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Investigating the contribution of the HNF-1αG319S gene variant to early-onset type 2 diabetes (T2D) using mouse islets and MIN6 β-like cells

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Background:

40% of Manitoban Indigenous youth with T2D harbor the HNF-1αG319S variant; the strongest genetic predictor of this disease currently known. Despite clinical evidence implicating pancreatic β-cell defects under post-colonial dietary stress, the mechanistic impact of HNF-1αG319S on β-cell dysfunction remains unknown.

Objective:

We hypothesize the G319S variant alters β-cell metabolism and insulin secretion depending on nutrient availability.

Methods:

To create appropriate models to test our hypothesis, CRISPR/Cas9 was used to knock-in the G>A.955 single nucleotide substitution into clonal MIN6 β-cells ("G319S-MIN6") and a C57/BL6 mouse model. Glucose-stimulated insulin secretion (GSIS) assays were performed on MIN6 β-cells (N=6) and isolated islets (N=3) from 3-month male wildtype and heterozygous mice. Follow-up *in vitro* measurements included qPCR (N=6) and GSIS following 24-hour exposure to 0.25mM palmitate (N=3).

Results:

HNF-1αG319S did not affect GSIS in MIN6 β-cells (8.73 vs. 8.91 ng insulin/μg DNA/hr) or isolated islets (1.76 vs. 1.98 ng insulin/μg DNA/hr); however, basal insulin secretion decreased 3.2-fold relative to WT-MIN6 and 1.8-fold relative to wildtype mice. Metabolic gene expression was altered in G319S-MIN6, including a 4-fold downregulation in glucokinase (glucose metabolism) and a 2-fold upregulation in carnitine palmitoyltransferase-1A (mitochondrial fatty-acid uptake). With a seemingly increased capacity to shuttle fatty acids towards beta-oxidation, G319S-MIN6 cells maintained 15-fold GSIS under chronic lipotoxic stress, which otherwise severely impaired GSIS in WT-MIN6.

Conclusion:

HNF-1αG319S alters the gene expression profile of MIN6 β-cells. Suprisingly, the HNF-1α variant does not impair GSIS in MIN6 β-cells or islets from male heterozygous mice, and may even confer resistance to palmitate-induced impairments in GSIS. Rather, G319S-expressing MIN6 β-cells and islets display reduced basal insulin secretion, that although beneficial for maintaining whole-body glucose

homeostasis during fasting, may trigger hyperglycemia when combined with excessive carbohydrate intake in the fed state. Future studies will address whether reduced dietary glucose and increased lipid consumption protects against T2D in G319S-expressing mice.