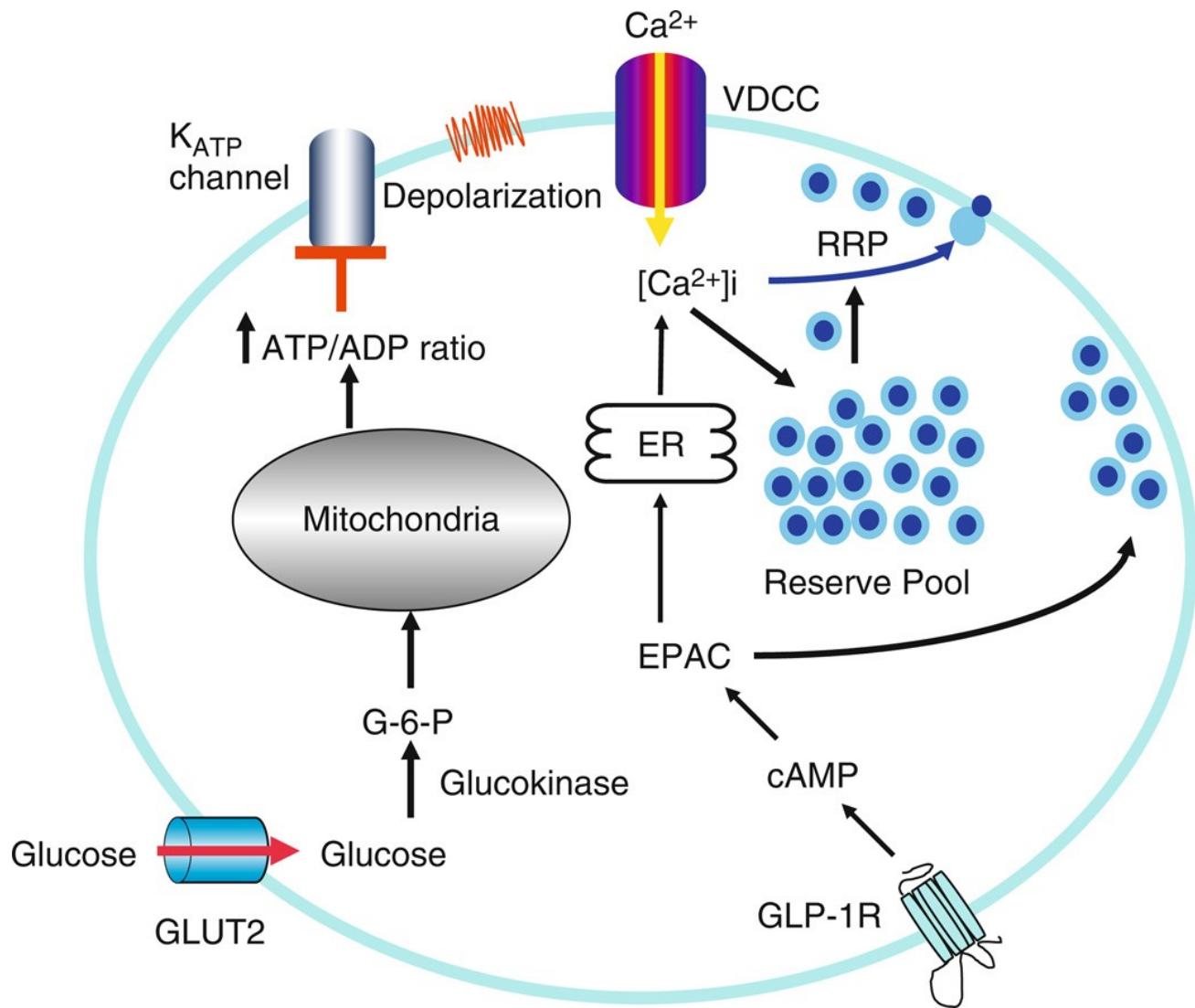


Measuring Slow Ca²⁺ Dynamics in MIN6 Monolayers and Pseudoislets

Clayton Seitz, Scherer Lab

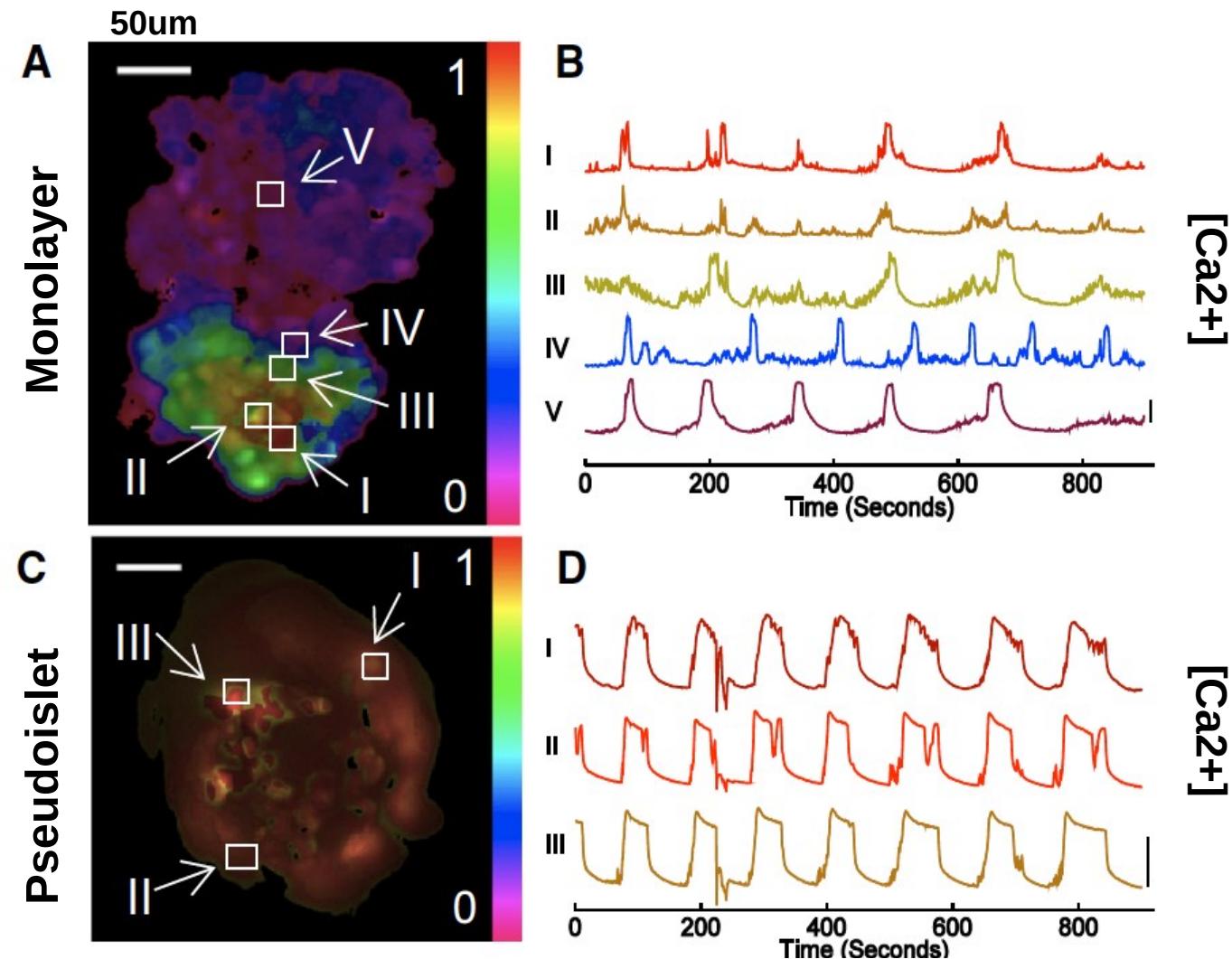
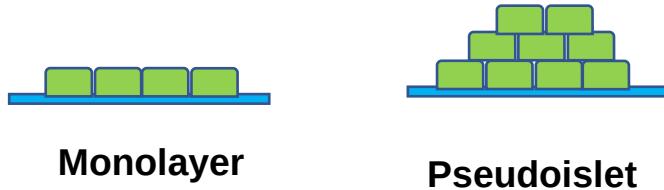
Intracellular Ca^{2+} is involved in Insulin Granule Exocytosis

- ATP-sensitive K^+ channels open and depolarize the cell
- Elevated $[\text{Ca}^{2+}]$ triggers insulin granule exocytosis
- Gap junctions allow cell-cell communication



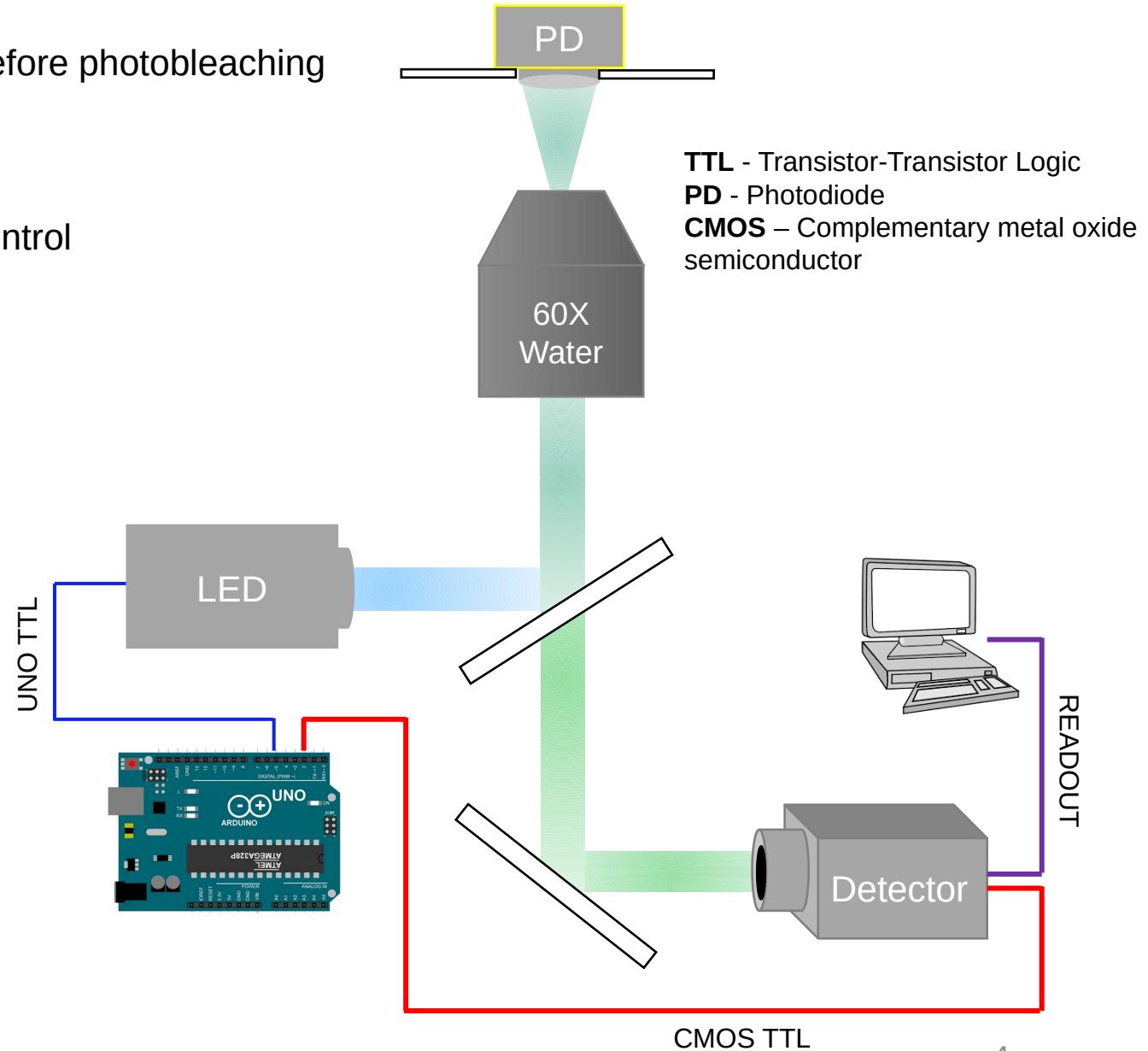
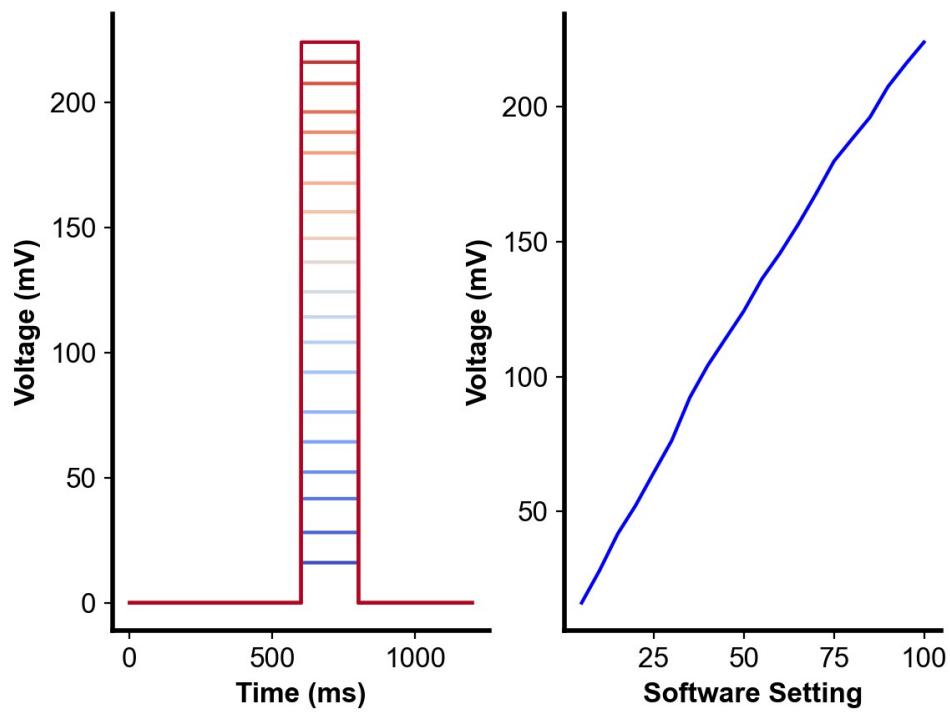
Synchronization of $[Ca^{2+}]$ oscillations is dependent on growth geometry

- $[Ca^{2+}]$ oscillations can be induced by incubation with glucose
- $[Ca^{2+}]$ can be read out using a fluorescent calcium indicator
- $[Ca^{2+}]$ oscillations are less synchronized in monolayers than pseudoislets



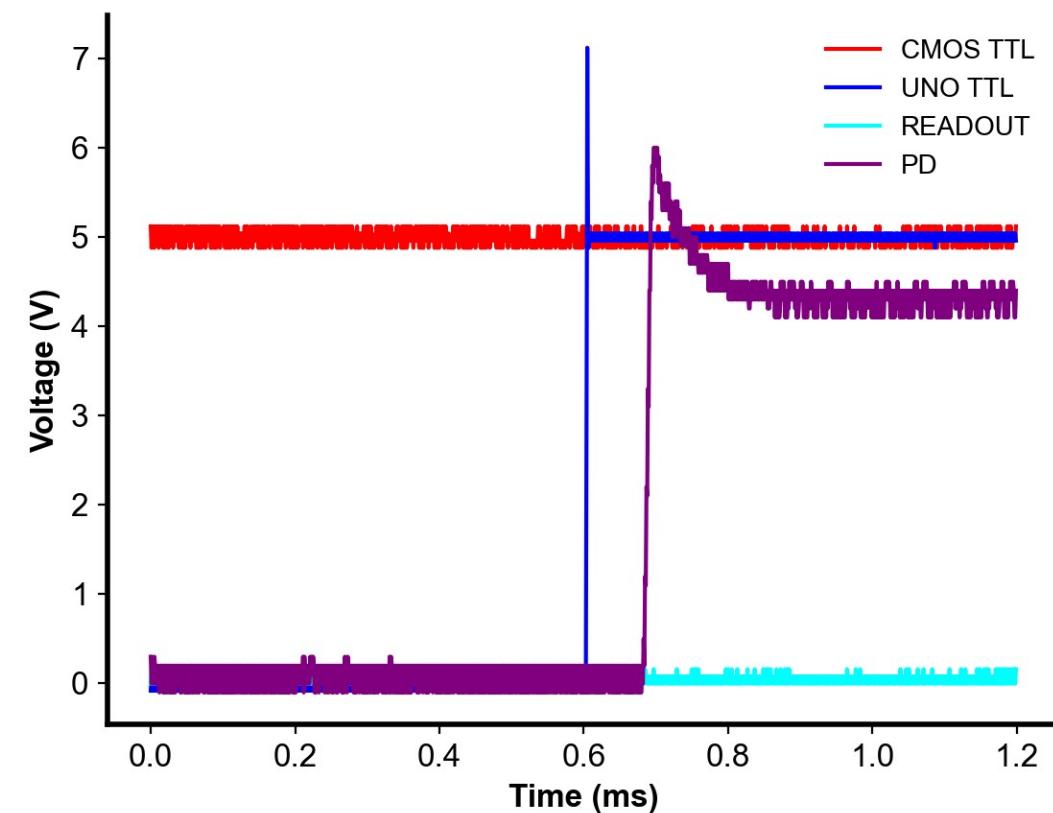
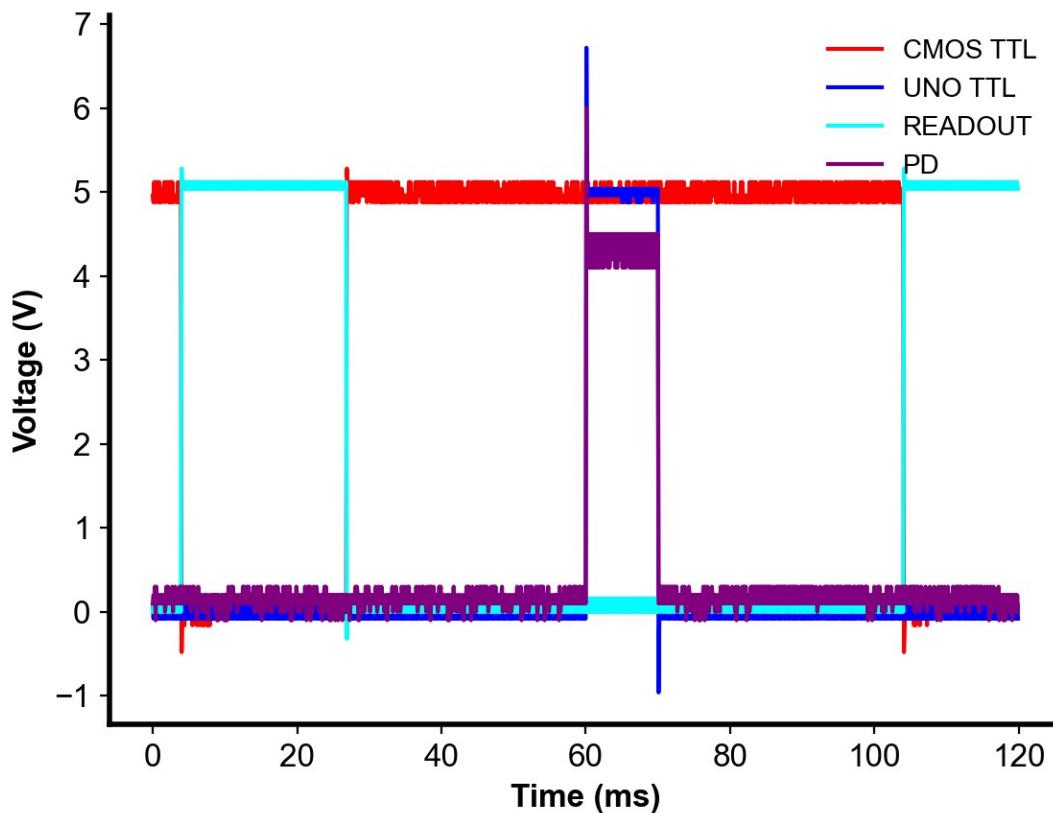
Photobleaching Imposes Constraints on Observable [Ca²⁺] Dynamics

- [Ca²⁺] calcium indicators have a finite photon budget before photobleaching
- Widefield LED excitation promotes photobleaching
- Photobleaching can be mitigated with timing/intensity control

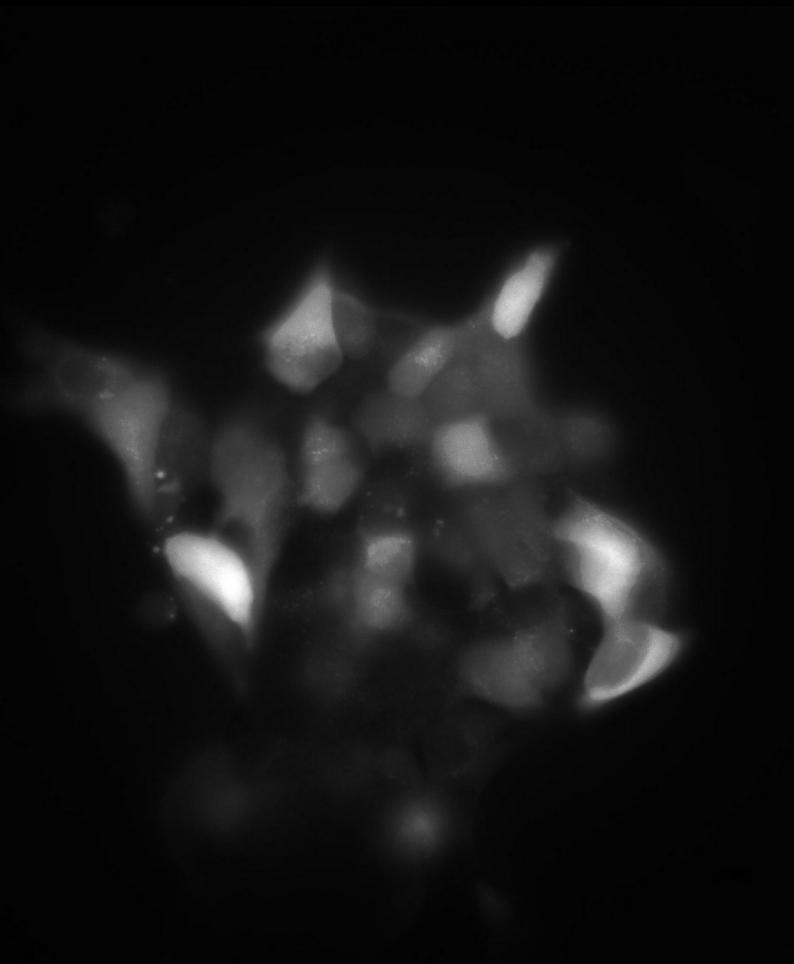


Transistor-Transistor Logic (TTL) Allows Precisely-Timed Pulsatile Excitation

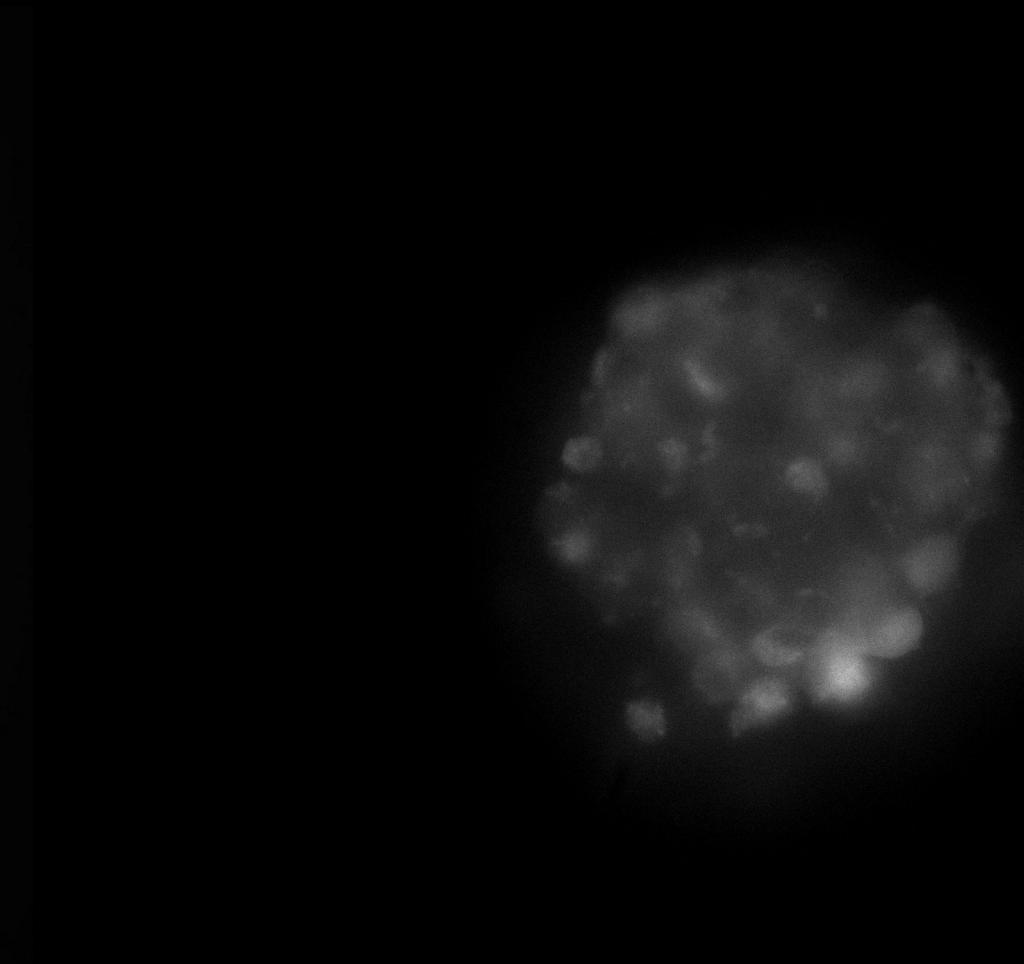
- Brief excitatory pulses during sensor exposure limits photobleaching at low frame rates



MIN6 Monolayer/Pseudoislet at 28mM Glucose



Monolayer



Pseudoislet