α-cell Response to Low Glucose

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Background

- Islet of Langerhans is made up of α-, β- and other cells

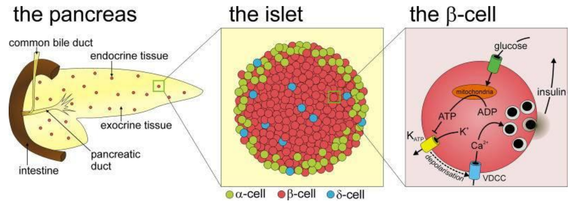

Figure 1: Islet composition.

- Islet plays a role in glucose homeostasis

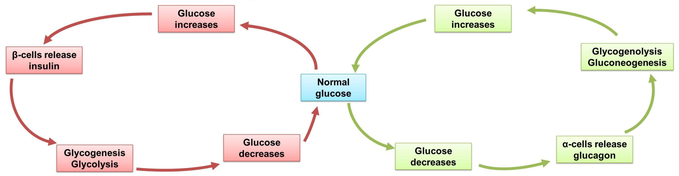

Figure 2: Glucose homeostasis cycle.

- α-cell response to glucose is poorly understood
- Studying NAD(P)H and calcium oscillations

Equipment

- Mouse model
  - Sacrificed at ~8 weeks
  - tdRFP under control of the glucagon promoter
  - Enzymatic digestion of acinar tissue to isolate islets
- Microfluidic device

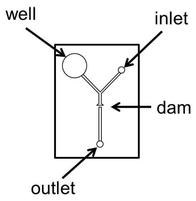

Figure 3: Microfluidic device schematic.

- Confocal microscope

Figure 4: Confocal microscope. Arrow indicates where microfluidic device is placed.

Ca^{2+} oscillations

- Measured at 0.1, 0.5, 1 and 5 mM glucose
- Active cells: Cells that are oscillating or showing steady increase in calcium
- Fluo4-AM: cell permeable calcium indicator dye

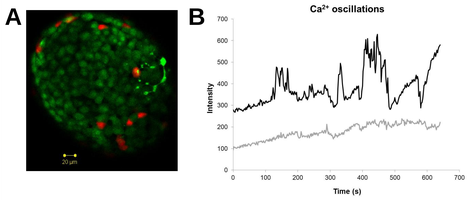

Figure 5: Ca^{2+} oscillations in the α-cell. A. Islet loaded with Fluo4-AM. Calcium is indicated in green. α-cells are indicated in red. **B.** Representative trace of change in calcium.

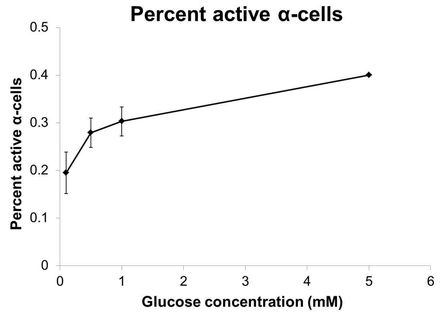

Figure 6: Percent active α-cells. n=1-4. Data shown is the mean S.E.

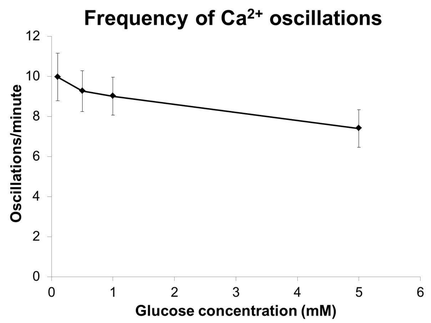

Figure 7: Frequency of Ca^{2+} oscillations. Time period of 5 minutes. n=1-4. Data shown is the mean S.E.

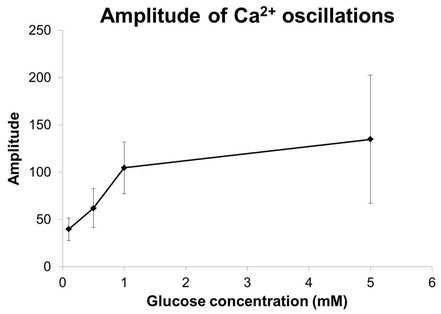

Figure 8: Amplitude of Ca^{2+} oscillations. n=1-4. Data shown is the mean S.E.

NAD(P)H

- Dose response from 0.01 mM to 5 mM glucose
- NAD(P)H is autofluorescent

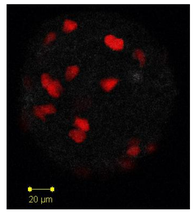

Figure 9: Overlay of NAD(P)H and tdRFP signal from an islet. Gray is NAD(P)H autofluorescence. Red is α-cells.

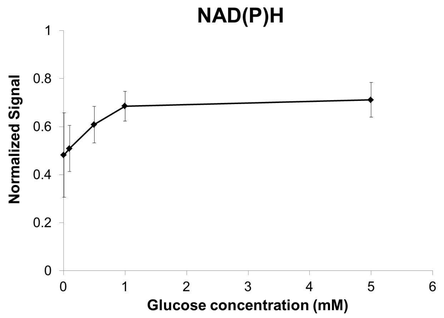

Figure 10: NAD(P)H autofluorescence. n=1-5. Normalized to NaCN (maximum fluorescence). Data shown in the mean S.E.

Summary

- Minimal activity at low levels of glucose
- Increases as glucose concentration increases
- Suggests left-ward shift compared to β-cells

- Next steps:
  - Glucagon secretion
  - Lower levels of glucose (0.01 mM)

References

1. Gromada J, Franklin I, Wollheim, CB. *Endrocrine Reviews* 28.1 (2007): 84-116.
2. MacDonald, PE, Rorsman P. *PLoS Biology* 4.2 (2006): 167-171.

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