Monoclonal alpha-cells display a heterogenous calcium response when subjected to glucose stimulation.

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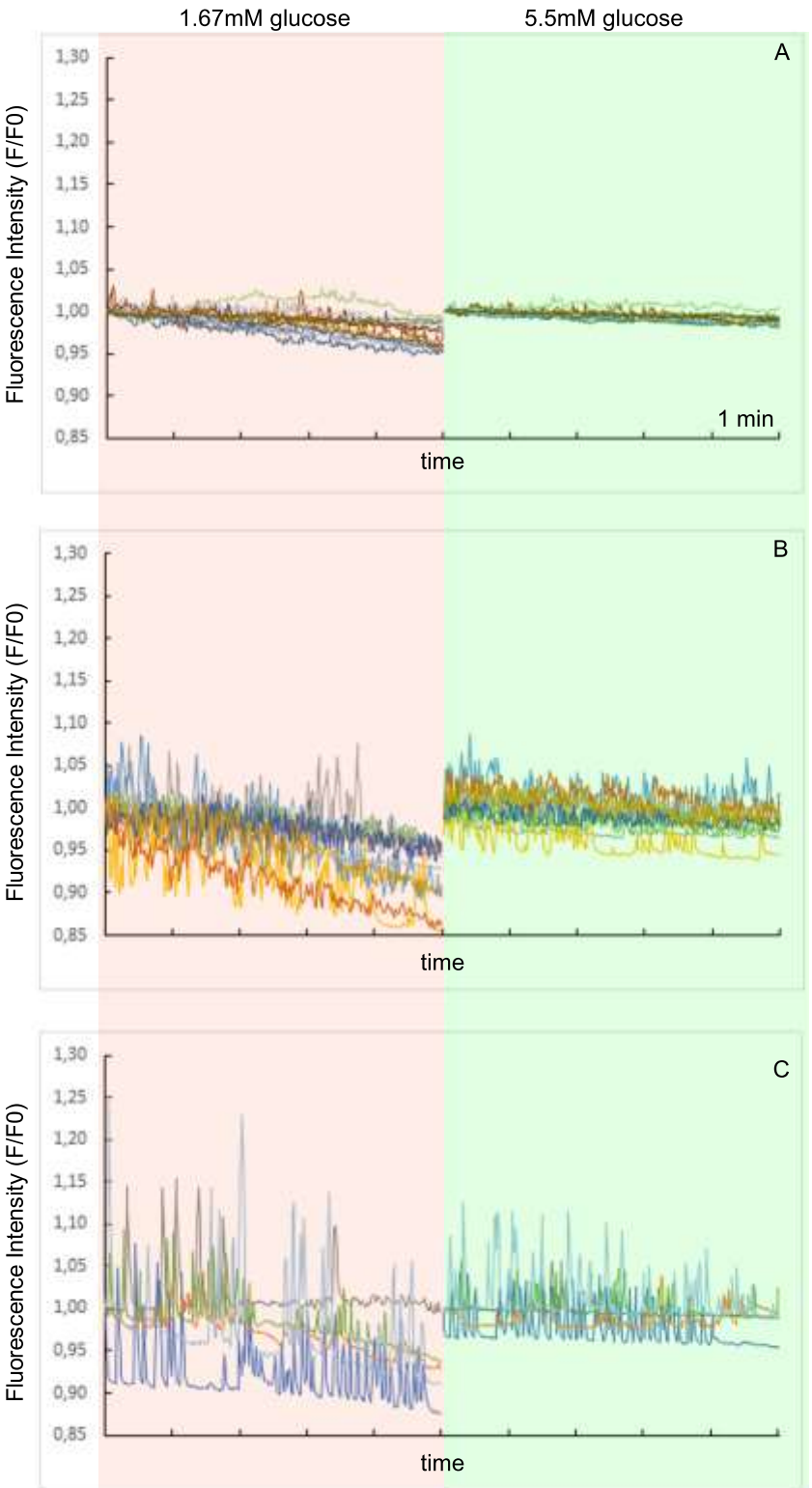
Background
In both health and disease, little is known about the mechanisms behind glucagon secretion, whilst there are indications for alpha-cell dysfunction in type 1 and 2 diabetes, contributing to inappropriate glucagon response in patients. For beta-cells, pacemaker-like behavior in calcium metabolism has been shown to be necessary for insulin secretion, but it is largely unclear how calcium fluxes in alpha-cells contribute to glucagon secretion. In this study, we monitor calcium activity of individual alpha-cells, in an effort to describe the general cell response to increased glucose levels.

Methods
The murine alpha-TC6 cell line was seeded as a monolayer at different densities, and incubated with 5uM of calcium indicator Cal-520, followed by imaging on an Andor Dragonfly spinning disk microscope in HBSS with 10mM HEPES containing low (1.67mM) or normal (5.5mM) glucose. Recordings were analyzed with Fiji software for mean intensity for Regions of Interest (ROIs) containing one cell per ROI and normalized to the first recorded fluorescence (F0).

Results
Individual alpha-cell calcium responses found, were dividable in 3 categories: low (A, continuous activity, with peaks<0.05), high (B, continuous activity, with peaks>0.05), and pacemaker-like (C, non-continuous activity defined by irregular but repetitive periods of on/off). Quantification of individual calcium responses are described as AUC. Preliminary data suggests that in low to normal glucose stimulation, the high-activity cells (19 cells analyzed) change their behavior after 5 minutes (P=0.0174, n=1).

Conclusion
We identified three distinct types of calcium flux exhibited by alpha-cells. Only the high activity-cells seem to show a difference after changing from low to normal glucose, suggesting that the other cells may not be inherently responding to their glucose surroundings.


1.67mM glucose
5.5mM glucose
A
B
C
Fluorescence Intensity (F/F0)
1,30
1,25
1,20
1,15
1,10
1,05
1,00
0,95
0,90
0,85
1 min
time