

Background

Brain organoids are 3D stem cell derived tissue models that replicate human brain structure, used to study neurological disease and drug response. Human brain complexity is poorly represented in *in vivo* and animal models, and as the FDA plans on phasing out certain animal testing, brain organoids are emerging as a critical alternative for preclinical research.

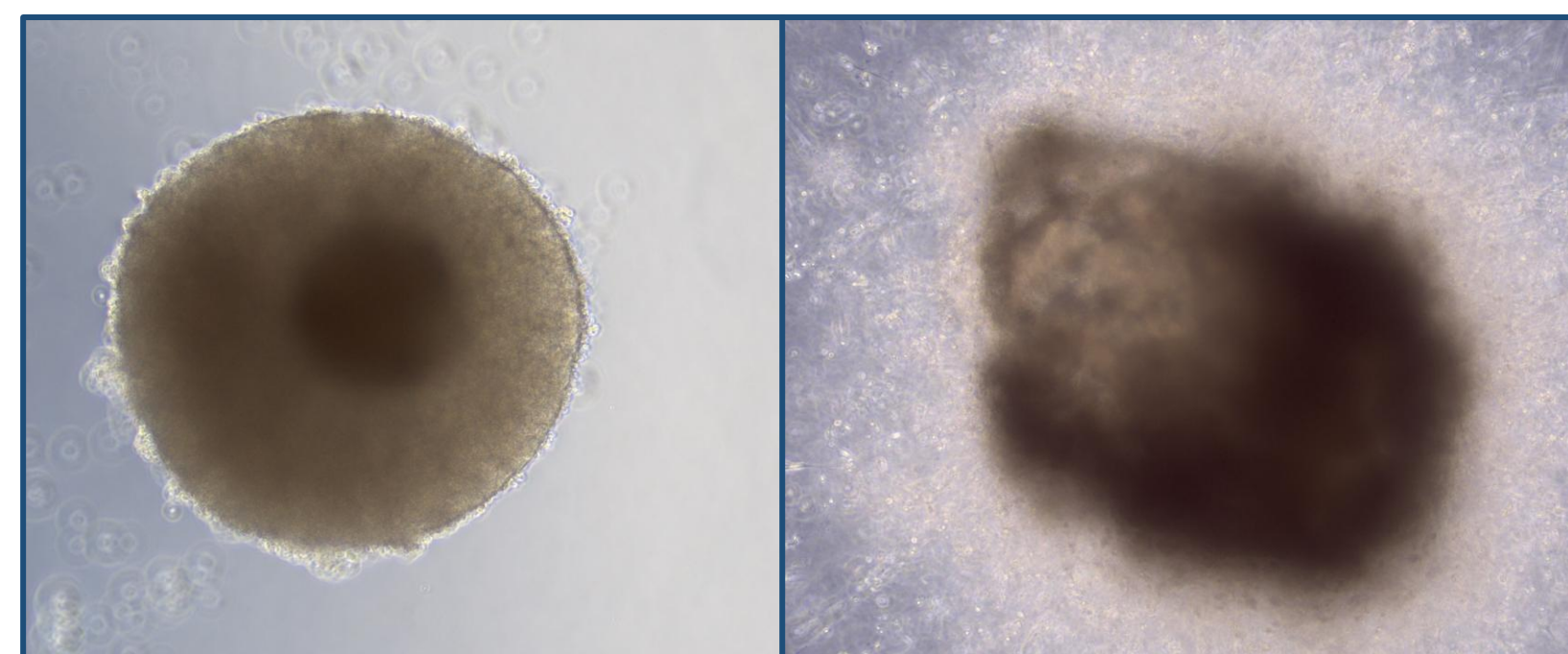


Fig 1. Healthy (left) vs unhealthy (right) human cerebral cortical organoids

Problem: Organoids within a batch vary in shape, size, structural organization, and quality → reduces reliability in experiments. Manual sorting is slow, subjective, and damages samples through repeated handling. Current commercial organoid sorting platforms are very expensive (\$55,000+).

Gap: No accessible/inexpensive solution exists for automated mm-scale organoid quality control.

Objectives

Goals	Target/Approach
Organoid transport	Handle 1-3 mm brain organoids with stable, continuous flow
Hydrodynamic flow focusing	Dual sheath flow to center organoids
Preserve viability during sorting	Maintain $Re < 100$ & shear stress < 0.01 Pa
Sterile, hands-off sorting	Binary pass/fail actuated sorting
Real-time organoid classification	Camera image → automated organoid classification algorithm
Biocompatibility	All fluid contact surfaces use PDMS, glass, & medical-grade tubing
Accessibility	Compatible for standard lab workflows

Design Solution

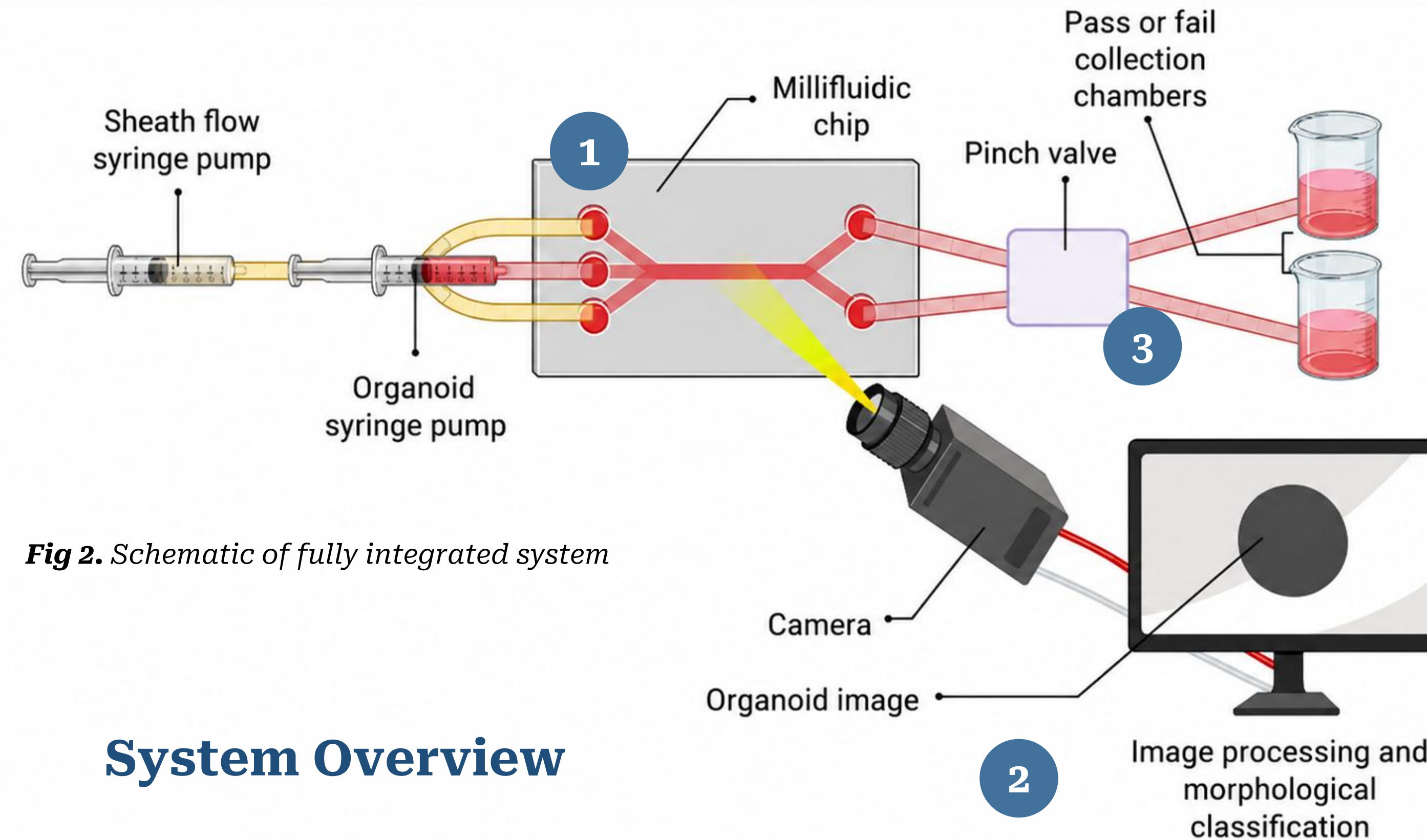


Fig 2. Schematic of fully integrated system

System Overview

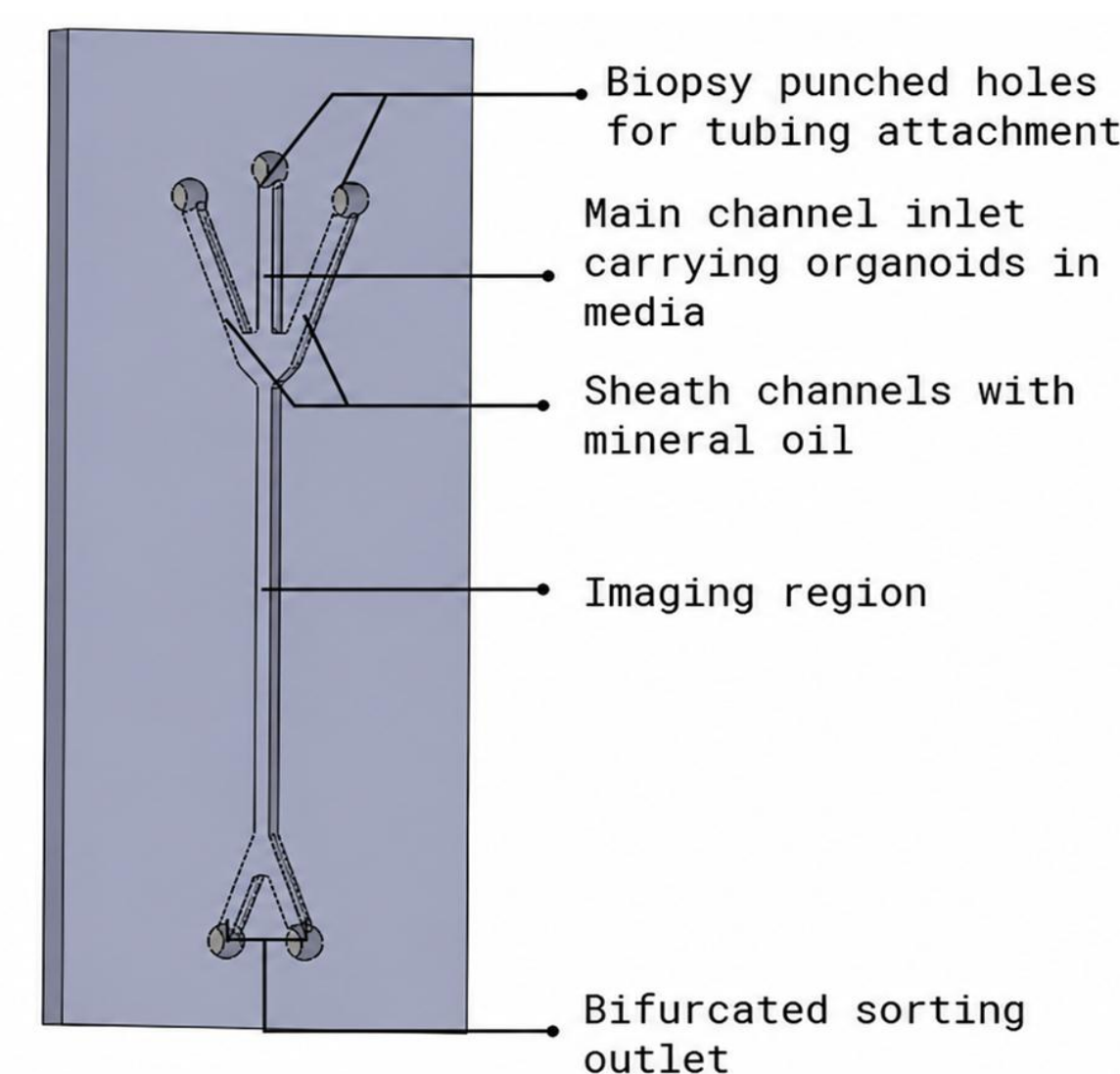


Fig 3. Channel layout of millifluidic chip

1 Millifluidic Chip

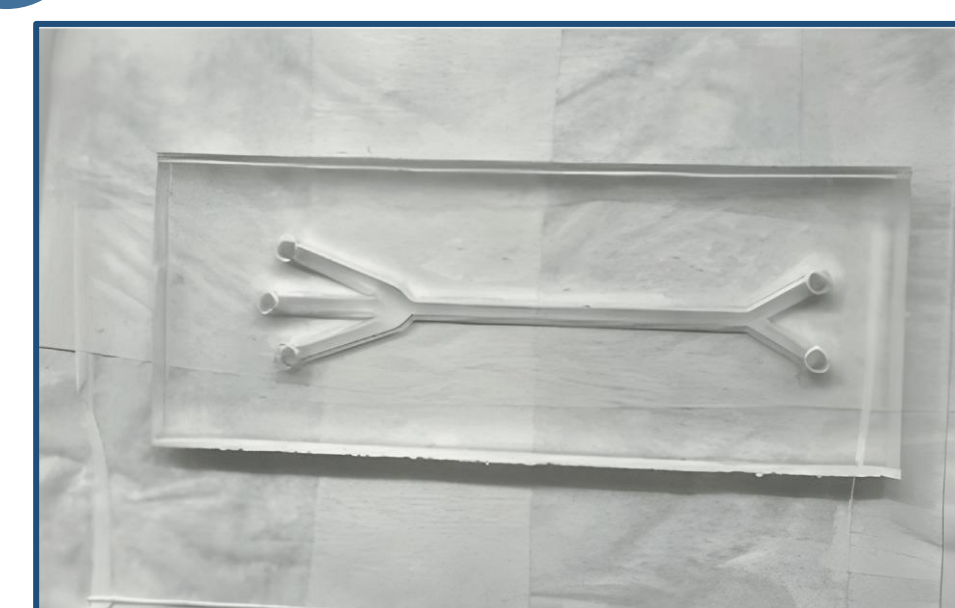


Fig 4. PDMS chip bonded to glass substrate

Channel width	3.5 mm
Chip dimensions	60 x 150 mm
Material	PDMS/glass
Mold Fabrication	Resin print

2 Image Classification

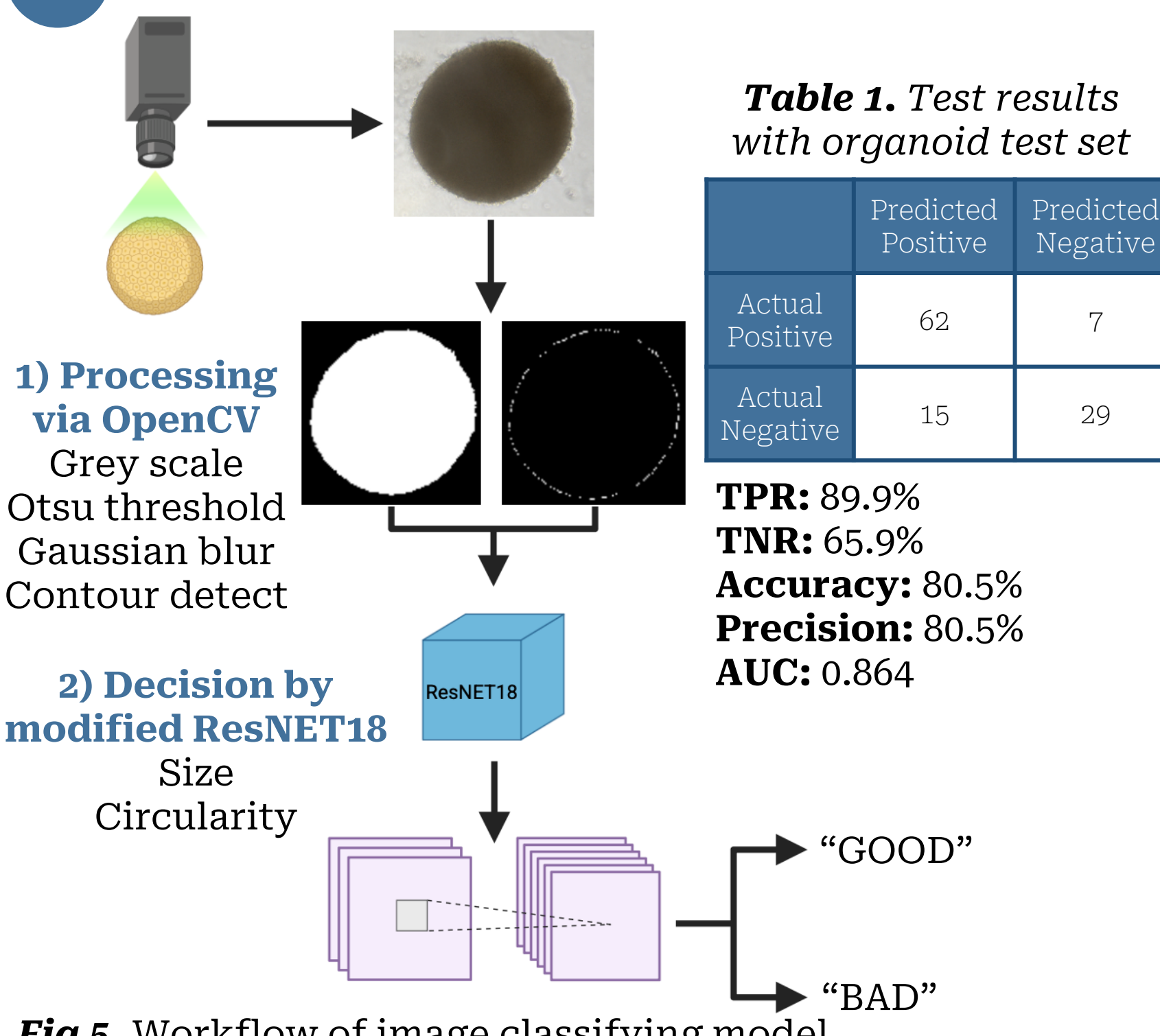


Fig 5. Workflow of image classifying model

3 Pinch Valve Sorting

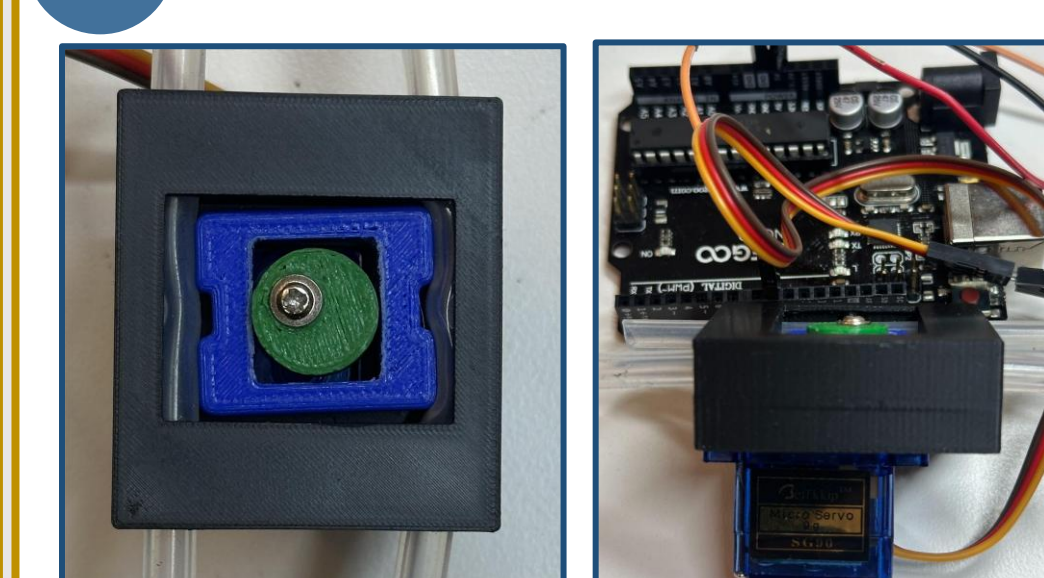


Fig 6. Pinch valve prototype

- 1) **Arduino Uno** receives "GOOD" or "BAD" Classification output
- 2) **Servo-driven eccentric cam in pinch valve** compresses one outlet tube while leaving the other open. **Switching time: ~1 s** from classification decision to flow redirection.
- 3) **Selective tube pinching** redirects flow toward the correct collection chamber.

Subsystem Testing & Validation

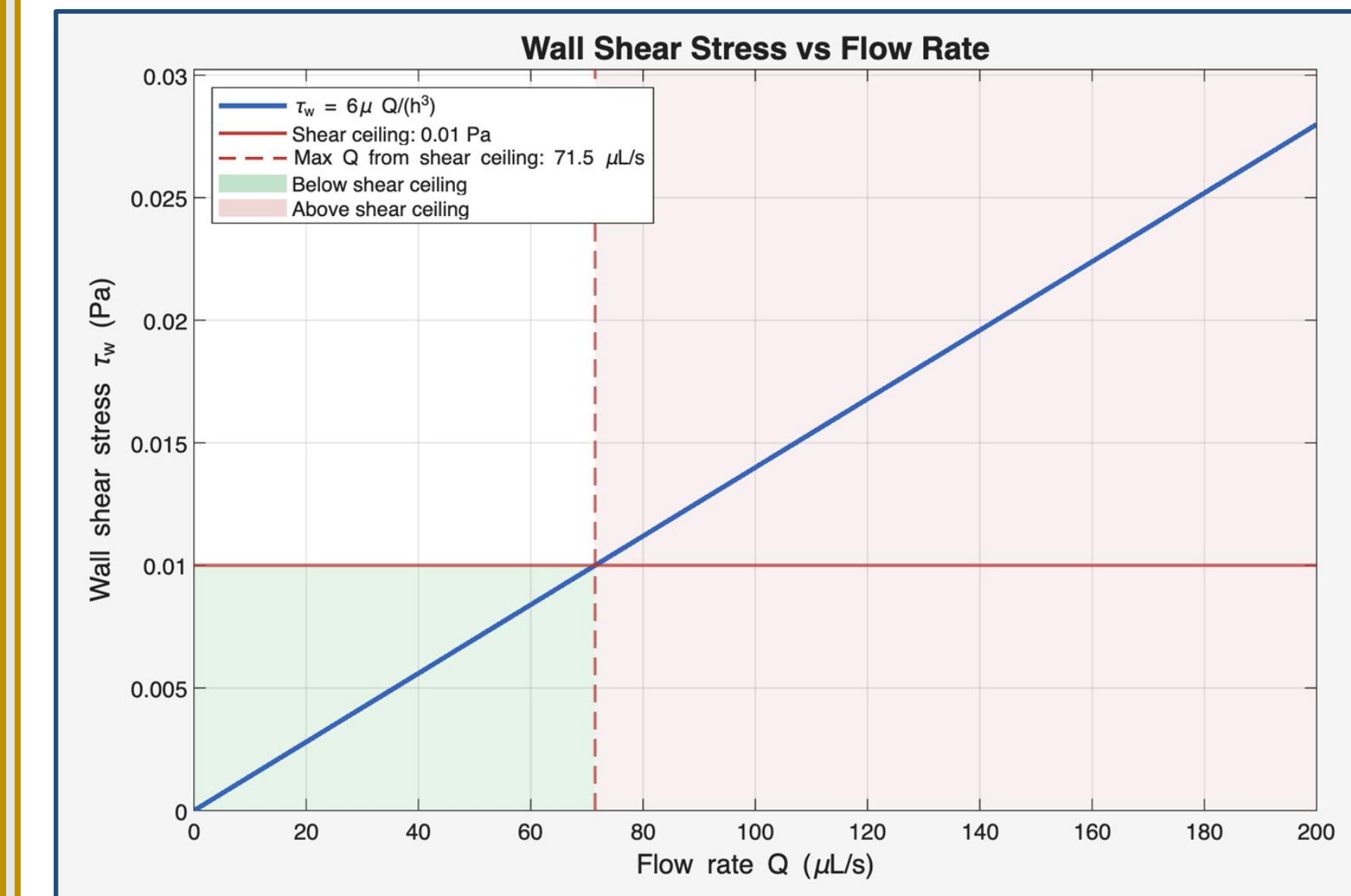


Fig 7. Wall shear stress vs flow rate in 3.5 by 65 mm imaging channel.

- At operating conditions ($Q=60 \mu\text{L/s}$) shear stress remains below the 0.01 Pa viability threshold with 5 sec imaging dwell time, confirming viability-safe conditions.

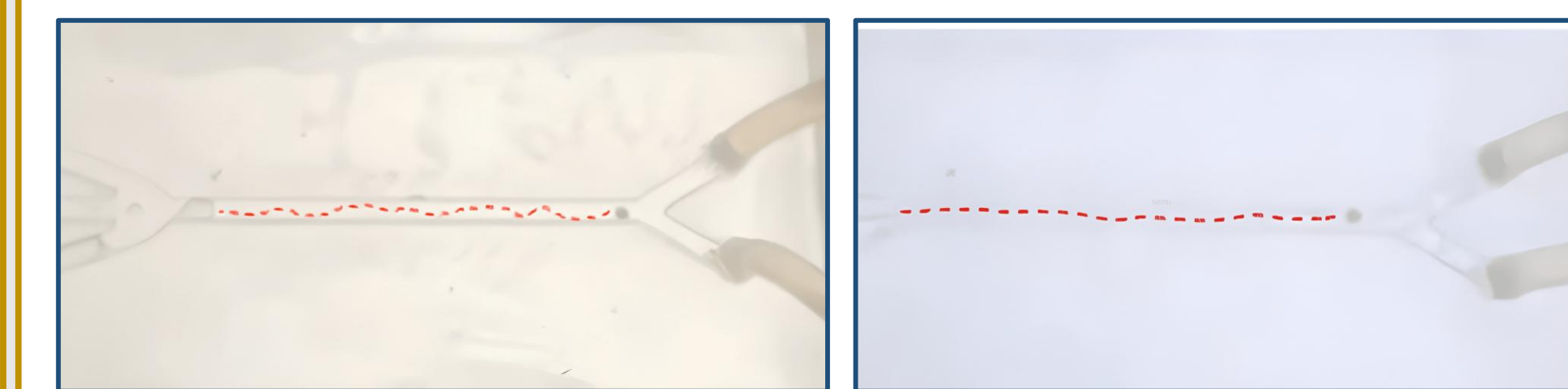


Fig 8. Bead phantom paths without sheath flow (left) vs. with mineral oil sheath flow (right) demonstrating improved centering of bead phantoms.

- Sheath flows prevent drifting towards the walls & keep beads in the middle of channel.
- Improves imaging consistency for classification.



Fig 9. Real-time raw camera image of bead phantom in channel (left) & image processing output of binary masks (right), which was accurately classified as "good".

- Classification algorithm is able to make an instant and accurate decision from raw image inputs.

Conclusions

Currently, our platform has achieved:

- Stable organoid transport and hydrodynamic focusing with viable flow conditions
- Hands-off sorting system incorporating fluidics, organoid classification algorithm, and pinch valve actuation that redirects flow
- Biocompatible, sterile, accessible materials and workflow with total cost $< \$200$

Future testing with complete integrated system:

- Sorting accuracy (% correctly classified and sorted to correct outlet)
- Organoid viability post-sorting
- Repeated valve timing consistency with classification algorithm input
- Throughput

Future scope:

- Integrate classification for advanced morphological features (e.g. surface texture, number of rosettes, internal organization, symmetry)
- Expand sorting criteria to support multi-class sorting beyond binary pass/fail
- Scale throughput for high-volume screening applications (e.g. drug testing)

Acknowledgements & References

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