Cellular Biomechanics

Linda Lowe-Krentz

Bioscience in the 21st Century

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Outline for today

- Tubes
- Vessel mechanics and disease
  - Models
- Lung mechanics
- Technology integration
Atherosclerosis is Geometrically Focal Disturbed Flow Region

“Sticky” ECs

“Leaky” Endothelium

Smooth Flow Region

“Inflamed” vasculature

“Leaky” Endothelium

Meron Mengistu

Flow, along with other factors, contributes to risk.
Effects of flow on endothelial cell morphology

- Models are helpful in the study of molecular events in cell culture

12 to 24 hours
Stress Fiber Alignment

• High Fluid shear stress exposure results in three phases of cell changes:
  
  – (1) increase in stress fiber formation
  – (2) dense cortical band formation of actin cytoskeleton
  – (3) stress fiber alignment in the direction of flow

No Flow

\[ \text{Phase 1} \quad \text{Phase 2} \quad \text{Phase 3} \]

Meron Mengistu
Actin remodeling under shear stress

Low stress for 30 min

High stress for 30 min
Cells and their actin filaments begin to align in the direction of flow after 60’ of high shear flow exposure. No change under low shear stress.
Effect of Flow Exposure on Endothelial Cells

% Elongation of Long-axis

- 15 min
- 30 min
- 60 min
- 120 min

σ_{eff}

4 dyn/cm²  15 dyn/cm²

ADINA models

control  15 min  30 min  60 min  120 min

Jamie Maciaszek, Shannon Alejandro, Josh Slee, Samir Ghadiali
Finite-Element Models

- 4 dyn/cm$^2$
- 15 dyn/cm$^2$

control  15 min  30 min  60 min  120 min

$\sigma_{eff}$
Add into the model Actin structures we see

No Flow → 5’ – 15’ \( \uparrow \) FSS → 30’ \( \uparrow \) FSS → 60’ – 120’ \( \uparrow \) FSS
Maximum Effective Stress

Effect of Flow Exposure on Basal Surface of Cell

- **4 dyn/cm²**
- **15 dyn/cm²**

<table>
<thead>
<tr>
<th>Time</th>
<th>4 dyn/cm²</th>
<th>15 dyn/cm²</th>
</tr>
</thead>
<tbody>
<tr>
<td>5 min</td>
<td>1 x 10^4</td>
<td>3 x 10^4</td>
</tr>
<tr>
<td>15 min</td>
<td>1 x 10^4</td>
<td>4 x 10^4</td>
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<tr>
<td>30 min</td>
<td>1.5 x 10^4</td>
<td>2 x 10^4</td>
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<tr>
<td>60 min</td>
<td>1 x 10^4</td>
<td>1 x 10^4</td>
</tr>
<tr>
<td>120 min</td>
<td>1 x 10^4</td>
<td>0.5 x 10^4</td>
</tr>
</tbody>
</table>
MECHANOSENSING

β/γ

α

Tyr

Tyr

Tyr

Tyr

Tyr

Tyr

Ca²⁺

Influx

K⁺ Outflux

Na⁺ Influx

Ca²⁺ Influx

Integrin

Caveola

PECAM

Src

Shp2

Small GTPases

MAPK Signaling Cascade

MEK1/2

MEK4/7

MEK3/6

Biological Responses
How to continue this work

• Test the model
  • Do any appropriate signaling proteins turn on?
  • Are there changes in surface attachment proteins at the right time?
  • Use vinculin antibodies
  • Look for how many patches of staining are in the cells after flow and where they are.
Average number FAs/Cell

Number of Focal Adhesions (FAs)

Time Exposed to high FSS of 15dyn/cm²
How to continue this work

• Block the signaling molecule(s)
  • Interfere with the attachment proteins
  • Chemically block signaling enzymes

• Refine the model
  • Alter the attachment?
  • Test again
  • Identify signal targets
  • Test their identities

Infinitely Bounded:  Periphery Bounded:
More tubes

- Global Lung Mechanics (the project is from Professor Samir Ghadiali – Ohio State)
Acute Lung Injury

- Infections → Necrosis and Detachment of alveolar epithelial cells.

- ↑ permeability of alveolar-capillary barrier → Flooding of small airways/alveoli

- ↓ gas exchange, severe hypoxia

- Standard of care: Mechanical Ventilation

- Ventilators cause additional cell injury (Ventilator Associated Lung Injury)

  - 200,000 patients/yr, 30% to 40% mortality rates, 3.6 million hospital days

  - Major public health issue, significant burden on health care system

Ware LB & Matthay MA,
Quantification of Cellular Morphology

- Laser scanning confocal images of cells with cytoplasmic stain (Calcein AM).
- Cells in 100% monolayer are flatter and thinner.

Convert confocal cross-sections to finite element models.
Microbubbles and Cell Injury

- Ventilators reopen fluid-filled lung regions (airways/alveoli) with microbubble flows.
- Microbubbles generate forces that cause cell deformation, death and detachment.
- IDEA: Make cell’s more rigid to prevent cell deformation, death and detachment.

Experimental data:
Cell death (red cells)
Cell detachment (cell loss)

Ghadiali
Optical Tweezers Methodology

Output:

Displacement: $D(\omega)$

Phase Shift: $\delta(\omega)$

Forced Oscillation

$\omega$: 0.1 – 6,000 Hz

Mengistu and Ou-Yang
Studying Micro-Mechanical Properties of Endothelial Cells

Intracellular Probes

- Intrinsic Structure
- Endocytosed Bead

Extracellular Probes

- Integrin-bound Bead
- PECAM-bound Bead
- Microtubules
- Actin Filaments
- Intermediate Filaments

Mengistu and Ou-Yang
Biological/Physiological System are very complex!

Biologists provide powerful tools to probe these systems.

Engineers provide mathematical tools which can be used to understand how the different components interact.