Interactions of Neurons and Materials

Sabrina Jedlicka
Developmental Cell Biology
Some Perspective

- **Stem cells**: Undifferentiated cells that are capable of proliferation, self-maintenance and differentiation towards specific cell phenotypes.

- These cells can be further classified as:
  - Totipotent (all cells – arise from small cluster of cells that a fertilized egg turns into)
  - Pluripotent (all 3 germ layers)
  - Multipotent (can differentiate into a small number of cells suitable to their existing location)
  - And others (such as IPSCs)

How does this happen?

- Via a process known as signal transduction.
- Cells respond to a variety of signals:
  - ECM
  - Morphogens
  - Cytokines/Chemokines
  - Hormones
  - Growth Factors
  - Mechanical Factors
  - Etc.
In vivo vs. In vitro

- In vivo, cells respond to a number of stimuli at any given time. The process is coordinated and quite elegant.
- In vitro, we are often unable to simulate this environment effectively.
Traditional Approaches to Cell Differentiation

Petri Dish

Chemical Addition

Cells Differentiate, but functionality is limited

Chemical, Mechanical, Material Cues Depending on Location

Full Commitment

Functional Maturation
How to grow neuronal cells in culture...From proliferation to differentiation

There is a subset of bioengineering that focuses on developing biomaterials for integration with neural systems, either *in vivo* or *in vitro*

- *Used in development of cell models to understand disease*
- *Used in development of cell models for drug testing*
- *Used in developing therapies for damaged tissues*
Cell-Material Interactions

Neural Stem Cells

Cellular Mechanotransduction

Cellular Biomechanics

Surface-Induced Chemotransduction
Definitions

- **Cellular Mechanotransduction**
  - The mechanism by which cells convert mechanical signals in biochemical responses

- **Cellular Mechanobiology**
  - Characterization of cell mechanics

- **Cellular Surface Induced Chemotransduction**
  - The mechanism by which cells respond to surface-bound signals (such as ECM proteins)
Why does chemo-mechanotransduction matter?

- **Cell Proliferation**
  - Disease States
  - Propagation of sufficient cell numbers for therapeutics

- **Cell Communication**
  - Force translation via cell-cell contacts
  - Downstream biochemical signaling

- **Cell Differentiation**
  - Development of useful cell-based models for research
  - Differentiation of cells into more mature phenotypes for transplantation
Mesenchymal stem cells differentiate based on substrate elasticity (Engler et al., 2006)

Our Interest: Neural Stem Cells

Most research in NSC development is focused on soluble growth signals or ECM molecules. However, NSCs are responsive to mechanical stimuli as well.
C17.2 Neural Stem Cells

- Neuronal differentiation is controlled via serum withdrawal (starvation), which usually results in a homogeneous population of neurons.

But...
Controlling Differentiation

- In vivo processes are highly coordinated and elegant.
- Some of the proteins involved in asymmetric division are known (Example: Numb), but some of their roles are not clearly defined.
- However, in a lab setting, when you are trying to regrow neurons for therapeutic purposes – you often start with stem cells in a petri dish, which does not have the elegant coordination of signals to trigger cell changes at the right time and place.
Goals: Enhancing Differentiation

Controlling the chemomechanical environment may allow us to provide the right signals to stimulate NSCs to become post-mitotic neurons of phenotype X for treatment of a certain disease.

Alternatively, it will provide a means to produce new model system to study novel drugs.

And lots of other applications!
NSCs and Chemo-Mechanosensing

How do material cues impact differentiation of NSCs?
Mimic the extracellular environment during early corticogenesis

Diverse Neuronal Population

ECM Is Critical for Appropriate Cell Differentiation and Migration
Material composition affects cell type diversity

RGD/YIG/NID: More Neurons

RGD/YIG/IKV/VSW: More Astrocytes
Mechanical Influences

Extent of C17.2 stem cell differentiation depends on the stiffness of the substrate. Softer substrates promote differentiation into neurons and the formation of longer neurite extensions, as shown by analysis of the expression of neuron-specific β-tubulin III.
Some Surfaces lead to 100% neuronal populations
Proliferation and Neurogenesis

- The degree of cell proliferation is controlled in part by the degree of asymmetric cell division.

- When a progenitor cell divides - it can make any variety of cell combinations:
  - Two progenitors
  - One progenitor and one neuron
  - One progenitor and one astrocyte
  - Etc.
Differences in Division Type?
Results

Asymmetric Division during Differentiation on Various Substrates

During differentiation, number of asymmetric divisions is greater on glass over the “soft” surfaces.
Results

Cells grown on "softer surfaces" form functional synapses (red: synaptophysin, green: Homer)

Softer surfaces yield a more homogeneous, more mature population of neurons
Working Hypothesis

Substrates are forcing cytoskeletal changes, altering the division dynamics, force transduction, and subsequent gene expression.

Day 10
Asymmetric Division Peaks

Day 5
Cell Division Peaks

Day 10
Nestin expression falls

Day 12-14
Most prominent morphological changes
TuJ1 expression peaks

Day 23
CNTs are interacting with cytoskeleton, altering the division dynamics
** early redistribution of actin (i.e. mechanical remodeling!)

CNTs are altering gene expression, but how?
Can we manipulate these processes from the “inside-out”?

- Nanomaterials
Nanomaterials

Figure 1.3: (a) DNA wrapped SWNT (Tu et al, Nature, 2009); (b) cholatemolecule from a SWNT surrounding. Cross-sections of stabilized SWNT-surfactant systems for: (c) SDS and (d) cholate (A. Quintill et. al., 2010)

Concentration Dependent Effect

Useful for Imaging, Sensing, Etc.

Long-term Impacts?
Single CNTs in cells
The presence of CNT increases the % of cells expressing β-tubulin III.

Internal destabilization or oompliance changes?
Implications?

- Establishing an understanding of how mechanical influences (both macro and nano) change cell fate => new therapies and models
Much more work to be done...
Questions?