Development of a Fluidic Microdevice for Engineering Pancreatic Islet Microenvironments

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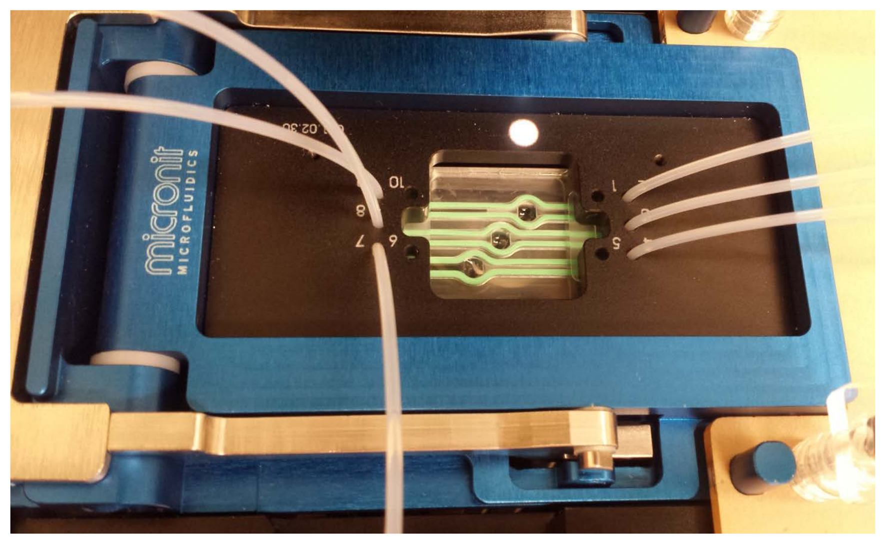
Design Goal: Simple Standardized Platform

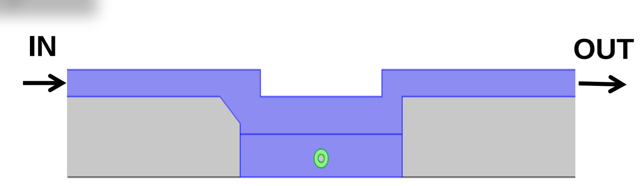


- Fluidically addressable wells
- Oxygen control
- Ready for optical microscopy
- Offline samplers
- Multiplexing ability
- Easily assembled
- Reversible sealing
- Versatile platform integration

Computational Fluid Dynamics (CFD)

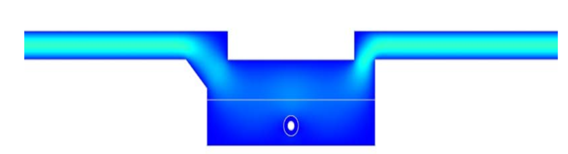
CFD to forecast fluidics and diffusion properties of the device:

- Mass Transport and Hill-type Reaction simulation in water, alginate and islet
- 2D parametric study
- Channel filled with water, capsule is made of alginate, islet is solid
- Time dependant study (0.5s time-steps, 3600s total)
- Laminar Flow ($v = 10^{-4}$ m/s)



$$\frac{\partial c}{\partial t} + \nabla \cdot (-D\nabla c) = R - \mathbf{u} \cdot \nabla c$$

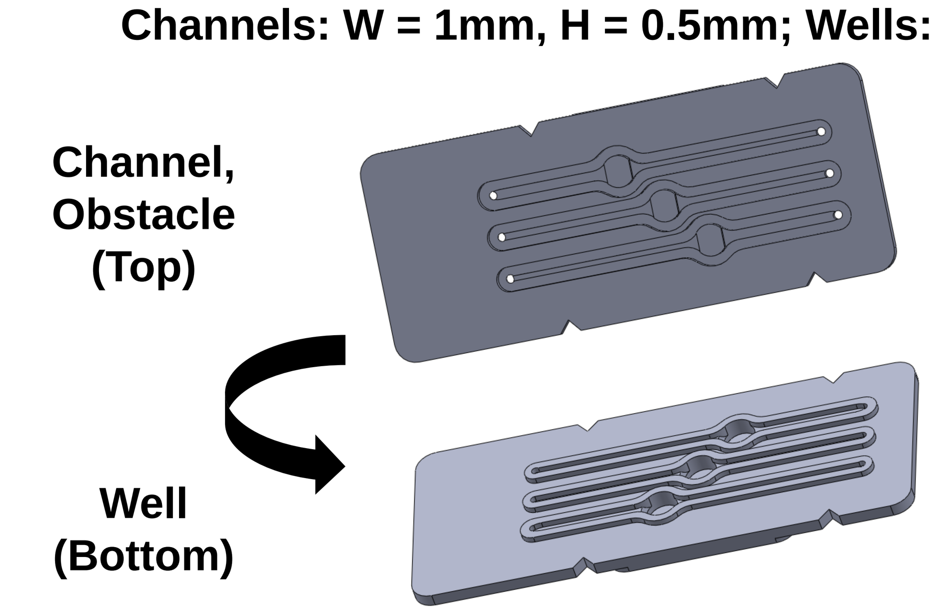
$$R = f_H(c) = R_{max} \frac{c^n}{c^n + C_{Hf}^n}$$



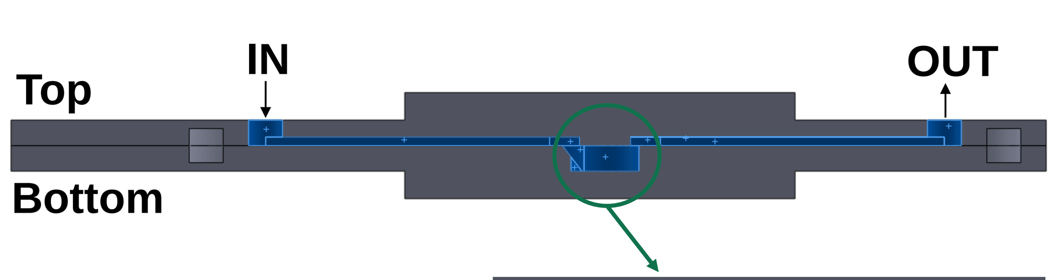
Insulin concentration | Glucose concentration | Oxygen concentration

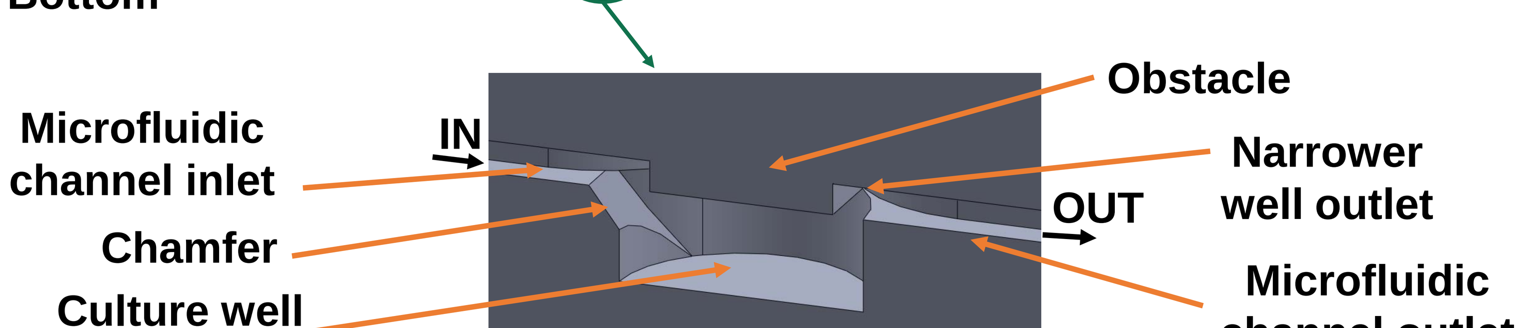
Computer Aided Design (CAD)

A 3D design of a 3 well-channel microfluidic device created in SolidWorks
Channels: W = 1mm, H = 0.5mm; Wells: W = 4mm, H = 1.5mm



- Walls-structures to isolate channels
- Parts fit into each other to impede liquid cross-talk
- No need of an insert for alignment





Section of the well (Dimetric view)

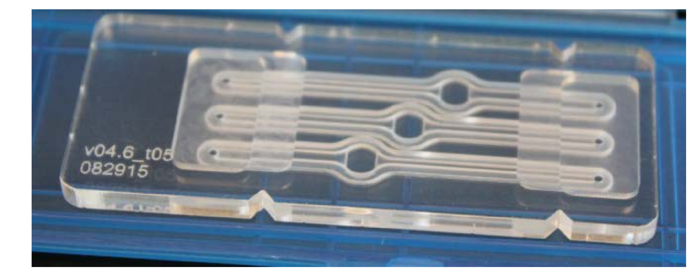
Prototype Fabrication

Roland MDX-540:

- CNC milling of wells and channels
- ±25µm tool accuracy
- Optically transparent acrylic
- Tight tolerances enable alignment

30W CO_2 Laser Engraver:

- Precision cutting
- Rapid processing



Microfluidic device parts milled from acrylic: Channels (top) and Culture wells (bottom)

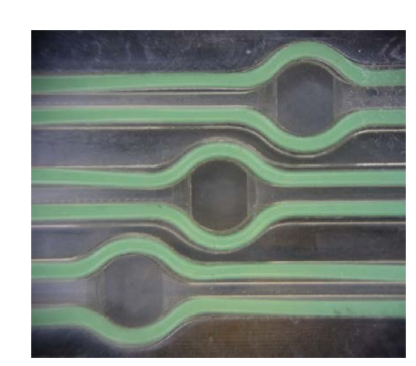
Experimental Set-up

Microdevice Assembly

- The device is designed to fit a commercial cassette (Micronit Microfluidics) that provides a convenient clamping system
- Standardized liquid ports (IDEX, Luer) enable reliable fluid connections
- Liquid sealing is guaranteed by a custom made gasket (DURASEAL 1533)
- A gasket enables a simple access to the well before and after islet seeding

Micronit Cassette

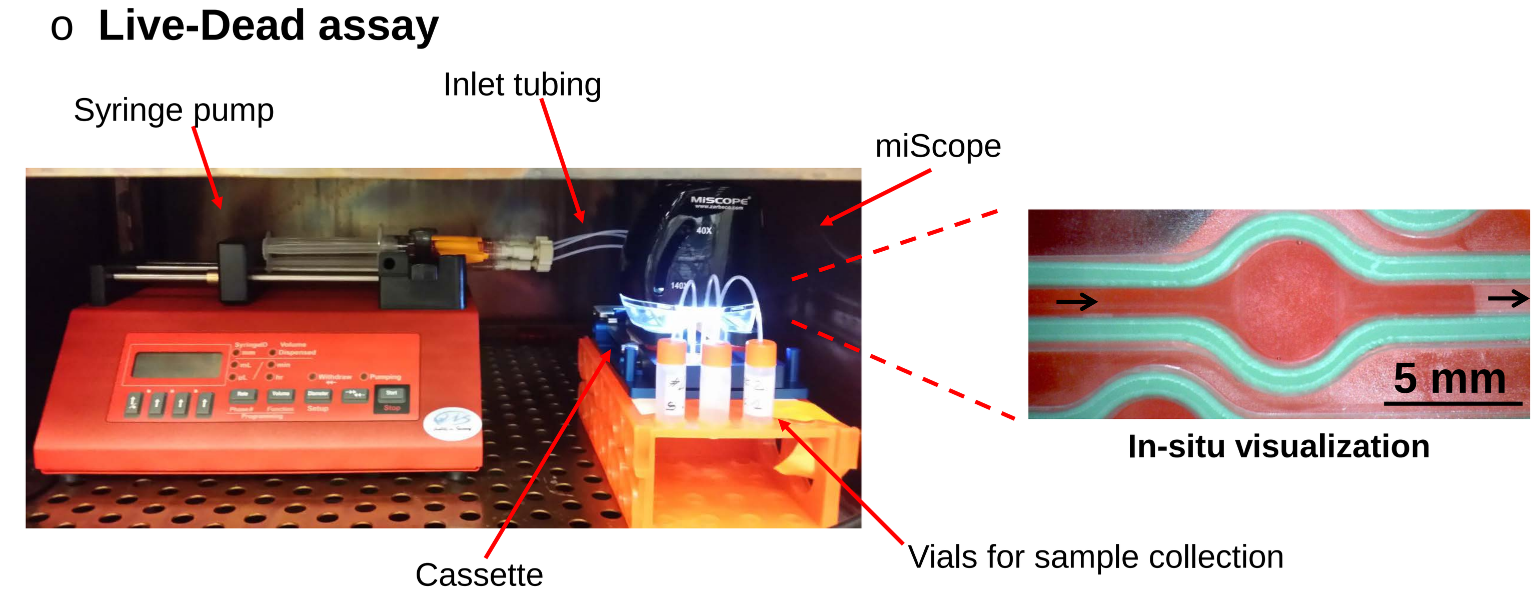
Microdevice is assembled with gasket (in green)



Microdevice is clamped and secured

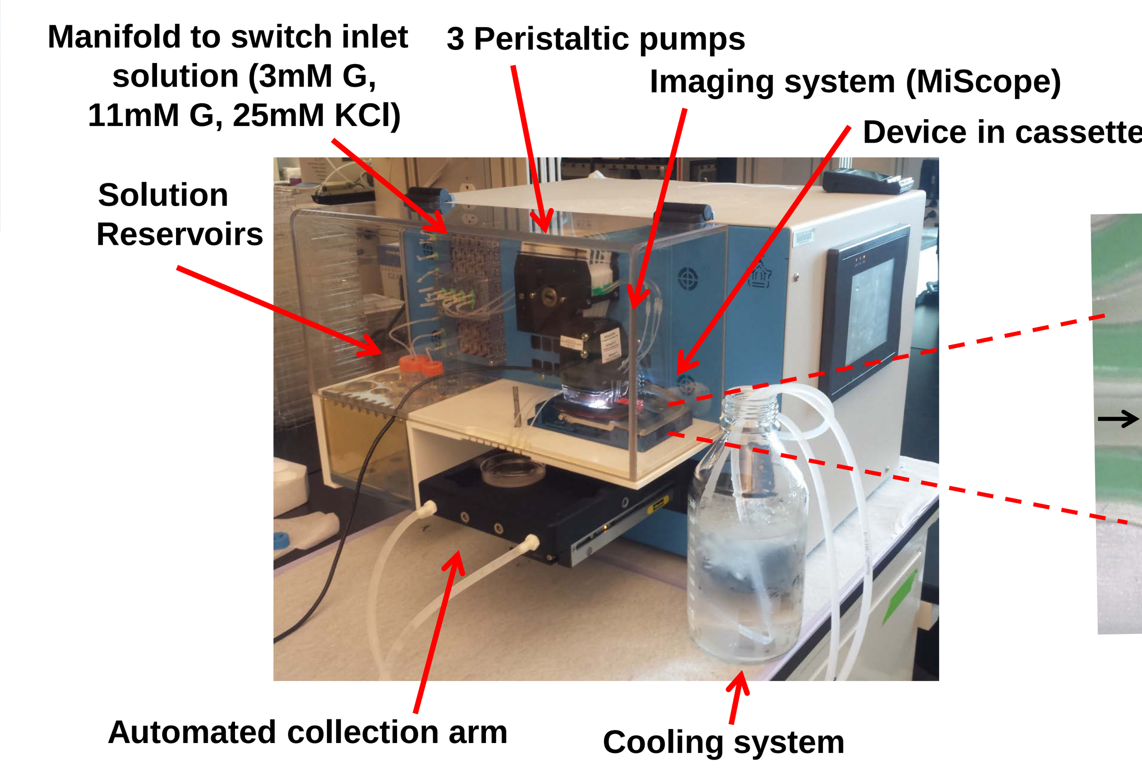
Set-up for tests in culture

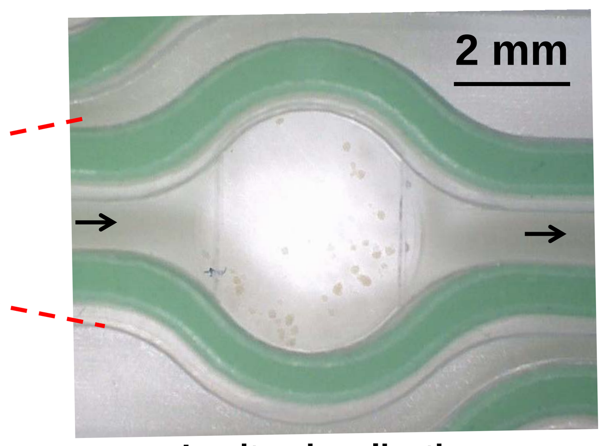
- Islets can be manually seeded and they do not drift with flow
- The microdevice can be set up in the incubator for long-term tests
  - o ELISA assay for off-line insulin measurement
- Multipurpose imaging (BF, FI, Confocal) feasible through milled surfaces
  - o Live-Dead assay



Set-up for acute tests

The device can be set up in the Biorep Perifusion Machine

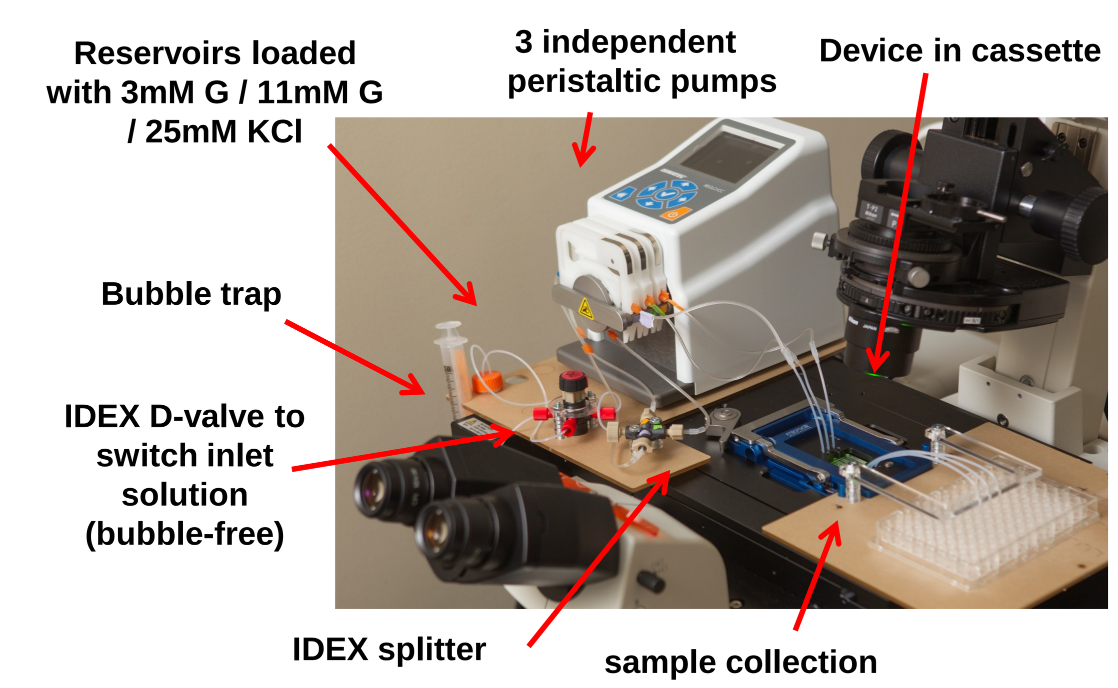


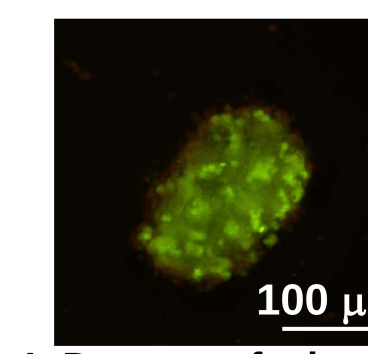


In-situ visualization

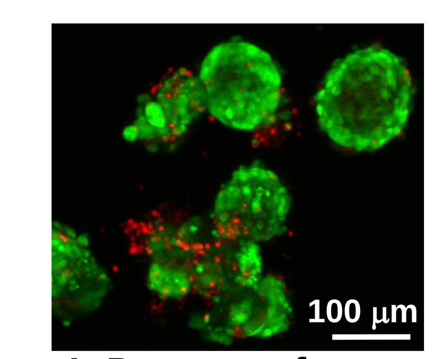
Set-up for live imaging

The device can be set up on any optical microscope





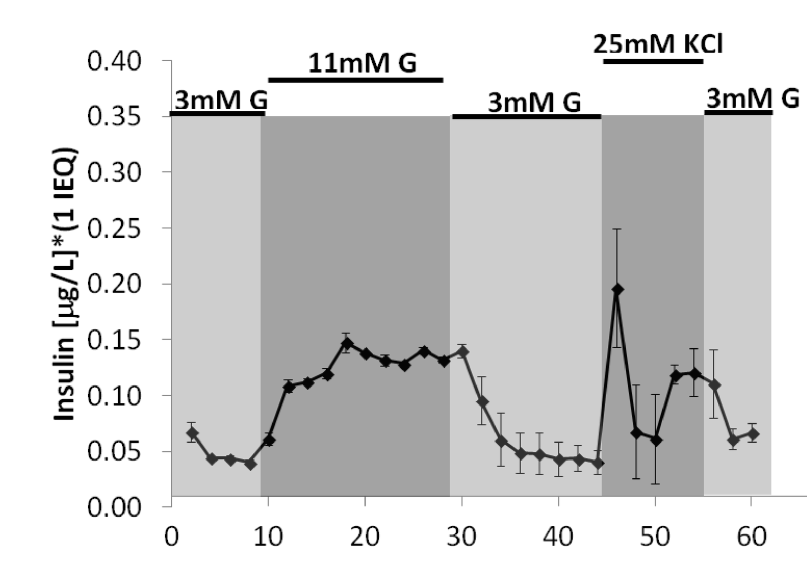
L-D assay of a human islet (FI image)



L-D assay of a rat islet (Confocal)

Perfusion Data

- Temporal insulin secretion of human islets perfused with basal (3mM) and stimulatory glucose (11mM), and KCl solution (25mM)
- Each well can host as little as <10 islets (or up to 100 islets)
- Samples are collected separately from each of the 3 wells of the device (data show average ±SEM)



Acknowledgments

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HIRN
NIH National Institute of Diabetes and Digestive and Kidney Diseases