

Exceptional service in the national interest



Engineered surfaces and microfluidic structures for controlling cell morphology, migration, and cell-cell interactions

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Sandia National Laboratories, Biosensors & Nanomaterials Department



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- Biological computation
- Neural engineering
 - Background
 - Neuron polarization
 - Microfluidic compartmentalization
 - Embedded microfluidic structures
 - Porous nanostructures
- Conclusions and Future Work

Biological neural networks have substantial computational capabilities

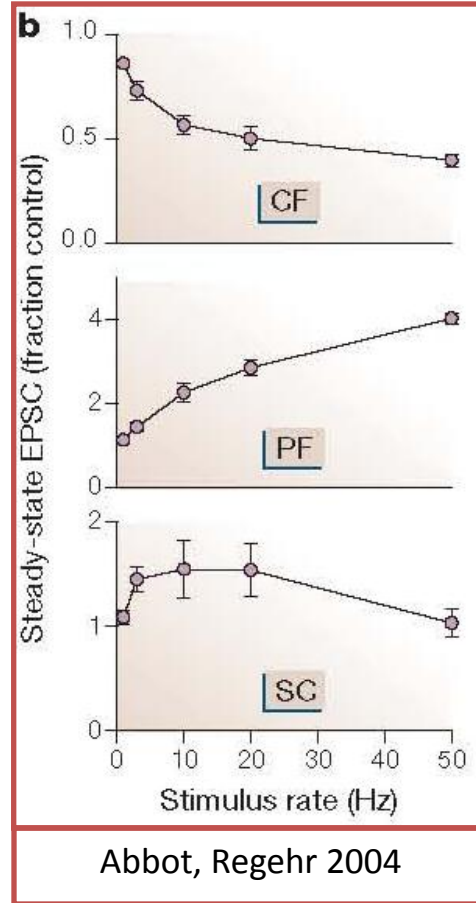
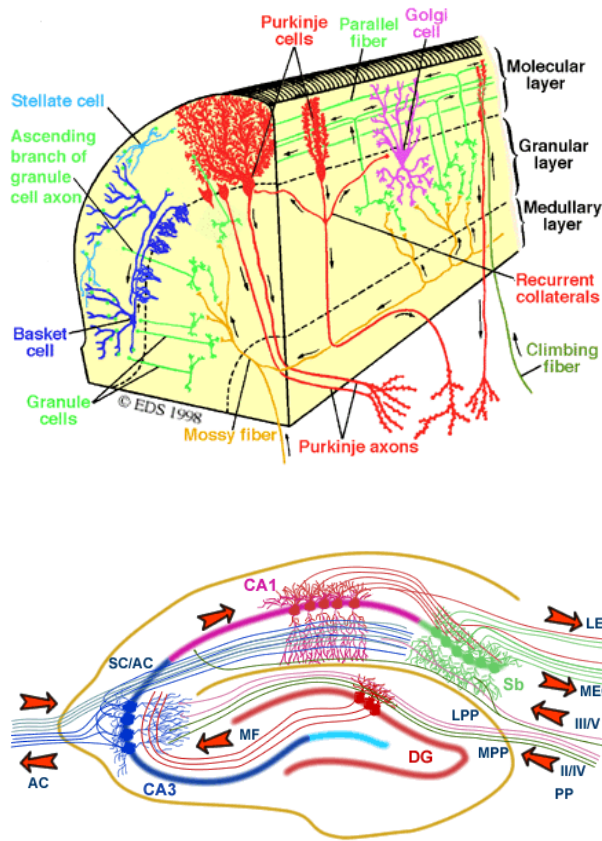


Cray XT5 Jaguar (ORNL), hex-core
~ 2×10^{15} flops/s, ~**10 MW**, **room-sized**
~**100 Gbits/in²**
access-limited (processor vs. memory)

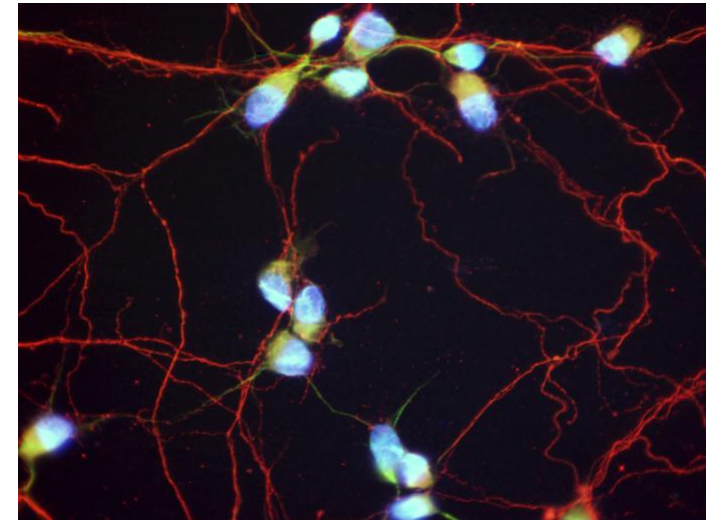


- Brain, 10^{11} neurons, 10^{15} glial cells
- $>10^{16}$ flops/s, **20W**, **1200 cm³**
- ~**large memory storage capacity**
- **fast memory access**

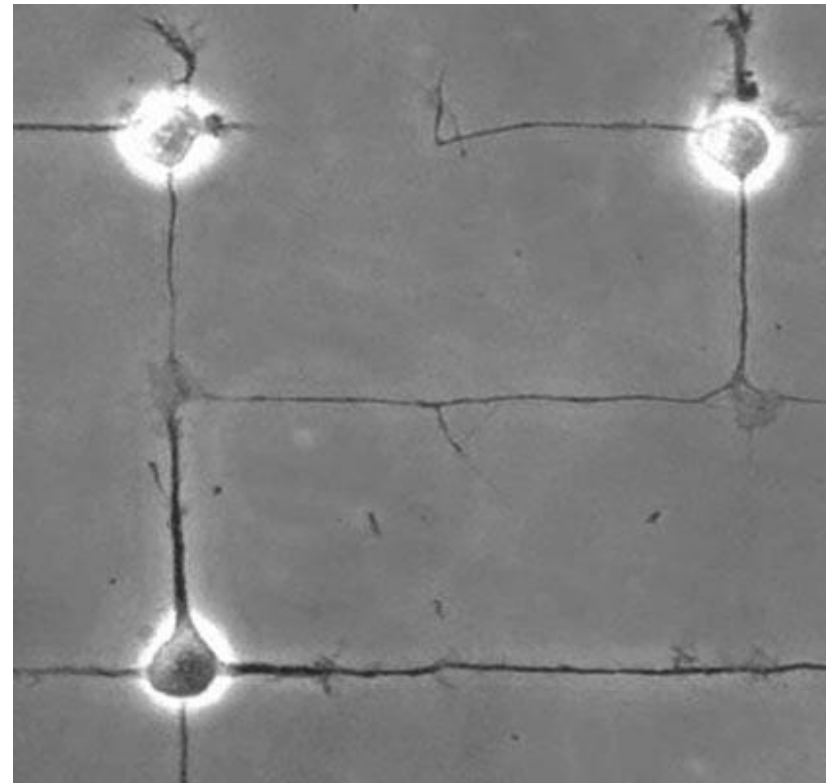
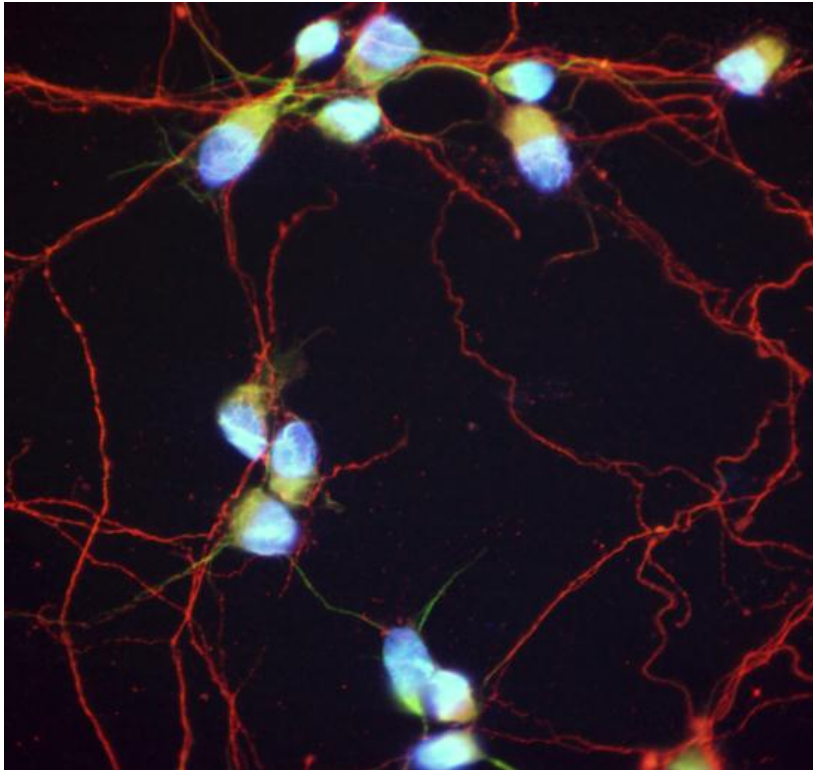
Influence of network architecture and stimulus frequency on network function



Re-engineer dissociated neurons into networks to examine the effect of architecture on function



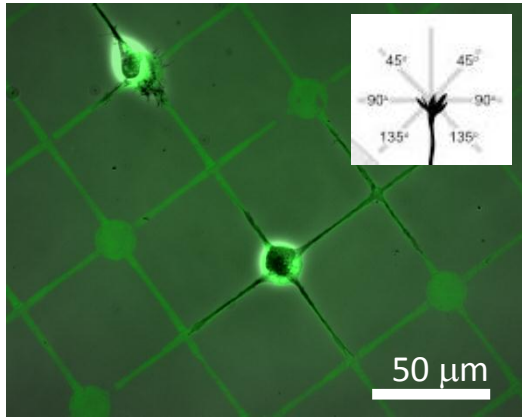
Utilize microfabrication techniques to engineer living neural networks



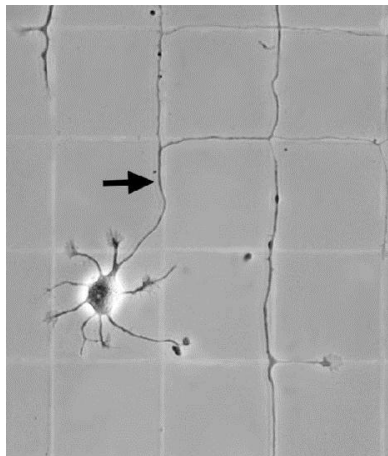
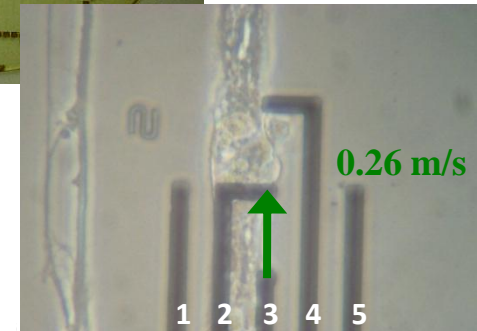
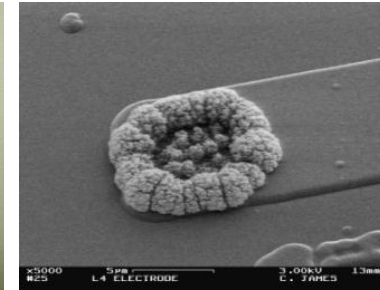
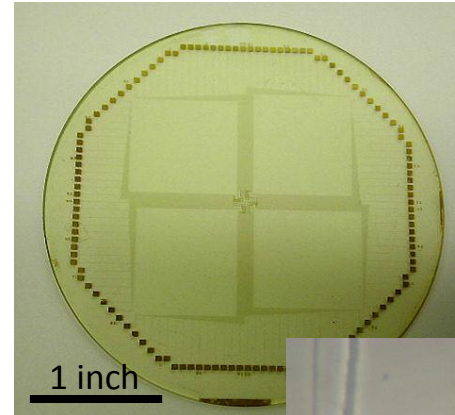
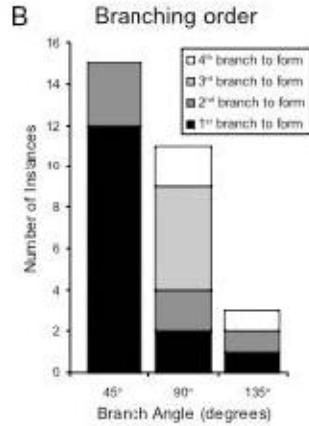
position cells, control polarization, and direct synaptic connections

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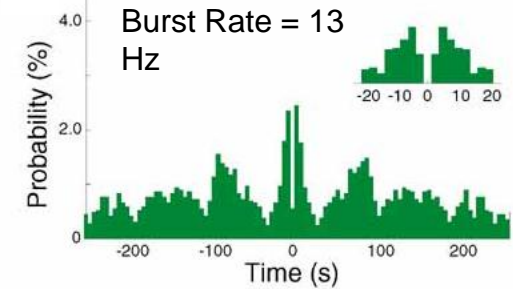
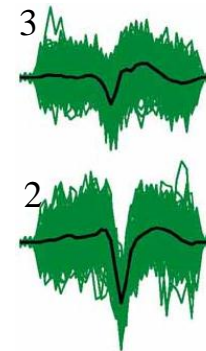
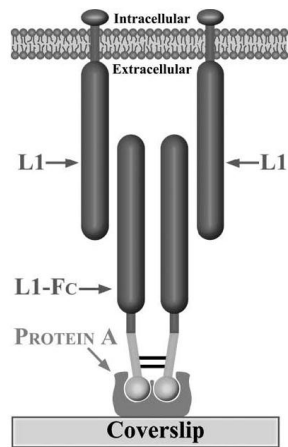
Engineered neural networks using microfabrication techniques



Withers et al., *J Neurobio* 2006, 66, 1183



Oliva et al., *Neurochem Res* 2003, 28, 1639



James et al., *IEEE TBME* 2000/2004, 47/51, 17/1640

Microsystems and Engineering Science Applications (MESA)

Highly collaborative research

- microsystems engineering (1700), biological science (8600), nanomaterials synthesis (1800), cognitive science (1400)



As an accredited Department of Defense Trusted Design Center, Sandia provides Trusted ASIC Design Services for both radiation-hardened and non-radiation-hardened foundries (350nm, 180nm, 130nm, 90nm), including in-house, IBM, National Semiconductor and other Trusted Foundries.

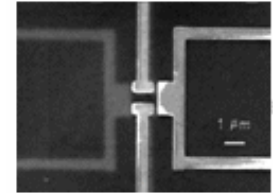
Center for Integrated Nanotechnologies (CINT)

US Department of Energy Office of Science User Facility and Nanoscale Science Research Center

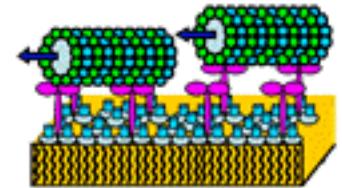
- Sandia National Laboratories (96K ft²)
- Los Alamos National Laboratories (36K ft²)



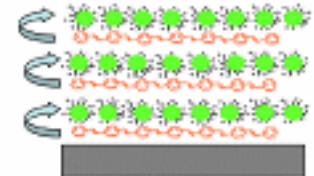
Nanoscale Electronics
and Mechanics



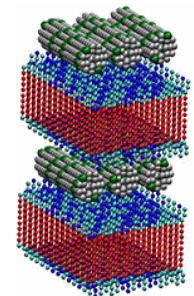
Soft, Biological and
Composite
Nanomaterials



Nanophotonics and
Optical
Nanomaterials



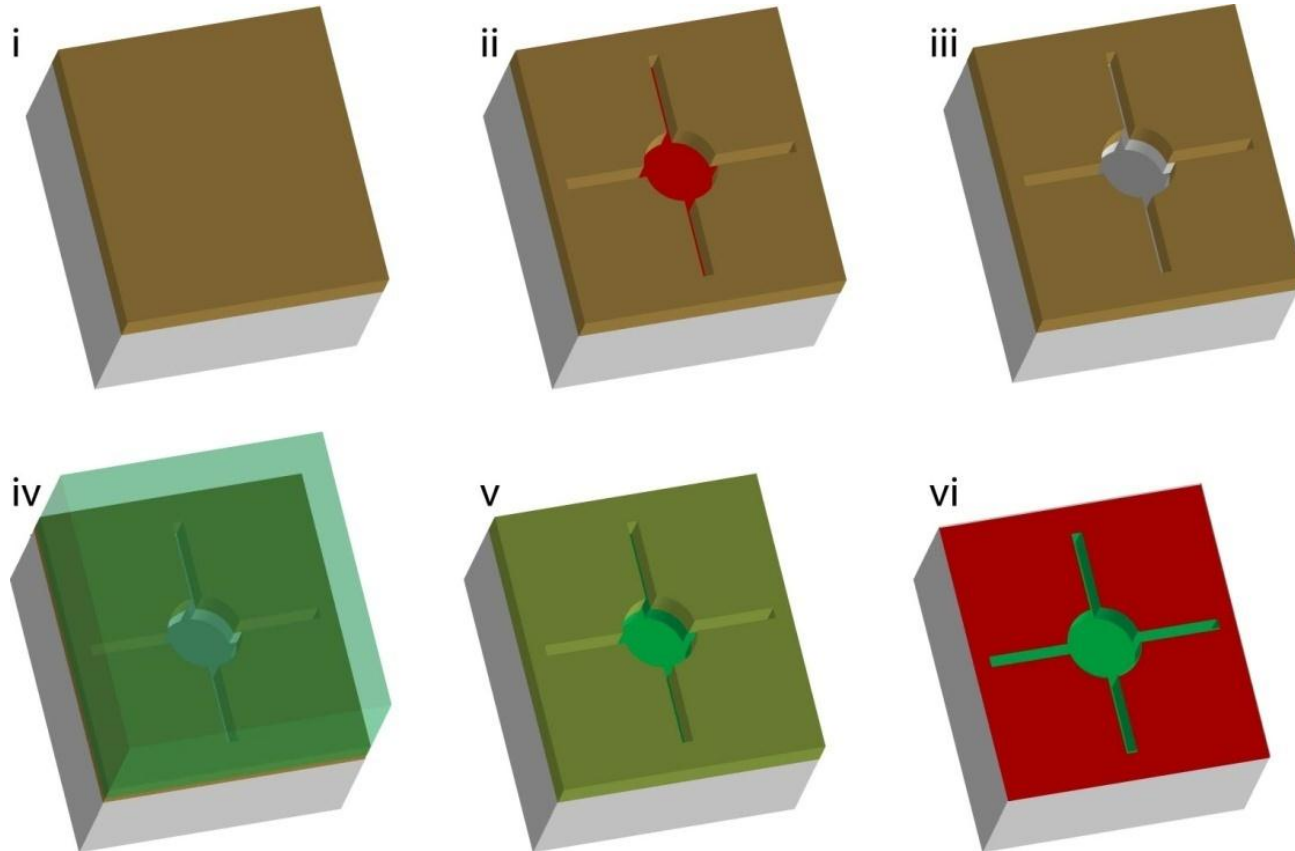
Theory and
Simulation of
Nanoscale
Phenomena



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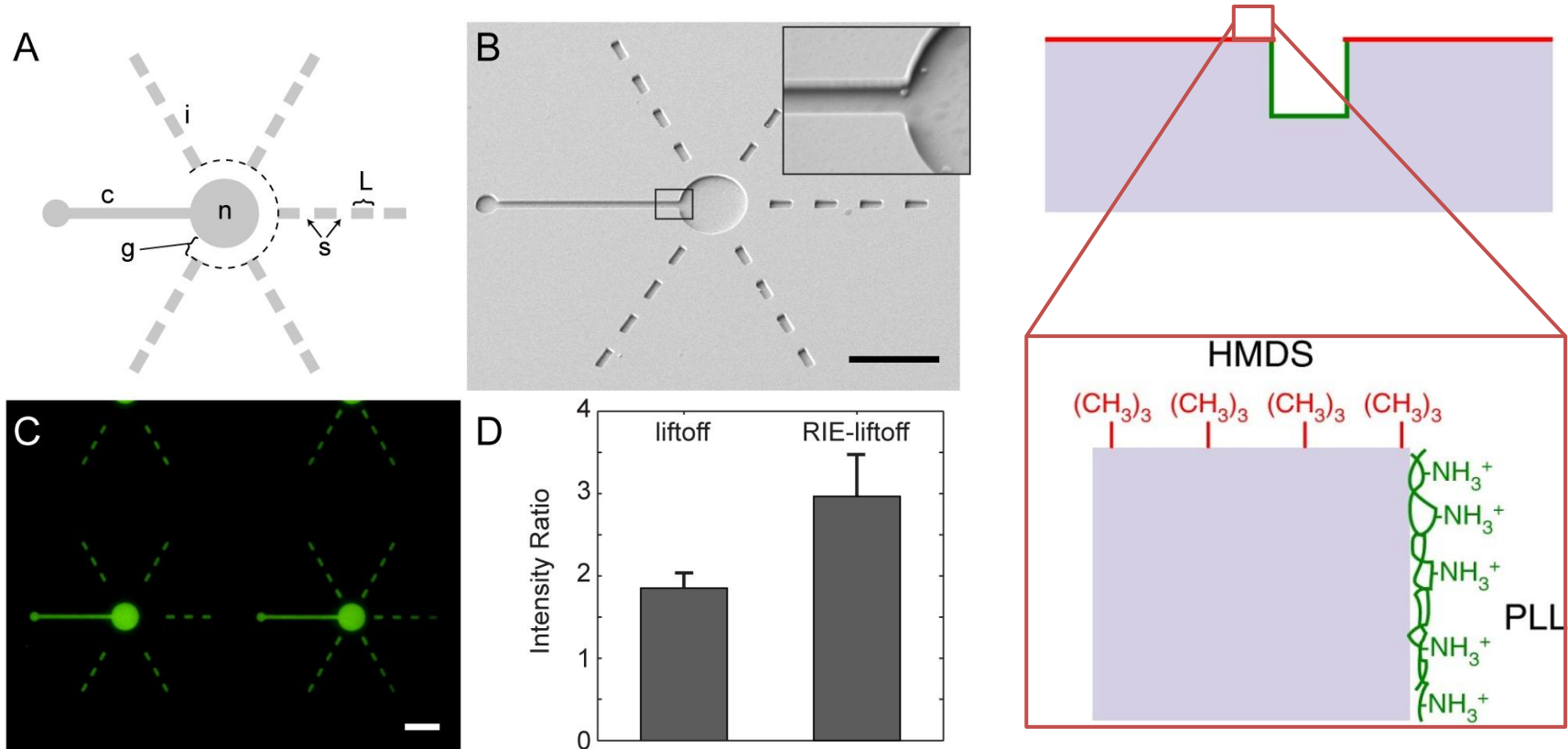
Topographical & chemical cues for network engineering

- Photolithographic techniques to create bi-functional surfaces



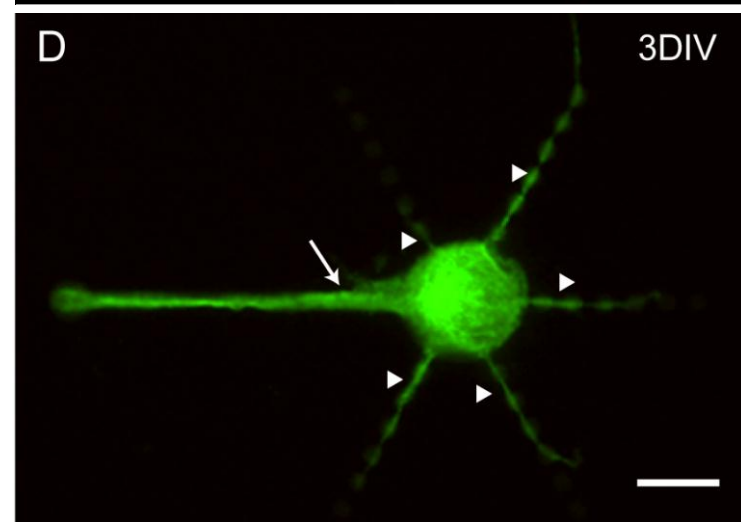
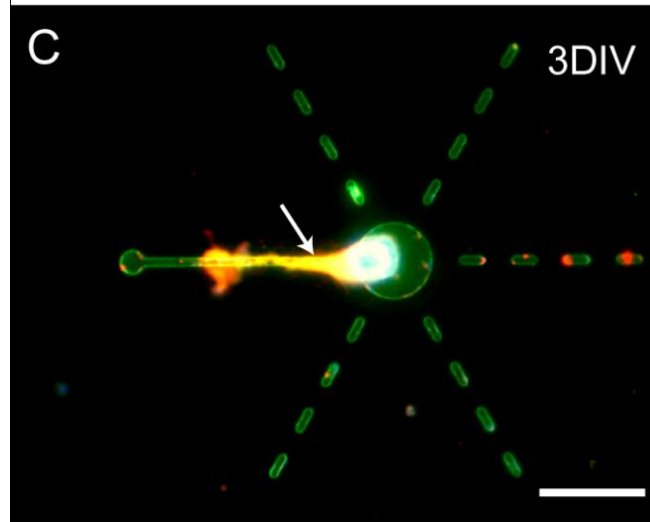
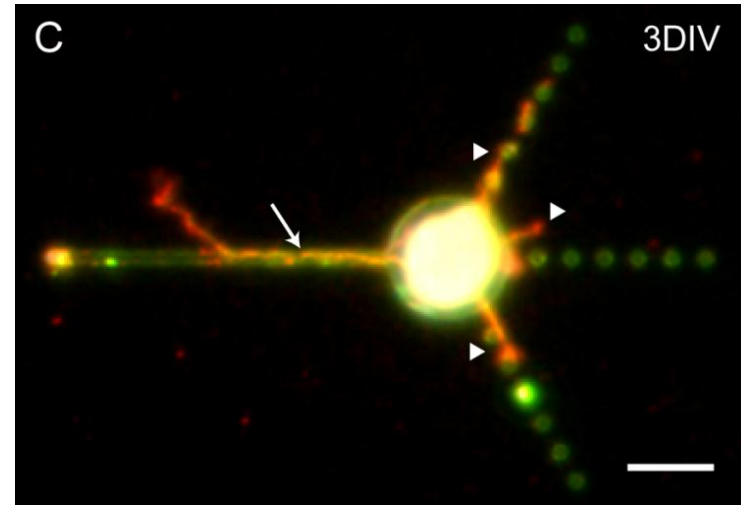
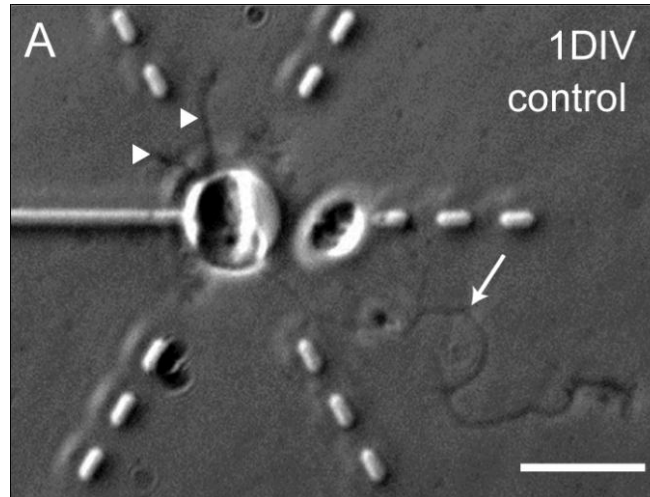
Bi-functional surfaces for promoting cell adherence and repulsion

- Individual neuron guidance cues consisting of adhesive (PLL) and repellent molecules (HMDS) embedded into a glass substrate



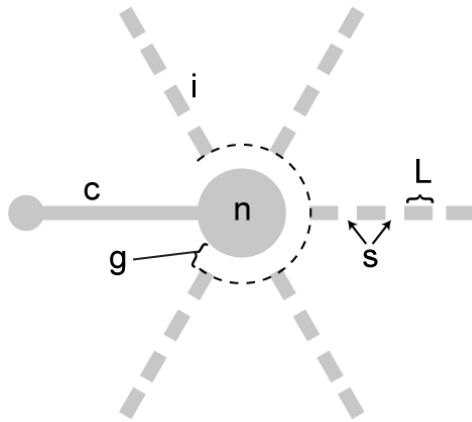
C.D. James et al., *Biomaterials* 2011, 32, 8860

Guidance cue geometry & neuron polarization

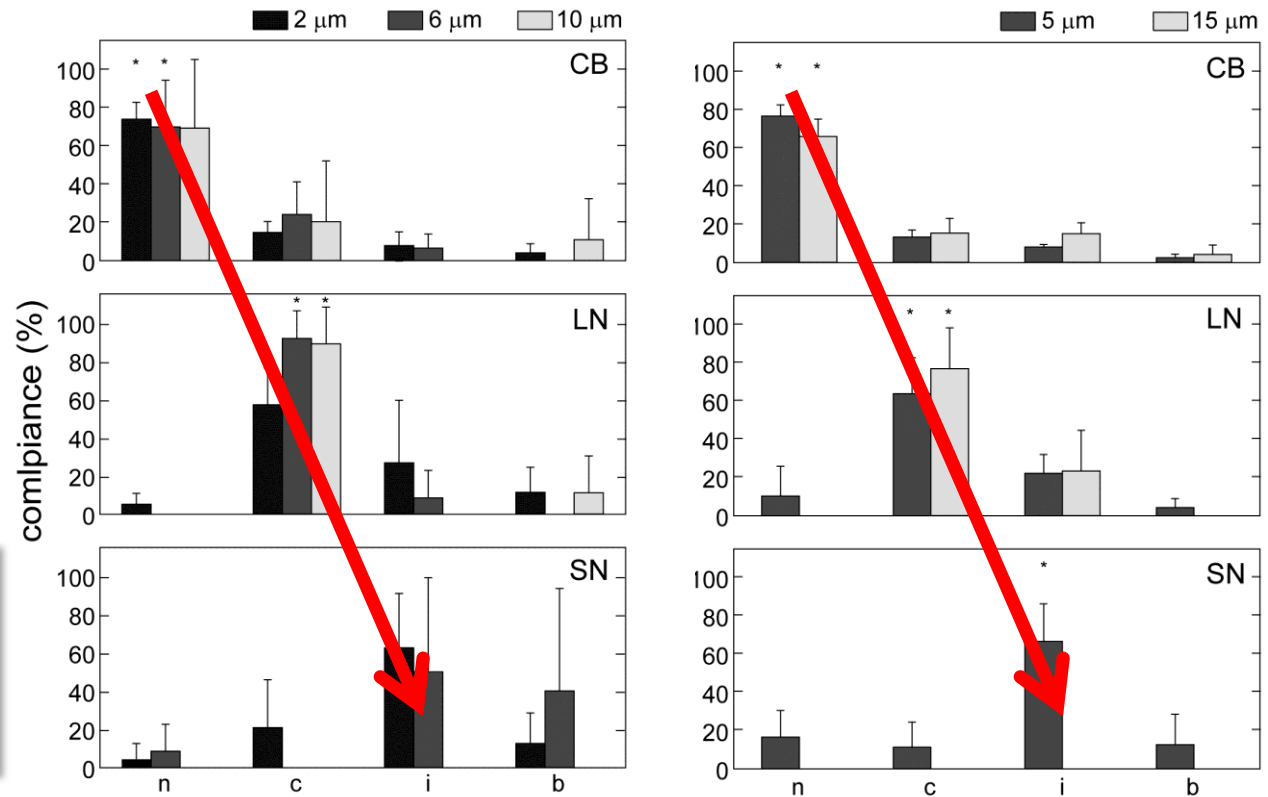


Specific guidance of cell attachment and neurite outgrowth

Cell bodies attach to nodes (n), long neurites extend along continuous lines (c), short neurites extend along interrupted lines (i)



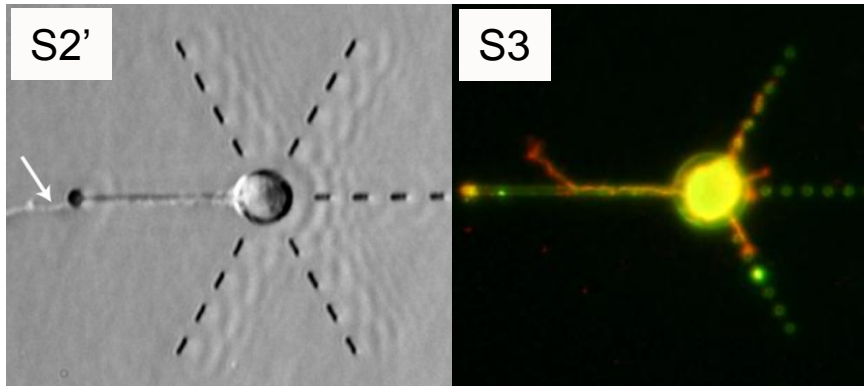
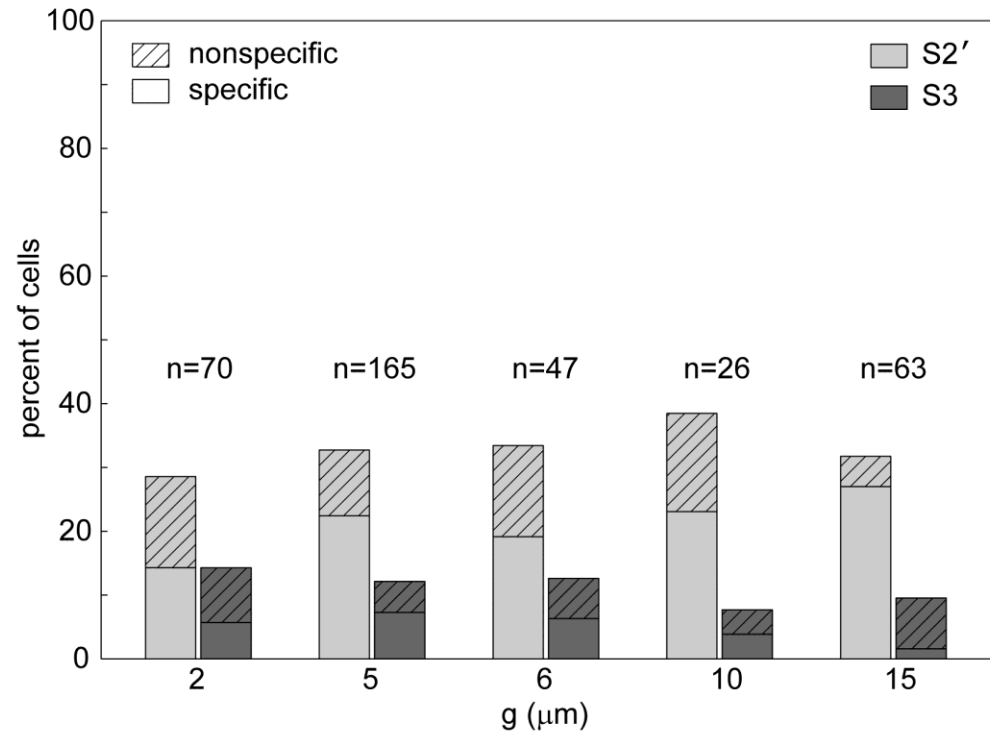
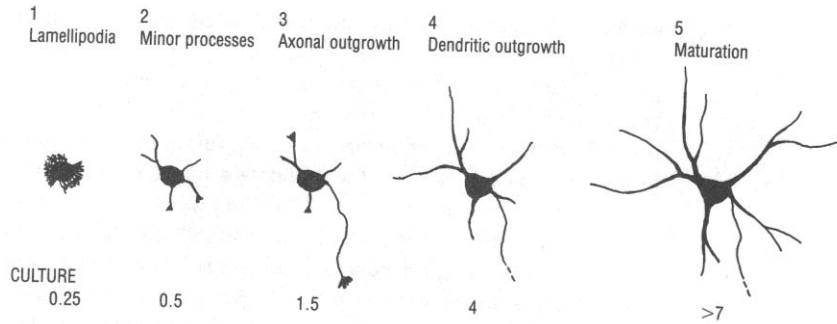
-small g provides no growth differential
 - large g prevents dendrite formation



C.D. James et al., *Biomaterials* 2011, 32, 8860

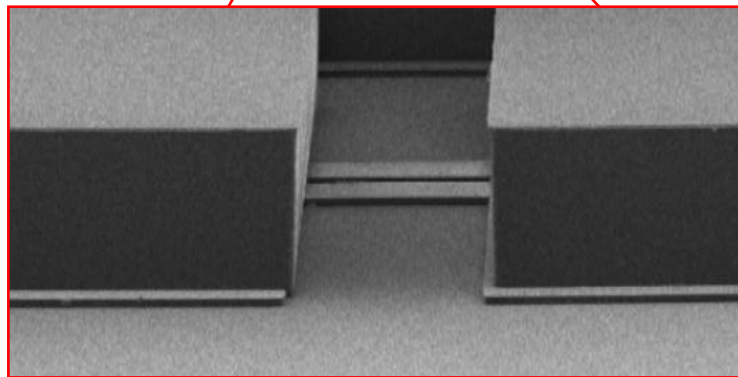
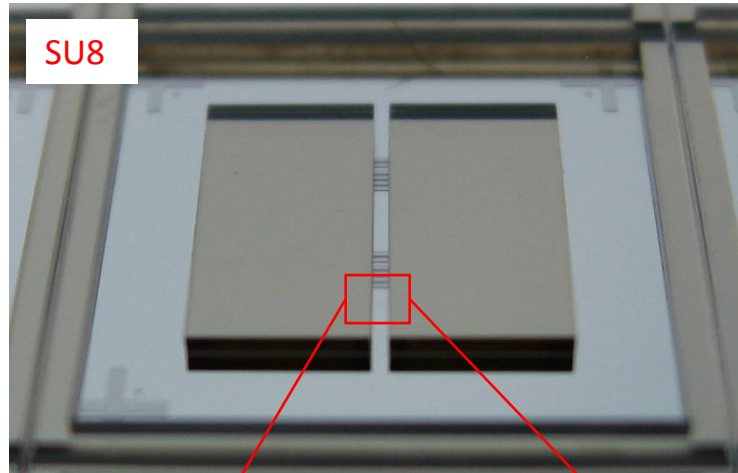
Guidance of neurons towards cytoarchitectural maturity

Banker and Goslin (1998):

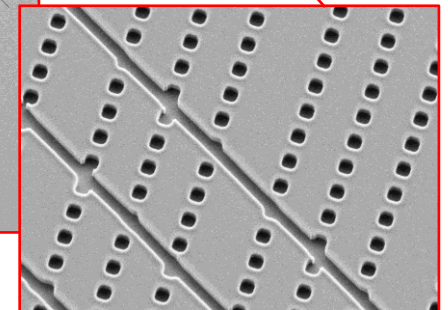
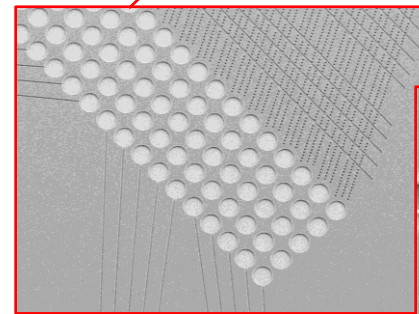
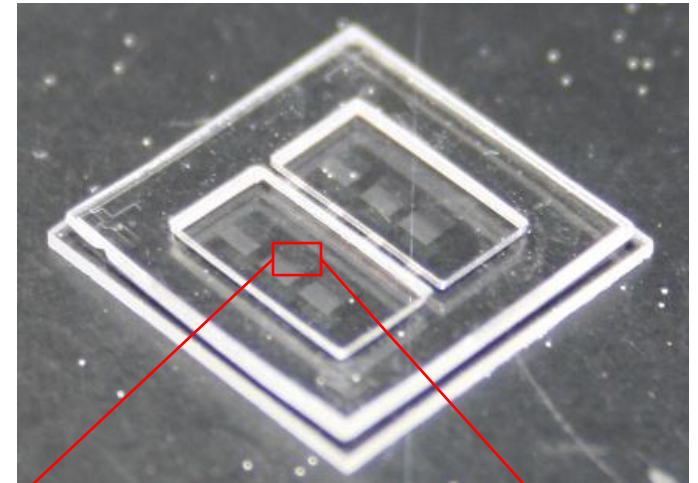


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Engineered networks using microfluidic compartmentalization and surface guidance cues

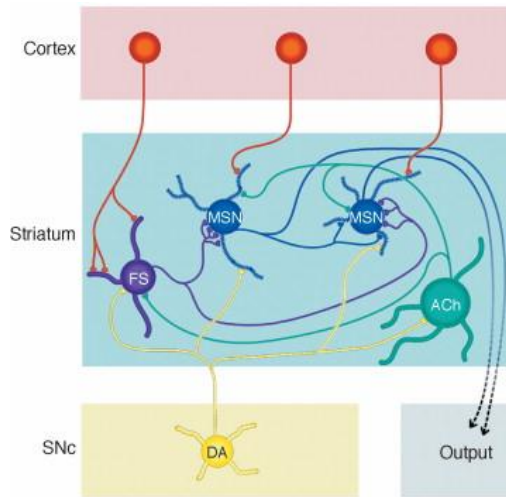


SU8 master for creating microfluidic compartments for engineered cell networks

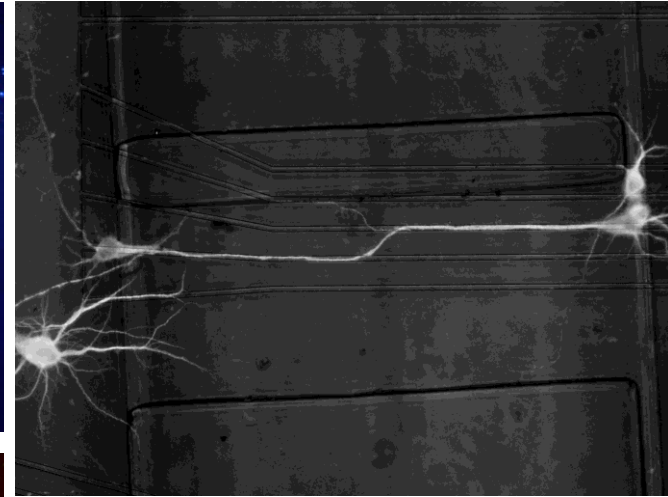
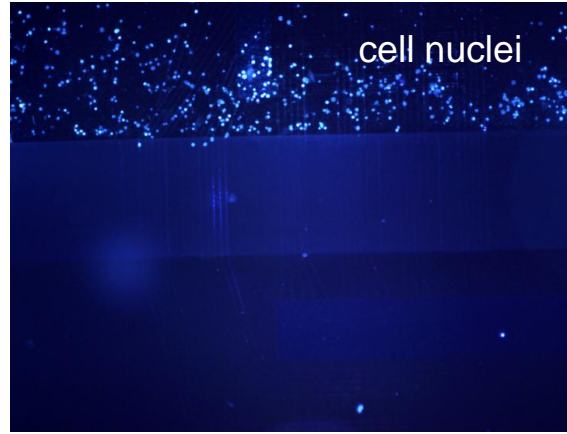


PDMS chambers bonded to glass substrates with guidance cues

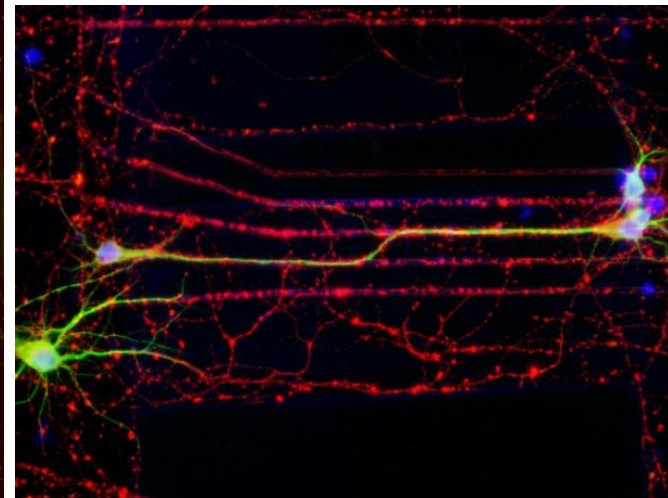
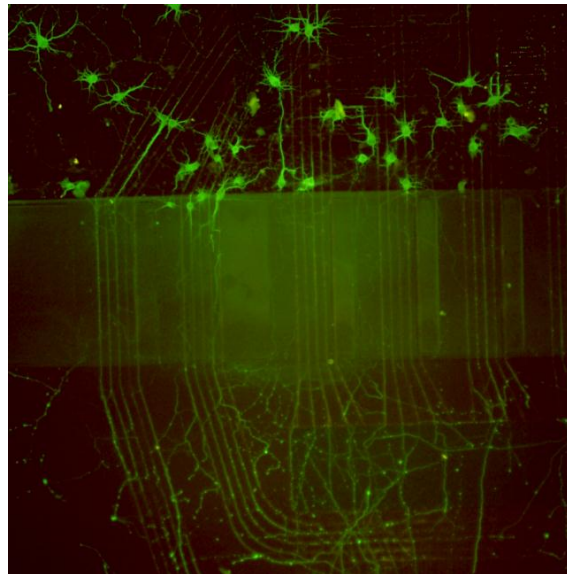
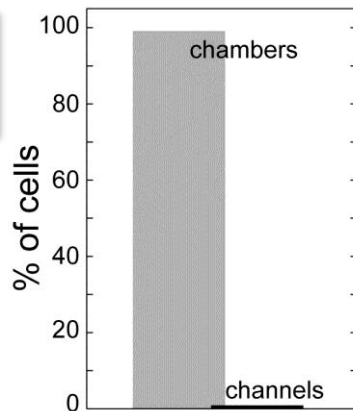
Compartmentalization of multiple neuronal cell types



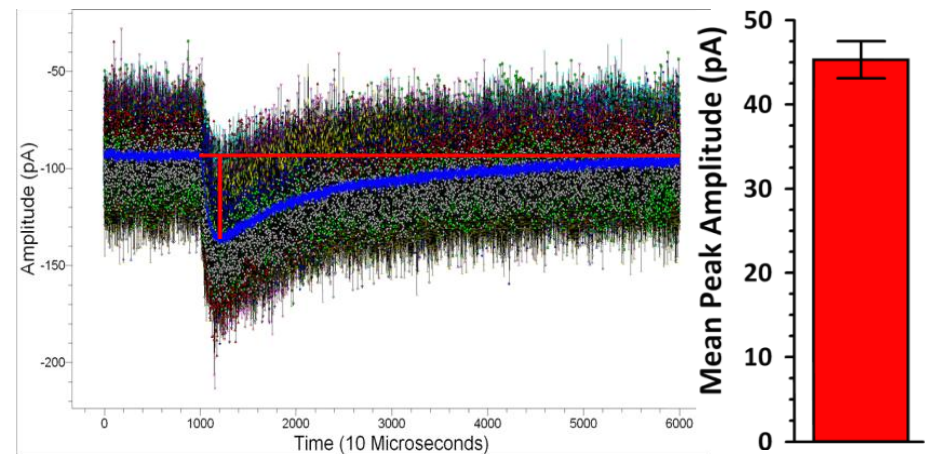
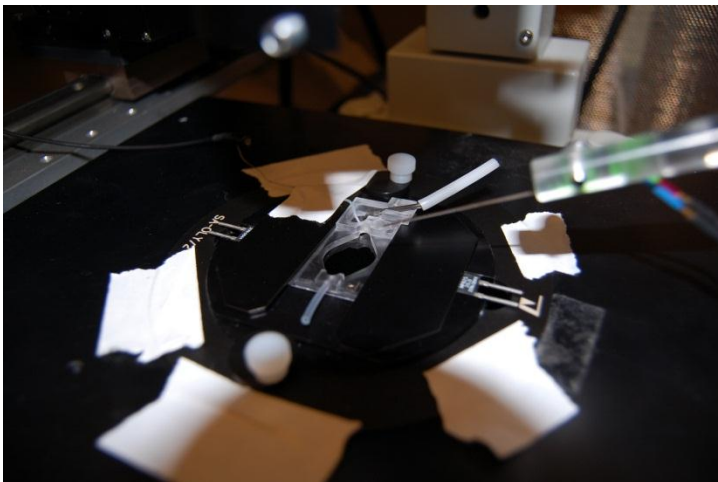
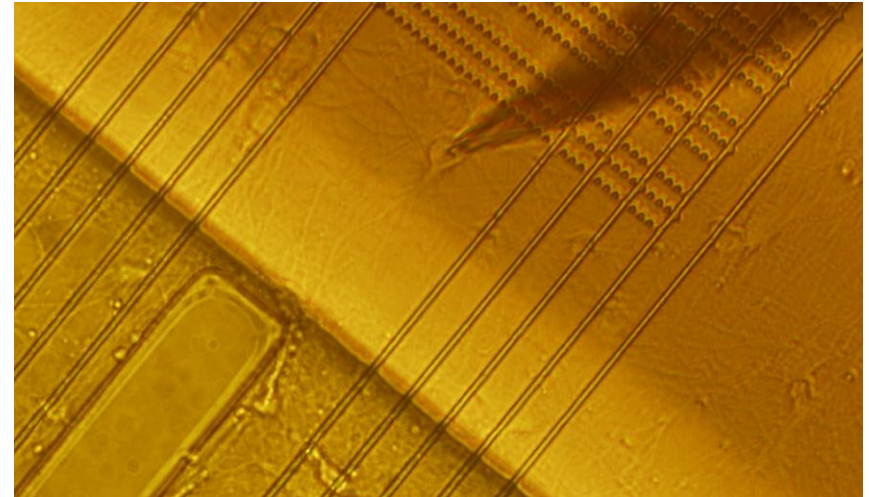
Tepper et al., *Curr Opin Neurobio*, 2004



10 μm
channels

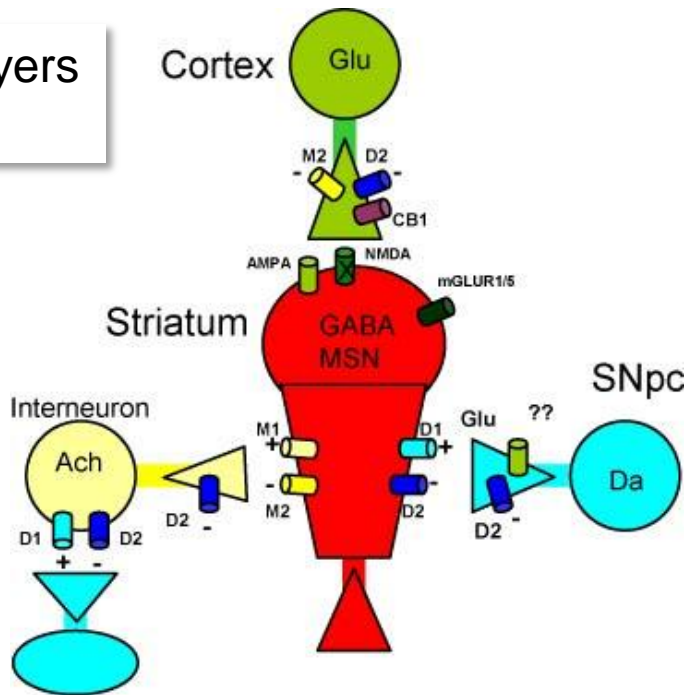


Electrophysiological analysis of engineered corticostriatal networks

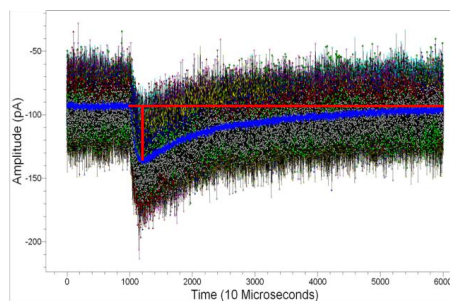
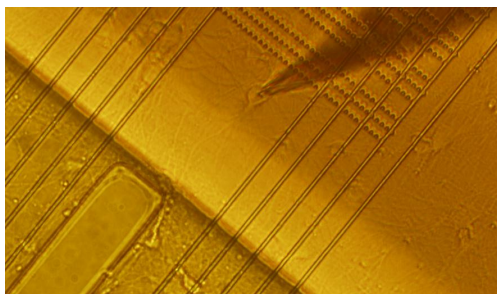
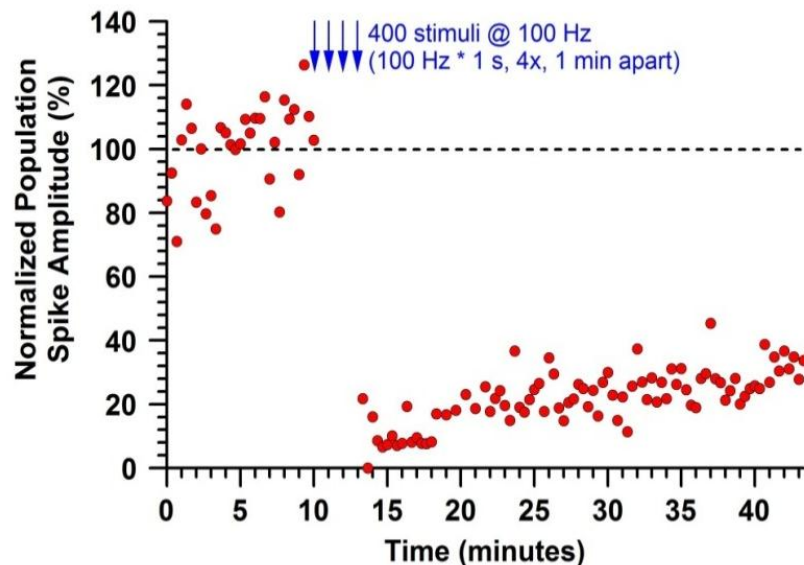
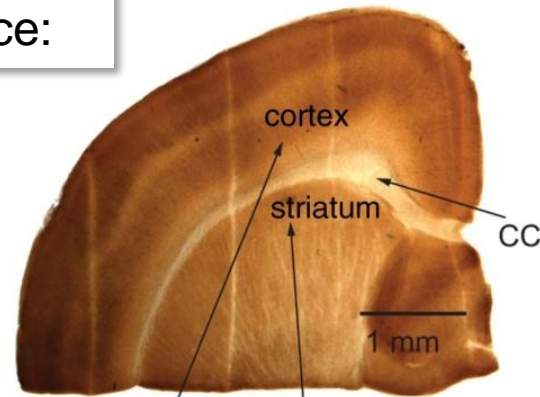


Engineered CSNs to identify important *in vivo* phenomena

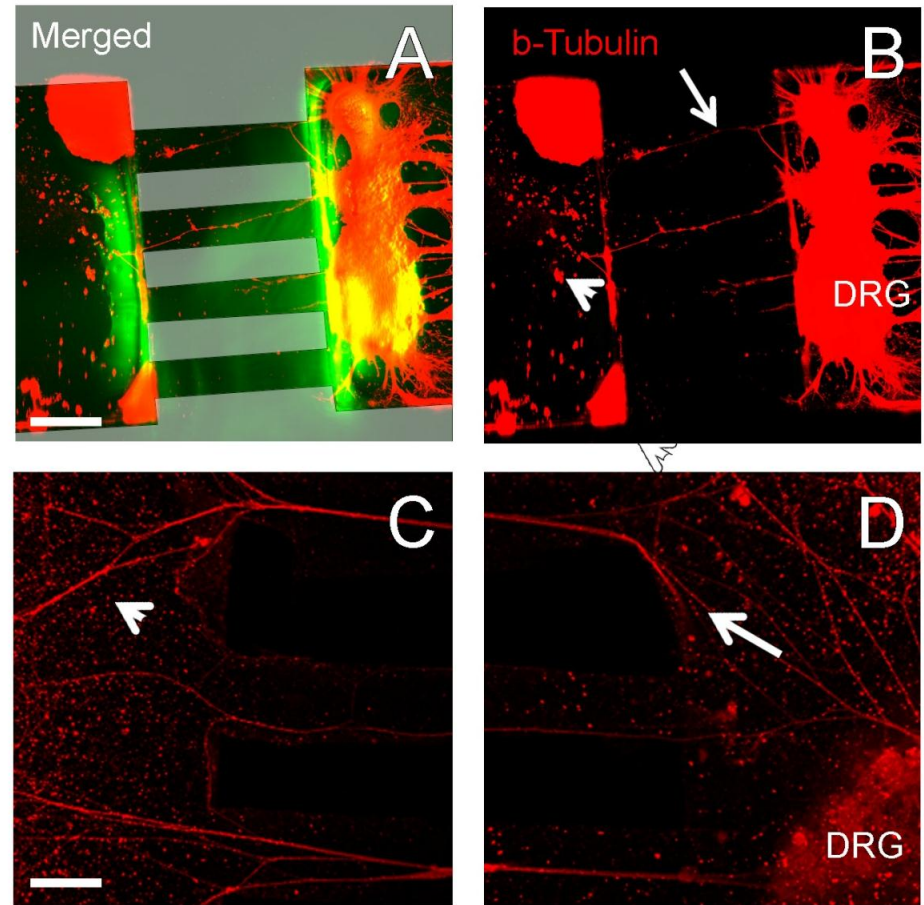
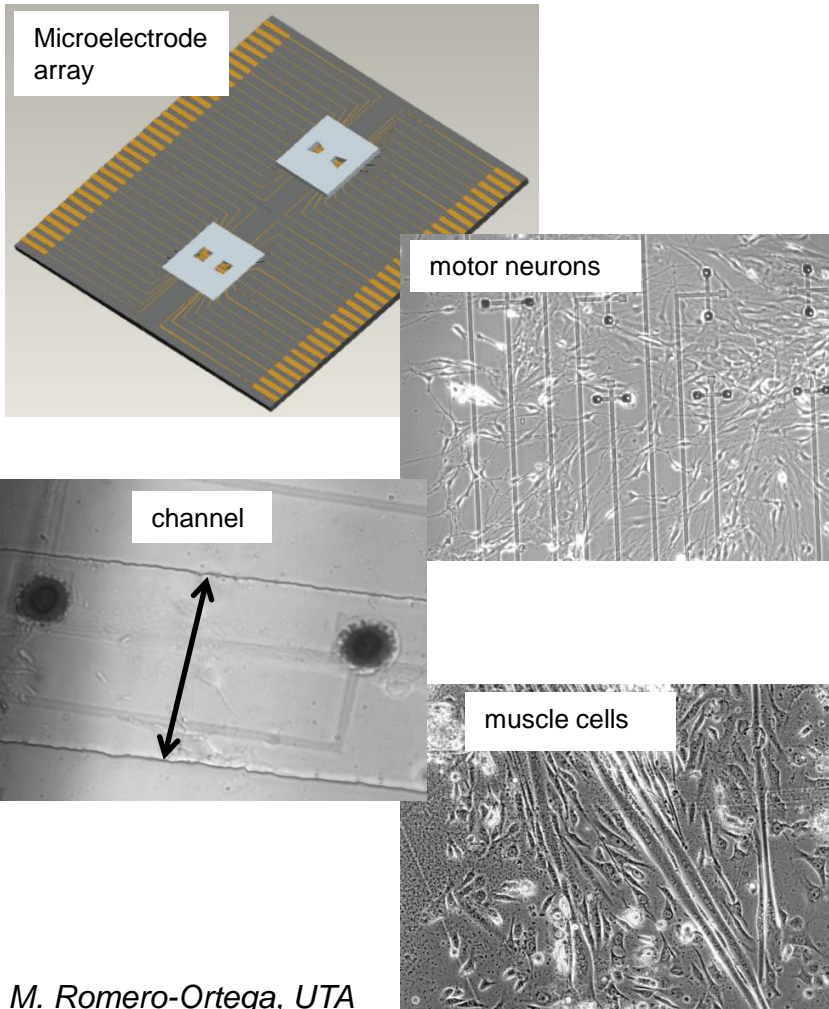
Major players in CSNs:



Brain slice:

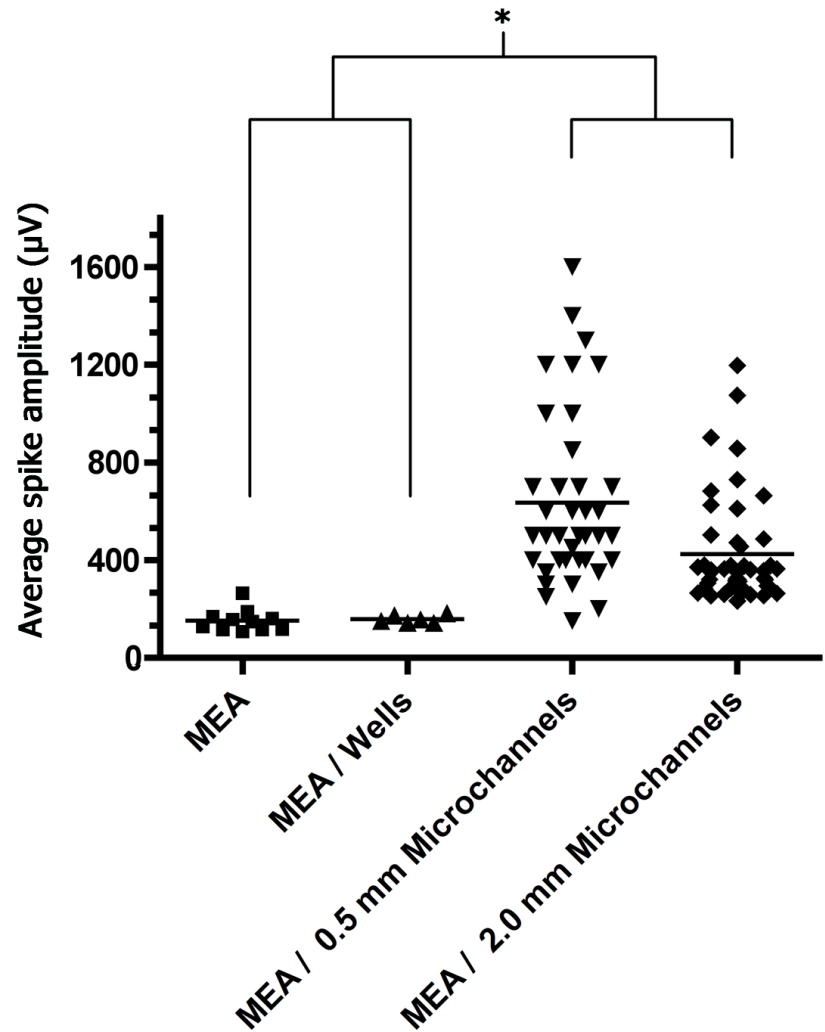
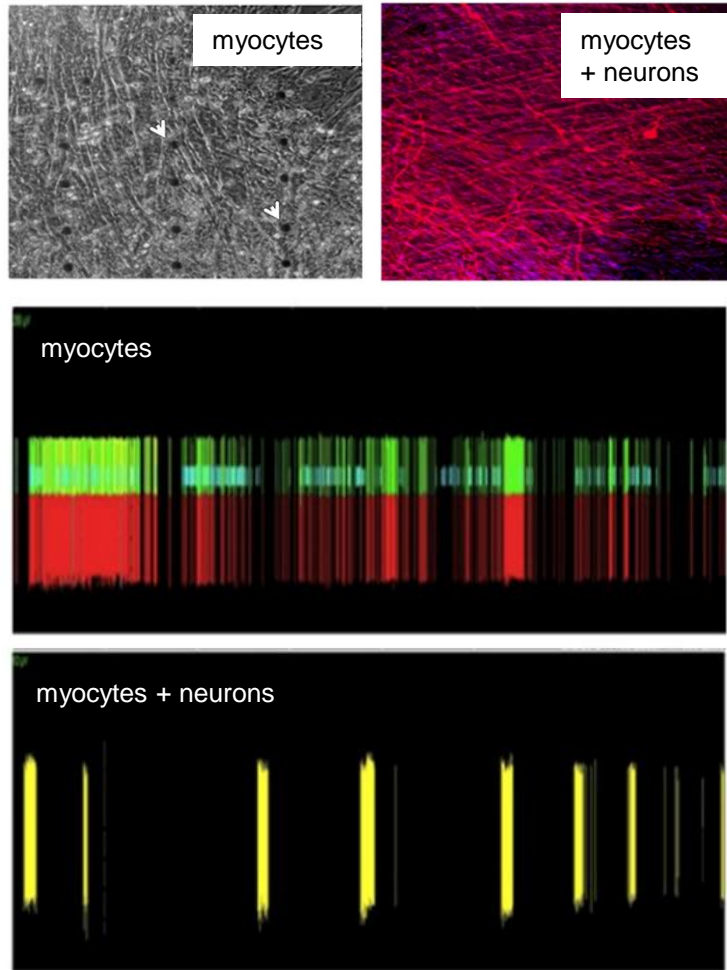


Engineered neuromuscular junction networks



M. Romero-Ortega, UTA

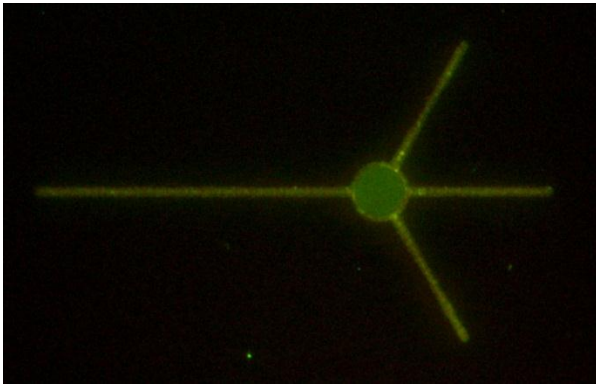
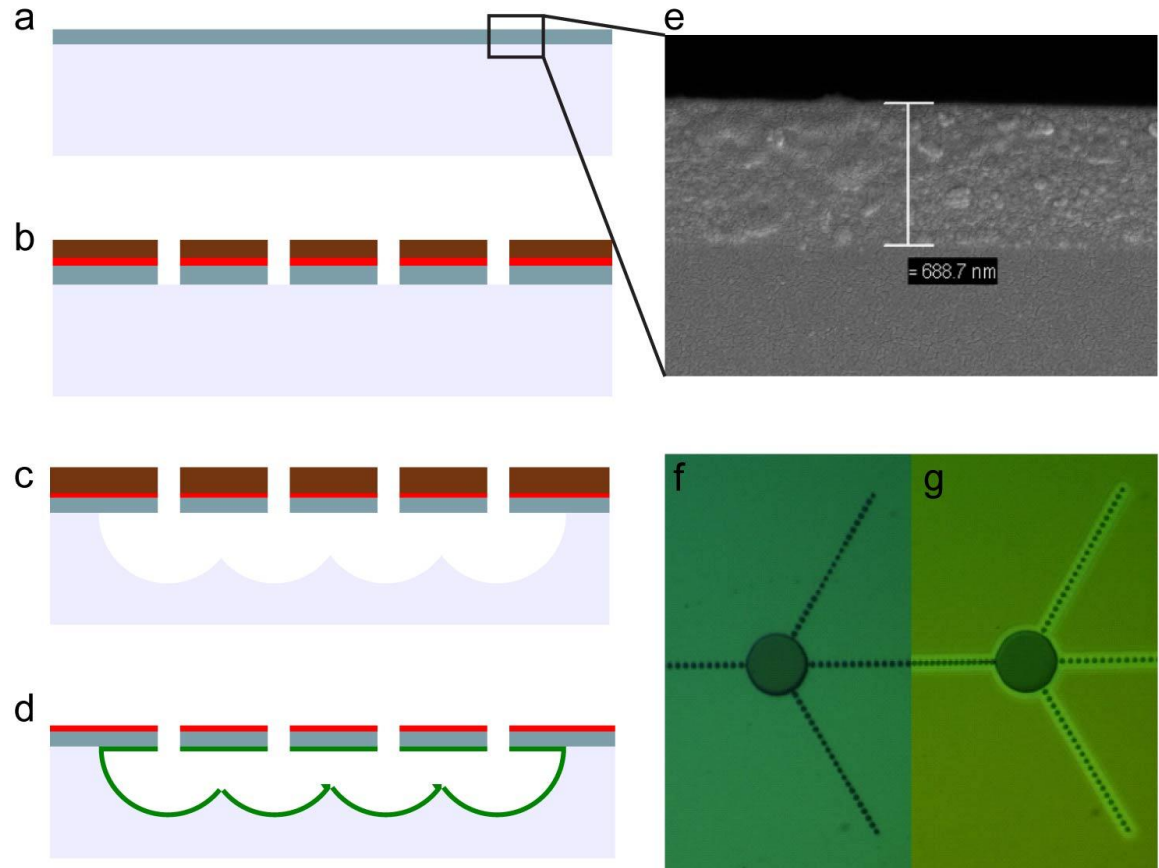
Modulation of myocyte electrical activity due to co-culture with neurons



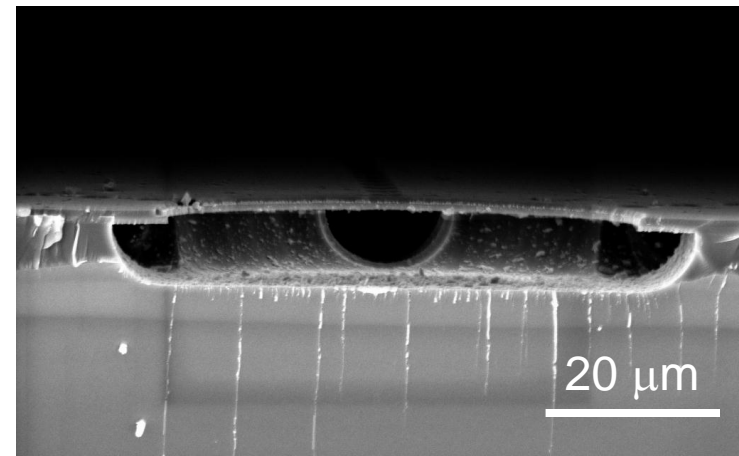
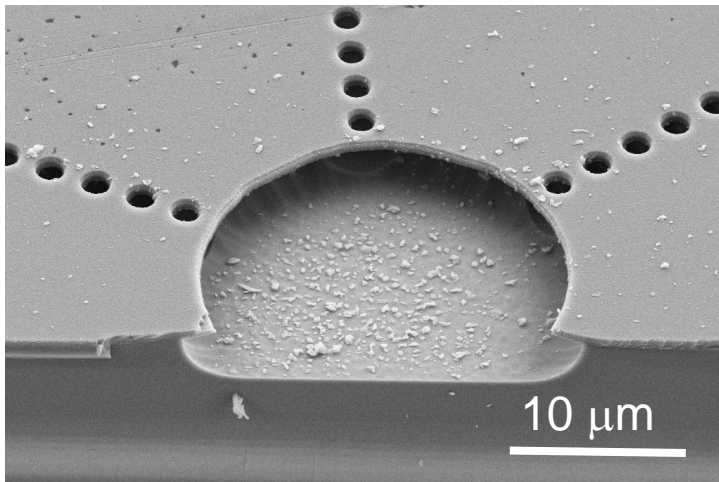
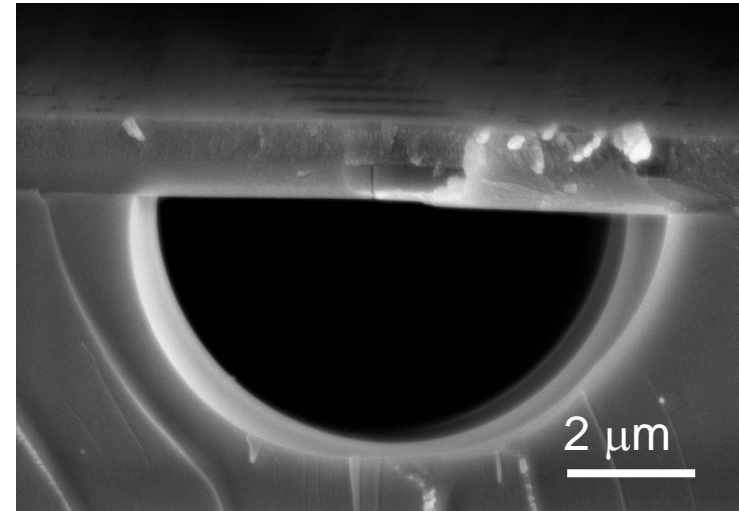
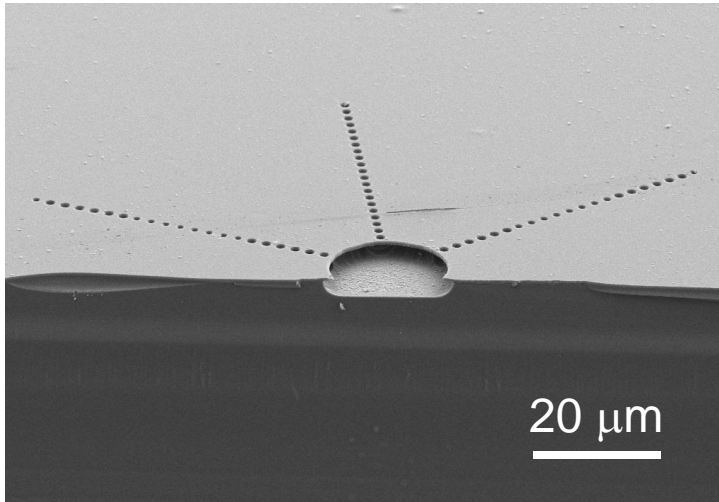
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Embedded microfluidic channels for 3D guidance of neurons

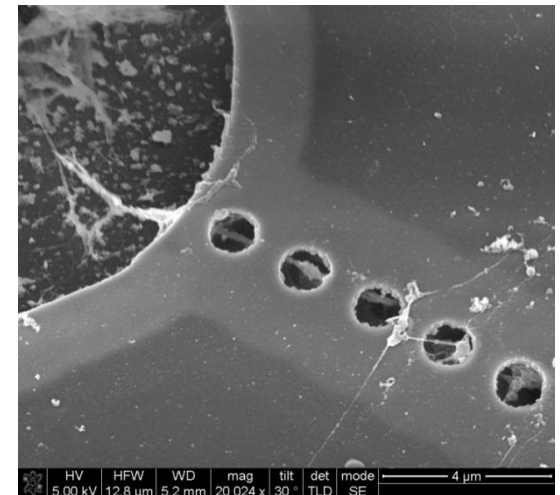
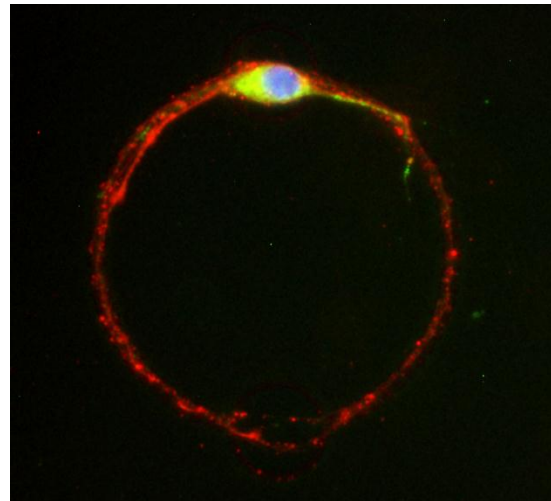
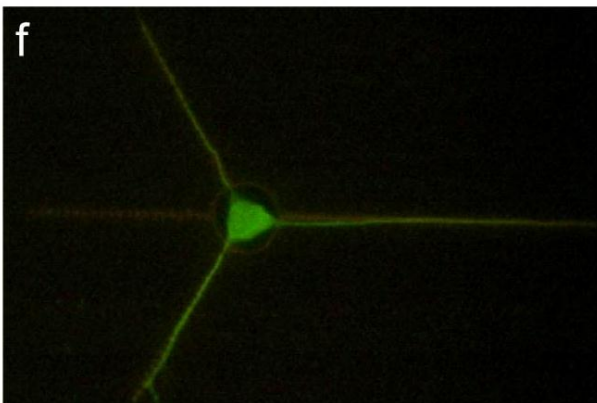
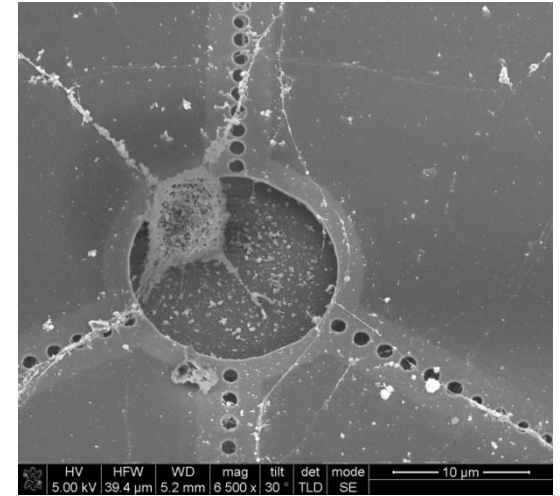
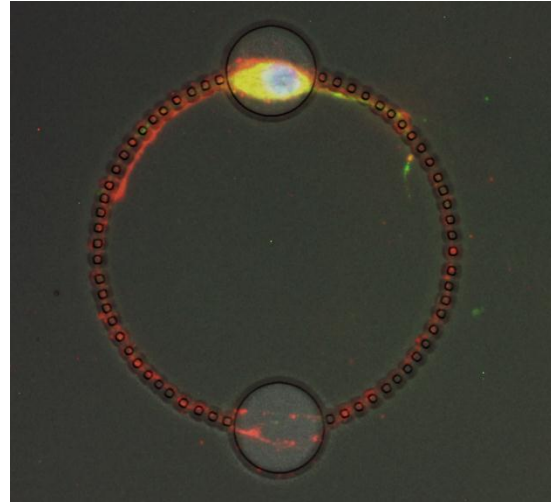
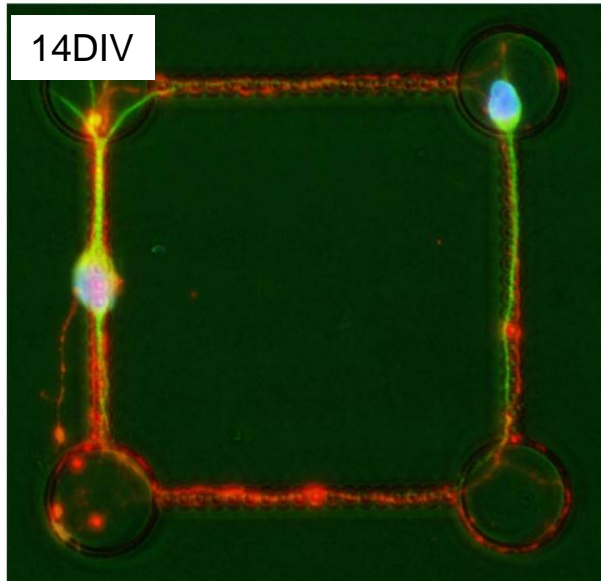
1. Deposit LPCVD SiN on fused silica
2. Pattern irrigation vias
3. Etch glass in BHF
4. Poly-lysine coating
5. Liftoff



Channels provide structural support while maintaining fluidic access for cells



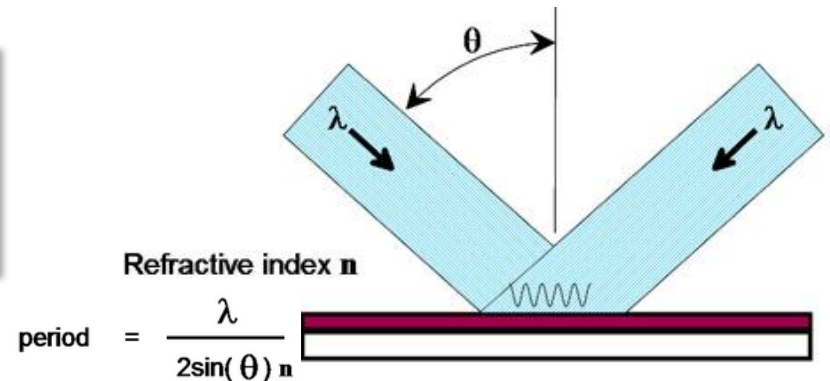
Long-term culture of neurons coupled to buried microchannels



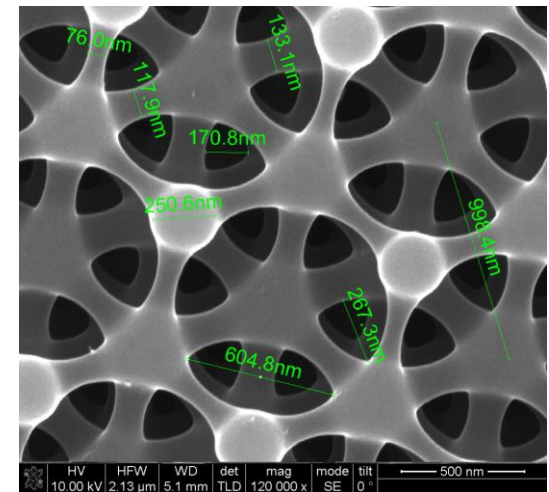
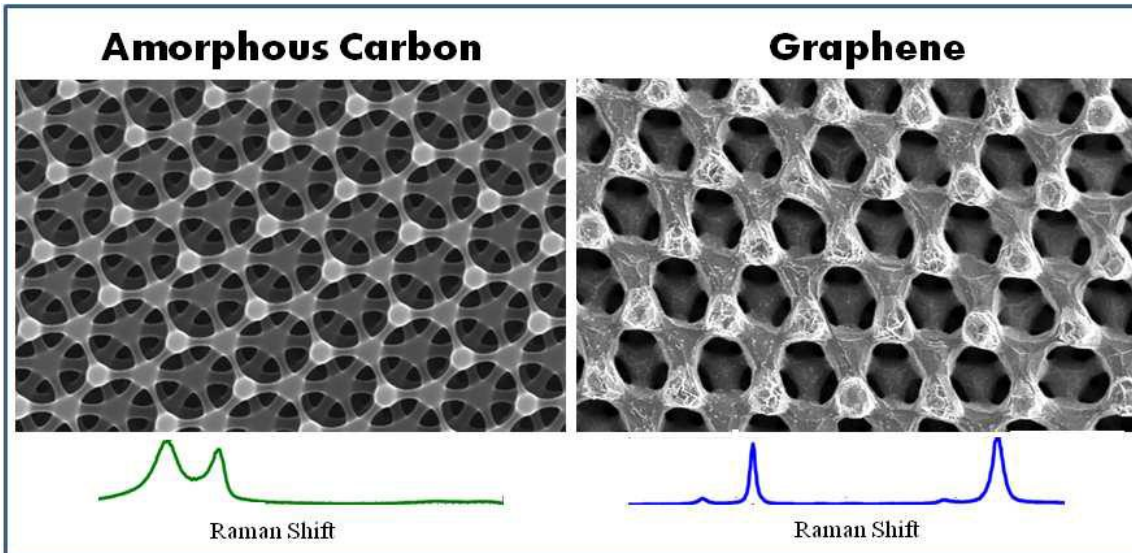
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Porous carbon nanostructures fabricated by interferometric lithography

- Microscale geometrical structures: cell body attachment, neurite outgrowth, polarization
- Nanoscale structures: dendritic spine development



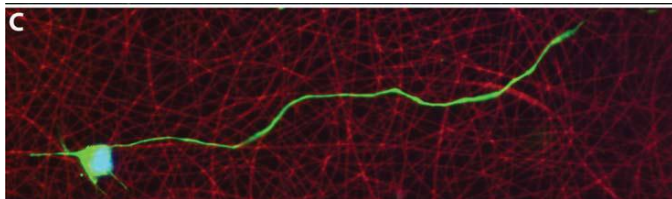
<http://www.almaden.ibm.com/st/chemistry/lithography/interferometric/>



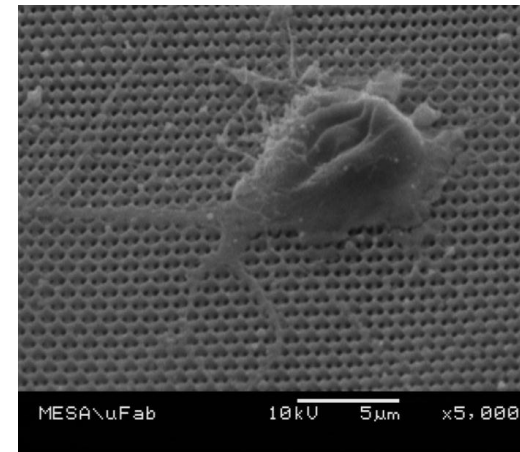
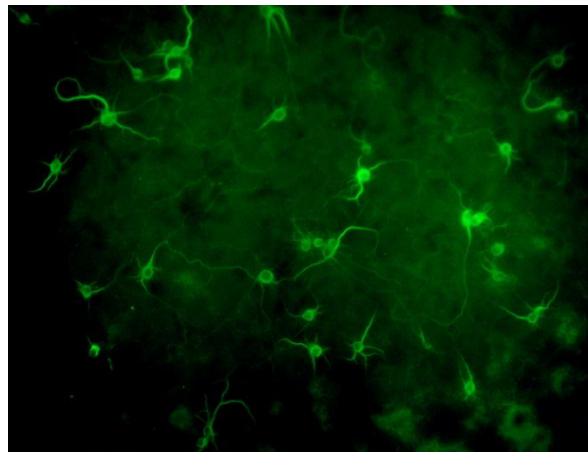
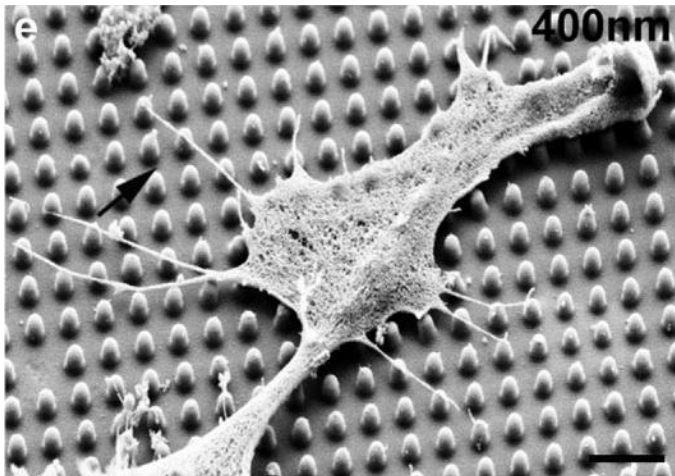
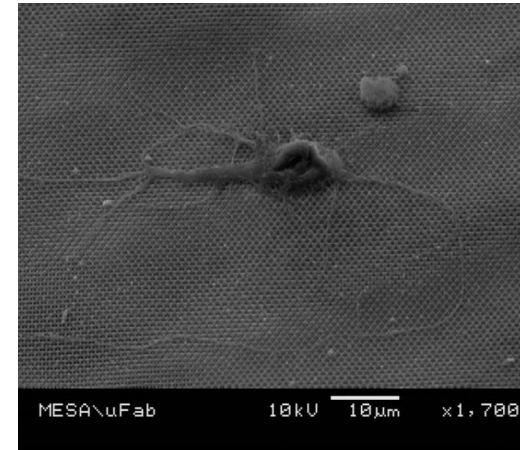
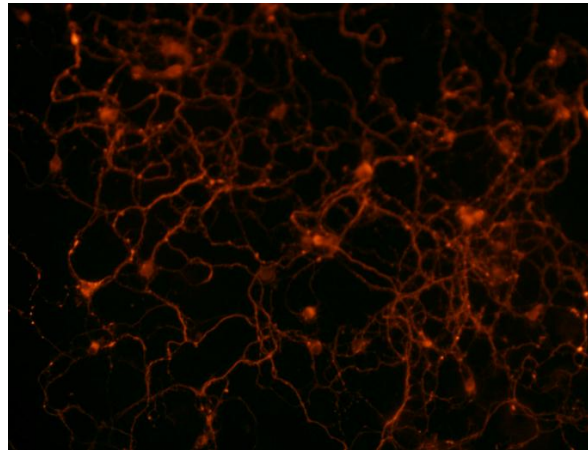
Burckel et al., *Small*, 2009, 24, 2792-2796

Neurons plated on nanoporous substrates

Nanostructured surfaces affect the maturation of dissociated cells in culture

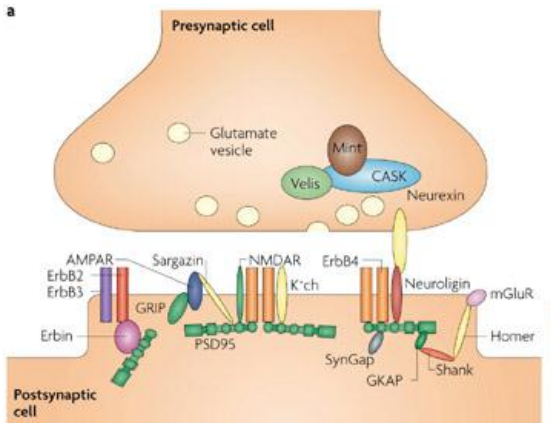


Gertz et al., *Dev Neurobio*, 2010

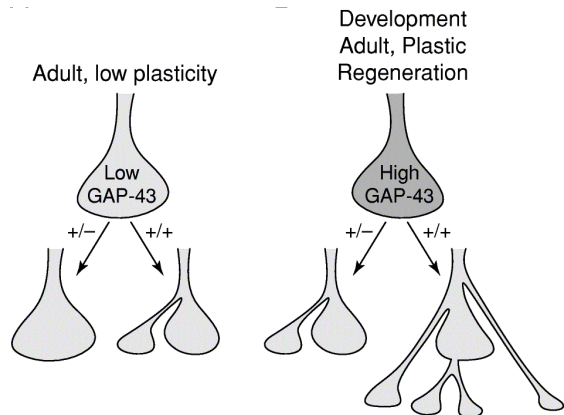
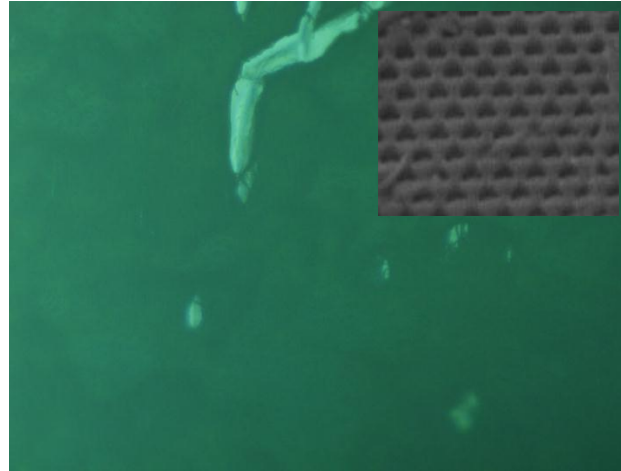


Migliorini et al., *Biotech Bioeng*, 2011

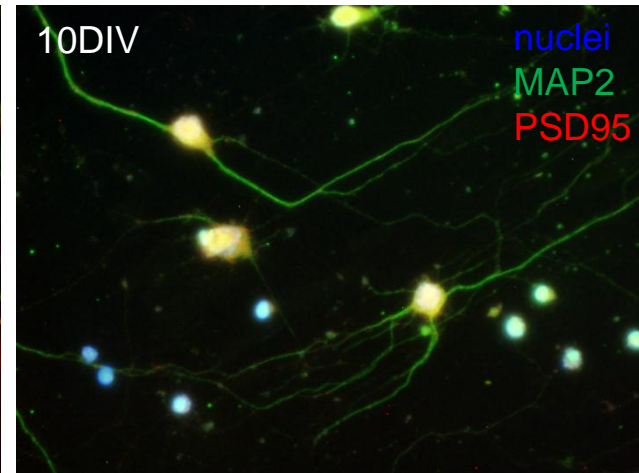
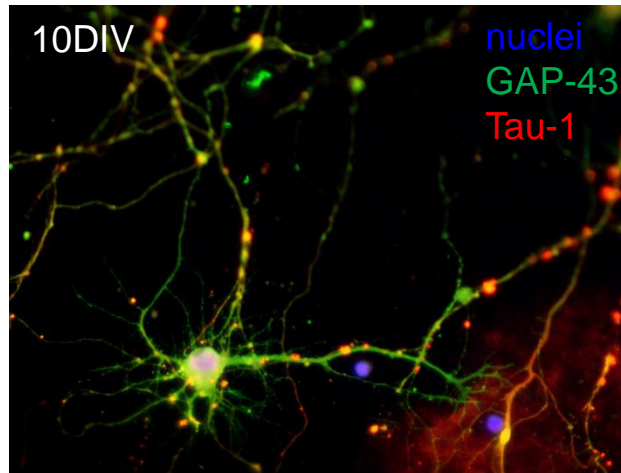
Synaptic staining of networks grown on nanostructured surfaces



Sekino et al., *Neurochem Inter.* 2007



Benowitz et al., *Trends Neuro.* 1997



Conclusions and Future Work

- Optimized geometrical cues can be used to engineer cellular networks
- Microfluidic compartmentalization can be used to generate heterogeneous multicellular networks
- 3D microscaffolds for engineering cellular networks to study granuloma formation during immune response
- Interrogation of corticostriatal network activity in tissue slices

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