Supplementary Information for

Automated cellular stimulation with integrated pneumatic valves and

fluidic capacitors

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Figure S1. **Mixing cross and intensity profiles. A.** Fluorescent image of the area on the microfluidic device where the flow from the islet chamber (left), Ins* (top), and Ab (bottom) channels intersect and enter the mixing channel. The image was captured when pulling buffer from the immunoassay reagent channels and fluorescein from the perfusion channel through the islet chamber and into the mixing cross. A line (shown in red) was drawn across the mixing channel to extract the fluorescence intensity across the channel. **B.** The fluorescence intensity across the mixing channel distance. The intensity at half-height covered 34 μm of the 100 μm wide channel.



Figure S2. Insulin secretion profiles on devices with integrated fluidic capacitors. **A-G** shows the insulin release profiles from single murine islets in response to a step increase in glucose (blue line, right y-axis) from 3 to 11 mM. **H.** Control experiment where a murine islet on the device was stimulated with 3 mM glucose for the first 5 min. At 5 min, the valves switched (indicated by the downward arrow) to deliver the same glucose concentration for an additional 10 min after which the 3 mM glucose solution was removed from the device and replaced with 11 mM glucose.



Figure S3. Insulin secretion profiles on devices without fluidic capacitors. Valves were used to direct 3 or 11 mM glucose (shown by blue line on right y-axis) to single murine islets while insulin secretion (black line, left y-axis) was measured. No fluidic capacitors were used in the device. The shoulder-like bump on each trace is shown by a (*) and occurred as the glucose level increased from 3 to 11 mM.