Islet on a Chip – Device Instrumenting for Real-time Electrophysiology

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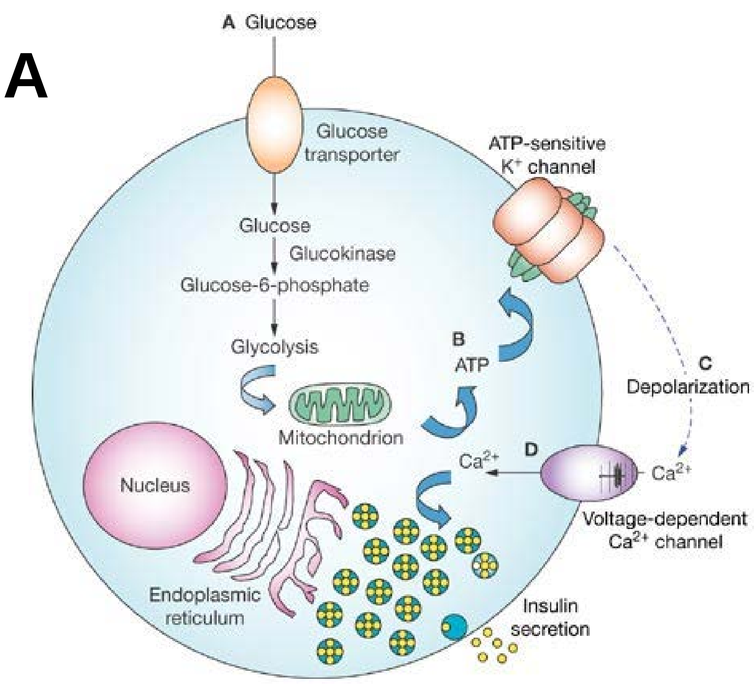
Background

- Glucose stimulated insulin secretion in beta cells is mediated by trans-membrane glucose transport, glycolytic ATP production, membrane depolarization by ATP-sensitive potassium ion channels, and calcium influx by voltage-gated calcium ion channels, leading to calcium-dependent insulin granule exocytosis
- Microphysiological systems containing SC-beta cells must recapitulate their cellular function to offer physiological insight into disease mechanisms or to uncover new therapies
- <u>Hypothesis:</u> Electrophysiological measurements of SC-beta cells signify a necessary predictor of phenotypic maturity
- Patch clamp recordings provide a fine resolution understanding of glucose sensitivity
- Integration of microelectrode arrays (MEAs) with our Islet on a Chip platform can continuously monitor the collective activity of multiple clusters



Leon et al. *Nature Clinical P. Endo.* (2007)

Adapted from Ashcroft et al. *Prog. Biophys.* (1989)

Figure 1 | Islet on a Chip and Cellular Mechanism of Insulin Release of Pancreatic Beta Cell.
(A) Glucose stimulation of insulin release. Glucose binds to glucose transporters on the cell surface. This initiates a signaling cascade that leads to K-ATP sensitive ion channels closing. Depolarization of the cell occurs by influx of calcium through L-type calcium channels and leads to insulin release. (B) Mouse primary beta cell action potentials at 10mM and 15mM glucose.

Main Achievements

1) Single cell electrophysiology of SC-beta cells in low and high glucose

2) Microelectrode Array (MEA) of SC-beta cells

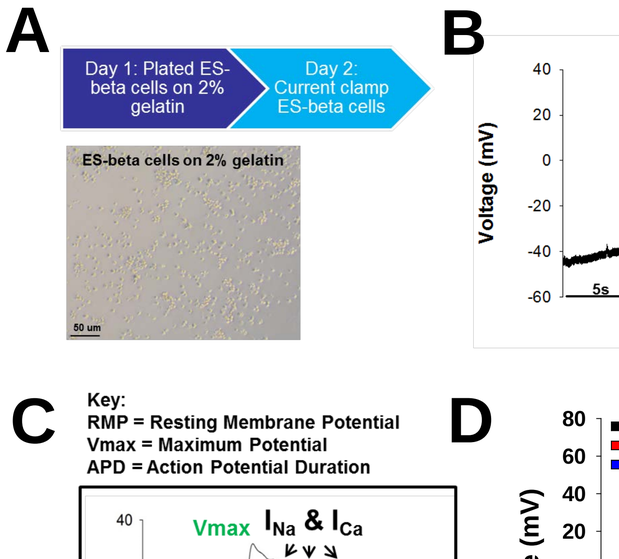


Figure 2 | Cellular Electrophysiological Response of SC-β cells to glucose.
(A) SC-derived beta cells are plated on 2% gelatin and cultured for 24 hours before patch clamp experiments (B) SC β cell response to low (3.3mM) and high (16.7mM) glucose (C) Single action potential at 5mM glucose with key for resting membrane potential, maximum voltage, and action potential duration (D) Summary of action potential parameters for SC-β cells at different glucose concentrations (E) Summary of action potential duration: action potentials are significantly shorter at 16.7mM vs 3.3mM and 5.0mM glucose (F) Upstroke velocity of action potentials at 3.3, 5.0, and 16.7mM glucose.

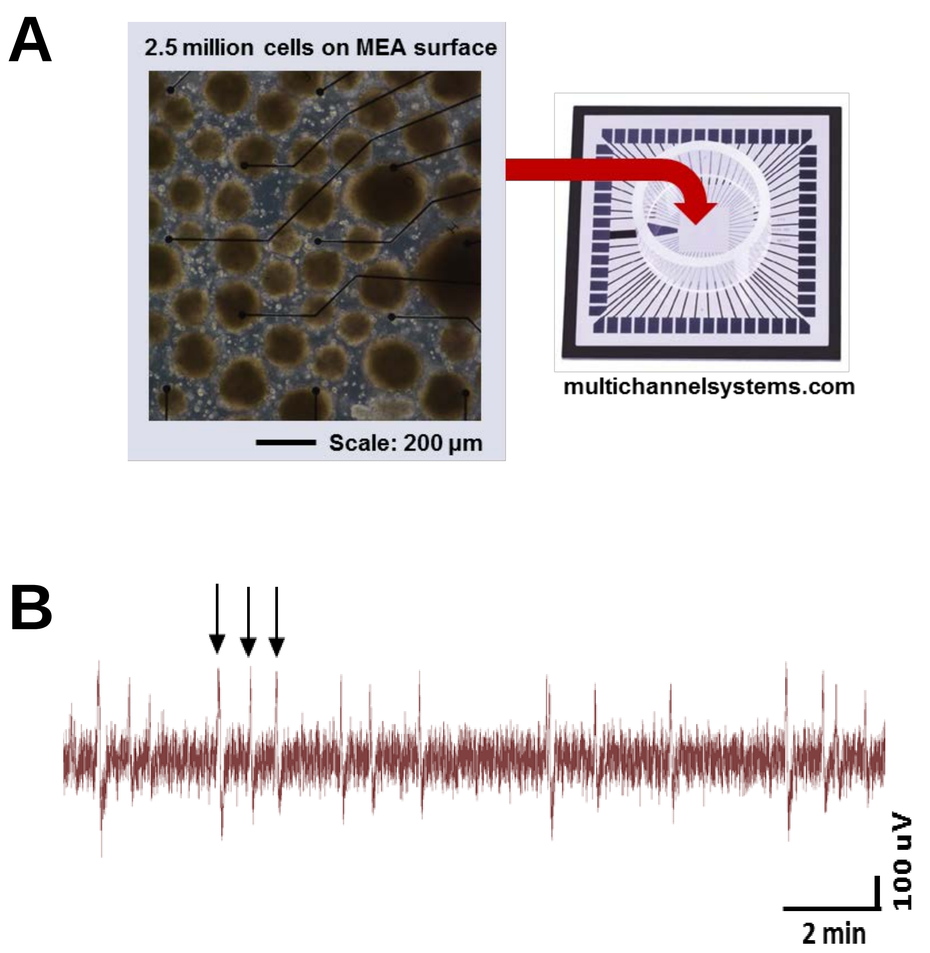


Figure 3 | Microelectrode array of SC-β cells.
(A) SC-β cell clusters elicit an extracellular electrical activity that may be measured when seeded onto a microelectrode array (MEA) (B) Electrical activity with representative spiking (black arrows) from one β cell clusters, filtered with a 4th order Bessel filter, low pass 100 Hz. Cells were loaded directly into an open-well MEA in CMRL-S medium with 5.5 mM glucose.

Future Directions

<u>Future aims of the project will focus on:</u>

1) Identifying cell types from ES-derived beta cells lines with single-cell RNA sequencing and developing reporter cell lines; 2) Investigating the electrophysiological response to glucose of ES-beta cells and comparing cell quality metrics to primary islets; 3) Engineering islets from ES-derived beta cells for MEA analysis; 4) Integrating MEAs with the Islet on a Chip platform for real-time electrophysiological monitoring

Acknowledgements

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