Impact of Mechanical Pre-stretch on Axonal Alignment and Growth

Chun Liu
Advisor: Dr. Christina Chan

Chemical Engineering and Materials Science
Spinal Cord Injury (SCI)

In the United States, the prevalence of SCI is 250,000 out of 300 million and the incidence is 11,000 new cases per year.

http://healthpages.org/health-a-z/spinal-cord-injury-function/
After SCI, **axons** break and **myelin** sheaths decompose.
What are axons and myelin?

- **Axon**
  - Electrical signals transmission

- **Myelin**
  - Prevent the electrical current from leaving the axon
  - Increase impulse propagation speed
Myelinating Cells

- Oligodendrocytes
  - Central nervous system
  - **Cell death** due to SCI

- Schwann cells
  - Peripheral nervous system
  - **Migrate** to spinal cord after SCI occurs

http://en.wikipedia.org/wiki/Oligodendrocyte
http://3.bp.blogspot.com/-Gvoes1JCslU/UilXjtIRpCI/AAAAAAAADk8/7AG4xvS2ILY/s400/schwann+cell.gif
Damages after Spinal Cord Injury

• **Primary damage**
  - Neurons and glial cells death
  - Blood vessel damage
  - Spinal cord swelling

• **Secondary damage**
  - Glial cells death
  - Breakdown of myelin sheath
  - Immune cell infiltration and inflammation

D.L. Stocum, et al. Regenerative Biology and Medicine, 2012

Demyelination

Demyelinated axons lose the ability to transmit nerve impulses.

http://www.sickkids.ca/images/Research/MSKids/37532-shutterstock_demyelinating%20neuron.JPG.
Chronic Progressive Demyelination accompanies SCI

- Oligodendrocytes death
- Remyelination is limited

Quantification of demyelinated axons throughout the craniocaudal axis of the spinal cord at 1, 14, 28, and 450 days post injury.

M. O. Totoiu, et al. Journal of Comparative Neurology, 2005
Neural Regeneration to Treat SCI

• Axon regeneration
  – Axon alignment
• Remyelination

http://medicalterms.info/anatomy/Myelination/
Axon Alignment is Important for Neural Regeneration after SCI

- Properly organized axonal alignment is required for axons to bridge the lesion site.

Transplantable Scaffold to Aid Axon Alignment

How to Enhance Myelination

Myelination is dependent on axon thickness
Research Goal

Axon alignment
Axon thickness

Spinal cord injury

Engineering approach
Rationale

Pre-stretch

Surface anisotropy

Axon alignment

Axon thickness

Myelination
Surface Anisotropy

**Anisotropy** can be defined as a difference, when measured along different axes, in a material's physical or mechanical properties.

Mechanical Pre-stretch Induced Surface Anisotropy

- **Effective Stiffness vs. directions**

Before

Isotropic surface (unstretched)

Anisotropic surface (uniaxially Stretched)

After

Cells tend to orient in the direction of maximum effective stiffness
Mesenchymal Stem Cell Alignment on Pre-stretched Surface

Crosslinking ratio: 35:1 (elastomer: crosslinker)
Young’s modulus: 270kPa
Membrane dimension: 5*3.5 cm²
Membrane coating: Poly-L-Lysine (PLL)

Axon Alignment on Pre-stretched Surface day6

DRG neurons on 16.7% pre-stretched vs. unstretched PLL coated 10:1 PDMS surface on day6, axons were stained by Tuj-1 antibody and Alexa 488 secondary antibody.
Axon Alignment on Pre-stretched Surface
day12

DRG neurons on 16.7% pre-stretched vs. unstretched PLL coated 10:1 PDMS surface on day12, axons were stained by Tuj-1 antibody and Alexa 488 secondary antibody.
Axon Alignment Quantification day3

Axon Orientation on stretched surface

Axon orientation on unstretched surface

0°

Orientation angle

Pre-stretch direction

90°

90 degree represents parallel to the stretching direction
Axon Thickness Quantification day6

Axon bundle thickness

Axon thickness (micron)

stretched

unstretched

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Summary

- Mechanical Pre-stretch
- Surface anisotropy
  - Axon alignment
  - Axon thickness
  - Remyelination
- Neural regeneration
- Spinal cord injury
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Thank you!

& QUESTIONS?