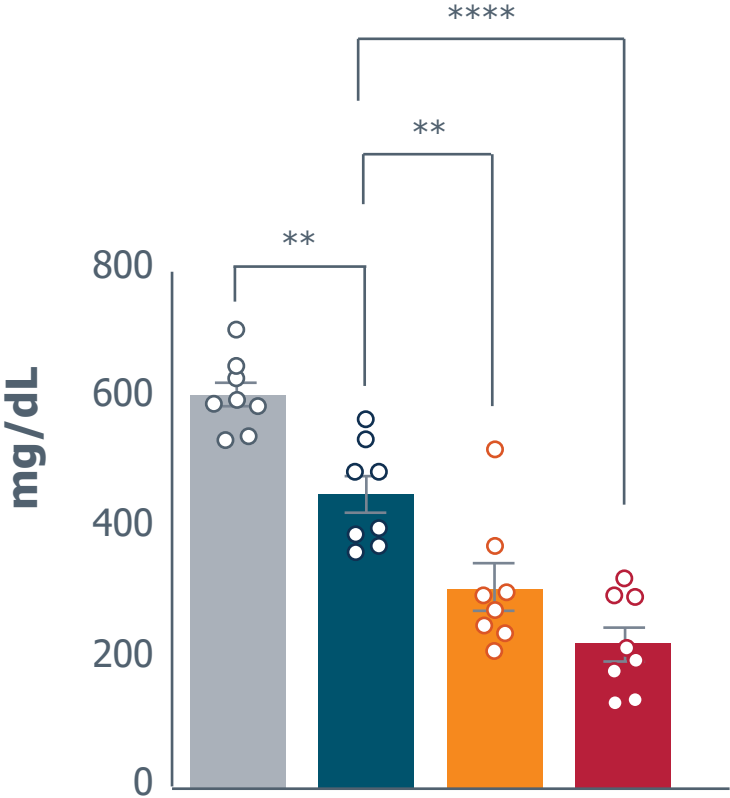
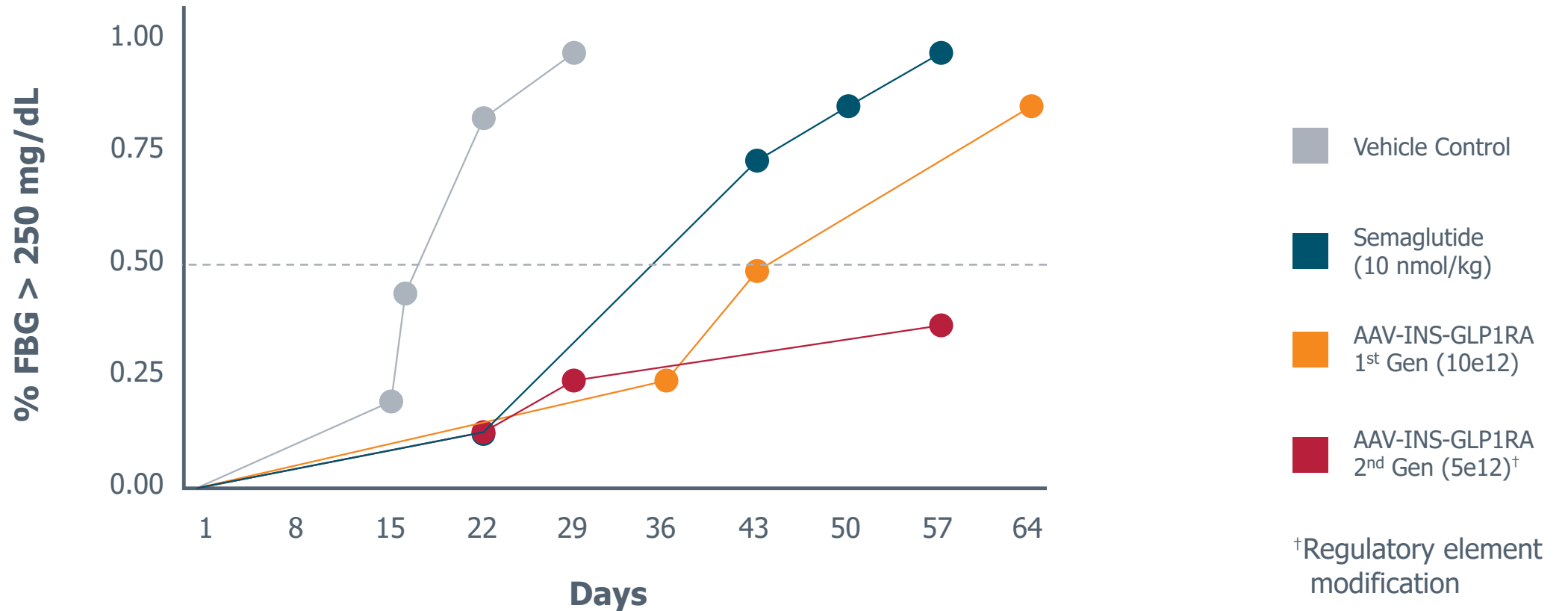**B) Fasting Insulin**
(Week 8, 4–6 hours fasted)

Vehicle Control

Semaglutide
(10 nmol/kg)

AAV-INS-GLP1RA
1st Gen (10e12)

AAV-INS-GLP1RA
2nd Gen (5e12)†

†Regulatory element
modification

Mean ± SEM shown; *p<0.05, **p<0.01, ****p<0.0001; n=8 per group. AAV=adeno-associated virus, Gen=generation, GLP1=glucagon-like peptide 1, GLP1RA=GLP1 receptor agonist, INS=insulin promoter, PGTx=pancreatic gene therapy

Disease Progression and Durability
GLP1 PGTx shifts progression of disease vs. daily semaglutide

AAV=adeno-associated virus, FBG=fasting blood glucose, Gen=generation, GLP1=glucagon-like peptide 1, GLP1RA=GLP1 receptor agonist, INS=insulin promoter, PGTx=pancreatic gene therapy
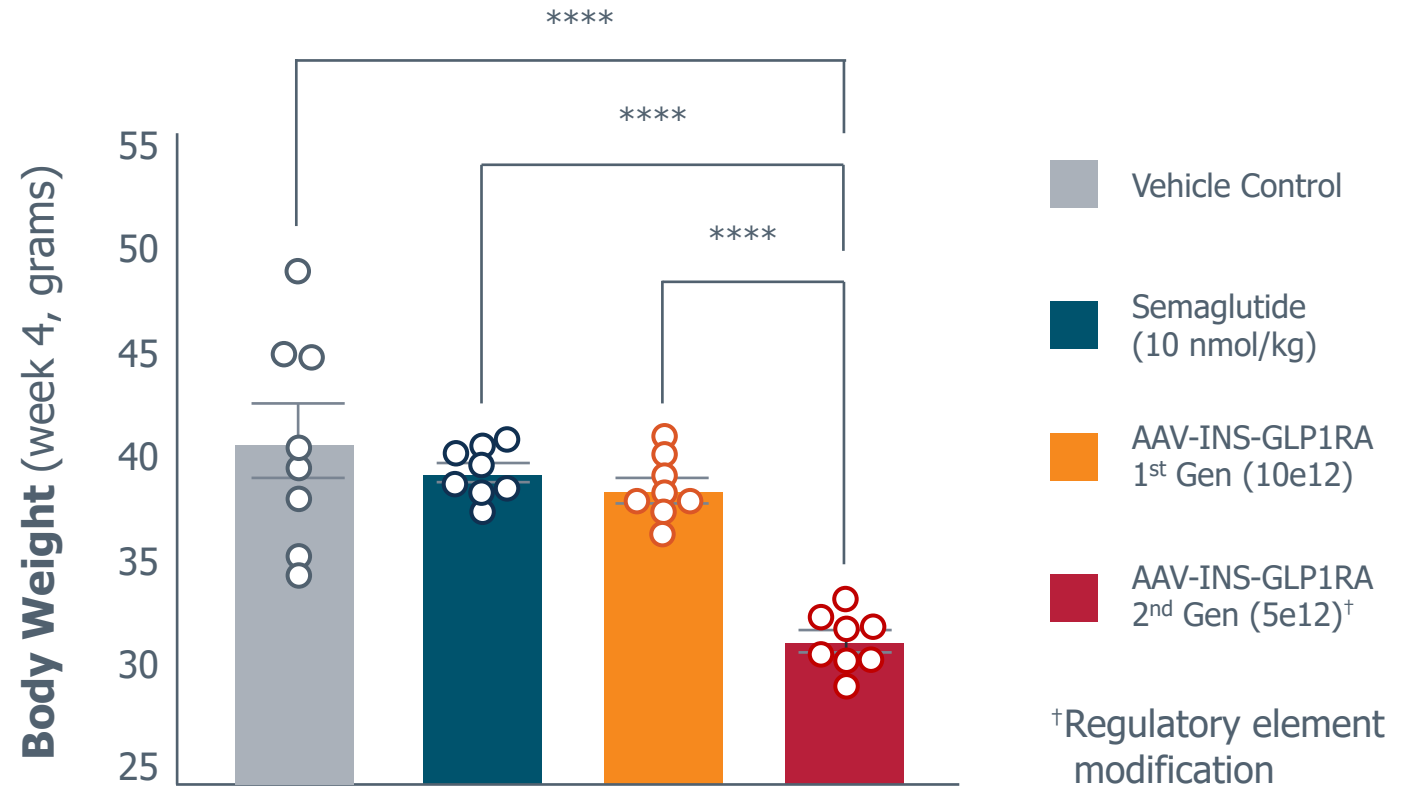
Body Weight Change
GLP1 PGTx prevents weight gain vs. daily semaglutide

23% lower total body weight with PGTx compared to vehicle

20% lower total body weight with PGTx compared to semaglutide

Mean ± SEM shown; ****$p<0.0001$; n=8 per group. AAV=adeno-associated virus, Gen=generation, GLP1= glucagon-like peptide 1, GLP1RA=GLP1 receptor agonist, INS=insulin promoter, PGTx=pancreatic gene therapy
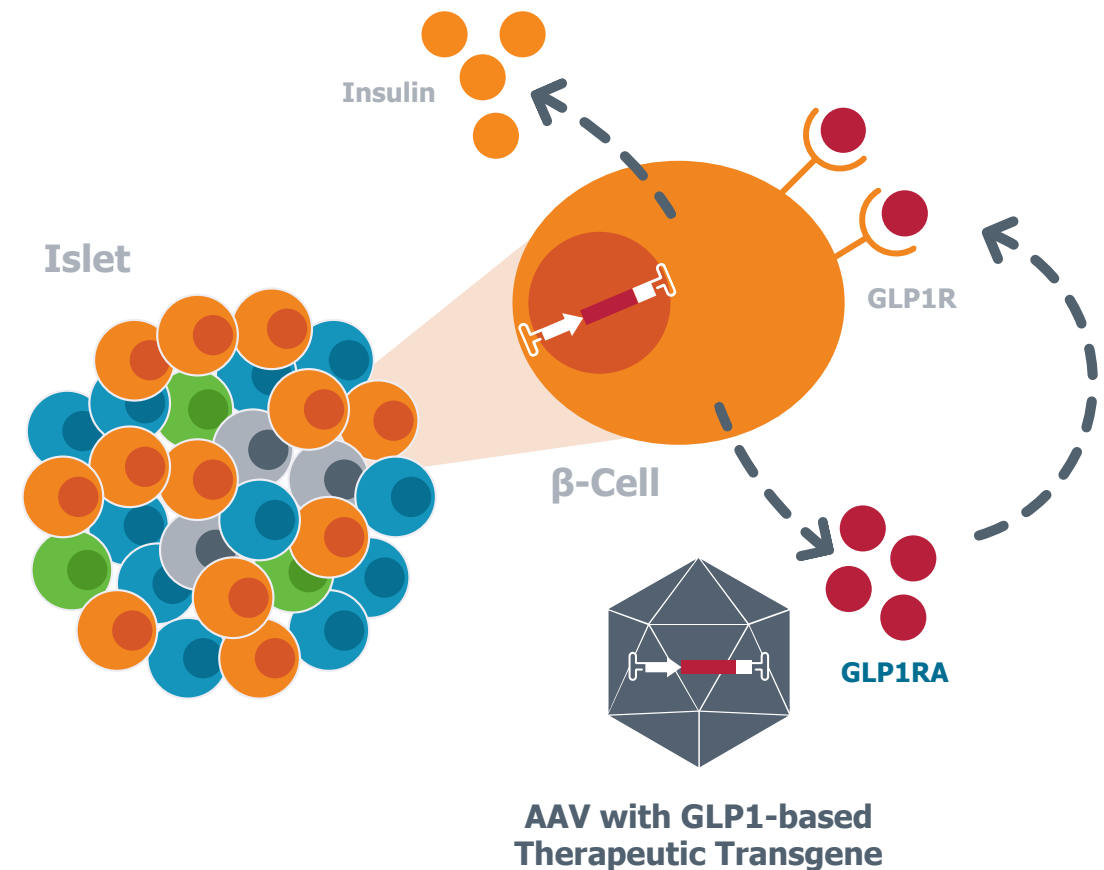
GLP1 PGTx Safety and Pharmacology Studies in Model Systems

Early feasibility and safety observations in *db/db* mice and Yucatan pigs are encouraging

Compared to chronic semaglutide, single-dose PGTx improves fasting glucose, delays T2D progression, and prevents weight gain in *db/db* model of T2D

PGTx lead optimization demonstrates potential for even greater efficacy in T2D and obesity with low pancreatic dose.

Data raise important questions about role of pancreatic islet in regulation of metabolic setpoint

Insulin
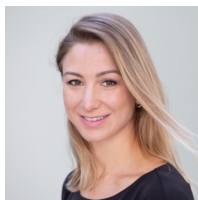
Islet

β-Cell

GLP1R

GLP1RA

AAV with GLP1-based Therapeutic Transgene

AAV=adeno-associated virus, GLP1=glucagon-like peptide 1, GLP1R=GLP1 receptor, GLP1RA= GLP1R agonist, PGTx=pancreatic gene therapy

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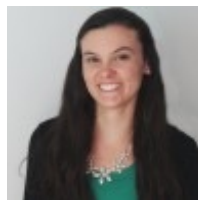
Virus and Gene Delivery

Lin Quek	Gary White	Suya Wang	Keiko Ishida

Device Engineering
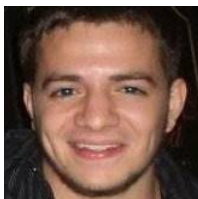
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