Lecture #15: Cell Migration
Cell Migration

Cellular Fate Processes

Migration

Adhesion

Replication

Differentiation

Apoptosis
Cell Migration

• Why do cells migrate?
  – Development
  – Immune response
  – Wound healing
  – Matrix remodeling
  – Uncontrolled migration $\rightarrow$ cancer

• How do cells migrate?
  – Majority of cells are stationary $\rightarrow$ Adhesion
  – In response to cues
  – Randomly
  – Via extracellular matrix
Cell Migration – A Dynamic Biochemical Process

- ATP $\rightarrow$ proteins $\rightarrow$ mechanical work
- Proteins polymerize into bundles and networks
  - Microfilaments (actin) 7-9 nm
  - Intermediate filaments 10 nm
  - Microtubules (tubulin) 24 nm
- Motor Proteins – myosin

Actin – green filament
Leading edge
Cell Migration

- Migrating cells in culture look as though they are moving in a particular direction.
- The shape of the cytoskeleton changes internally due to the pattern of adhesion.
- The “front” is called the leading edge or lamellipodium.
- The “rear” is called the uropodium.
Cell Migration

1. Protrusion of membrane lamellipodia
2. Adhesion to the matrix occurs via integrins
3. Contraction of the cytoplasm by myosin-based motors
4. Rear release and forward displacement
5. Integrin recycling
Cell speed depends on substrate density

- Cell migration and adhesion are linked by the ECM concentration and the receptor-ligand affinity.
Example: Determining direction of cell migration

- CHO cells are seeded uniformly at the beginning of the experiment
- Maximum migration speed: 20 µm/hour

- After 24 hours, are the cells at equilibrium? What if the strips were 500 µm wide?
Example: Determining direction of cell migration

40 \mu m

<table>
<thead>
<tr>
<th>Fn concentration in adsorbing solution (\mu m/mL)</th>
<th>Cell distribution after 24 hours</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>All cells on “high” strips</td>
</tr>
<tr>
<td>5</td>
<td>All cells on “low” strips</td>
</tr>
</tbody>
</table>

Interpret these results – offer mechanistic hypothesis

Suggest additional experiments
Example: Determining direction of cell migration

- Possible #’s for illustration purposes

<table>
<thead>
<tr>
<th>[Fn]soln (µm/ml)</th>
<th>[Fn]surface high</th>
<th>[Fn]surface low</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>5</td>
<td>3</td>
</tr>
<tr>
<td>10</td>
<td>10</td>
<td>7</td>
</tr>
</tbody>
</table>

- On high [Fn] strips, more R/L contacts
- On low [Fn] strips, less R/L contacts
- Proposed model: At 10µm/mL, cells on “high” strips because many R/L contacts and cell cannot generate enough force to break attachments
- At 5µm/mL, cells on “low” strips because there are very few R/L contacts and cells cannot generate enough force via such few contacts to form enough attachments to move
- Possible experiments: check cell morphology, integrin engagement (FAK), chemical perturbations, check [Fn] …
A problem on cell motility.

(A) A cartoon illustration of the migration of a typical mammalian cell on a flat surface. This teardrop shape is found in many types of cells.

(B) Cells confined to squares preferentially extend their lamellipodia from the corners. nu, nucleus.

(C) If a cell is confined to a shape of teardrop, will the cell preferentially extend its lamellipodia from the sharp end or from the blunt end? If released from confinement, in which direction will it likely move?
Results

- Cells initially confined to the teardrop patterns (with an area of ≈2,000 \( \mu m^2 \); observed a total of 45 cells) moved predominantly (82%) toward the blunt end!!

More Fun Shapes

1

2

3
Cell Migration

• Directed
  – Using ECM cues, mechanotransduction (varying stiffness of a material) or chemotaxis signaling

• Random
  – Persistent random walk at short observation times and Brownian at longer periods
Movement toward a chemoattractant

- We can evaluate the “strength” of an attractant by monitoring migration toward it.
- The strength is measured by the percentage of cells moving the “correct” direction up the gradient.
- What might this depend on??
  - Concentration of attractant
  - Steepness of the gradient
Chemokinesis vs. Chemotaxis

- **Chemotaxis** (directional)
  - Chemoattractant in fluid phase

- **Chemokinesis** (random)
  - Chemoattractant in fluid phase

- **Haptotaxis**
  - Chemoattractant molecules associated to the surface

- **Necrotaxis**
  - Necrotic cell + released substances

© Kohida, L. 2008
Transwell Migration Assays

(a) Random migration (chemokinesis)
(b) Directional migration (haptotaxis)
(c) Directional migration (chemotaxis)
(d) Chemoinvasion/transmigration

Eccles et al., Biotech Ann Review, 2005
How do we observe migration?

• Time lapse imaging
  1. Scratch test
  2. Label with GFP or other imaging protein
Migration Assays

• Simple scratch test
  – http://www.youtube.com/watch?v=eTWC4PFSpzQ

• Patterns in 1D mimic 3D movement
  – http://www.youtube.com/watch?v=uM8YT52H0l4
Neutrophil chemotaxis in linear and complex gradients of interleukin-8 formed in a microfabricated device

Noo Li Jeon†, Harihara Baskaran†, Stephan K. W. Dertinger², George M. Whitesides², Livingston Van De Water¹,³, and Mehmet Toner¹*

Published online: 1 July 2002, doi:10.1038/nbt712

Nature Biotech
Dilution network

[Diagram of dilution network with labels for PDMS, Coverlip, Inlets, Outlet, Neutrophils, and Flow]

[Graphs showing normalized intensity vs. channel width (μm)]
Table 1. Chemotaxis parameters of neutrophils

<table>
<thead>
<tr>
<th>Gradient</th>
<th>Chemotactic flux $\frac{\delta L}{\delta x}$ (ng·ml$^{-1}$·500 µm$^{-1}$)</th>
<th>Chemotaxis coefficient $\chi$ (10$^9$ mm$^2$·s$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>1.2</td>
<td>12.0</td>
</tr>
<tr>
<td>50</td>
<td>5.5</td>
<td>5.8</td>
</tr>
<tr>
<td>500</td>
<td>6.0</td>
<td>0.6</td>
</tr>
</tbody>
</table>

Chemotactic flux is obtained by calculating the initial average speed of neutrophil density data at different homogeneous linear gradients (Fig. 3). Chemotaxis coefficients were obtained by dividing the chemotactic flux by the corresponding gradient$^{26}$. 
3D Migration Platform

Han, Lab Chip, 2012
3D Migration Platform

Han, Lab Chip, 2012
3D Migration Platform

Han, Lab Chip, 2012
3D Migration Platform

a

ECM

PDMS

Glass coverslip

b

Number of Transmigrated Cells

Migration Distance (mm)

1h

fMLP 0nM
fMLP 10nM
fMLP 100nM
fMLP 1000nM

12h

c

Number of Transmigrated Cells

Migration Distance (mm)

1h

IL-8 0ng/ml
IL-8 1ng/ml
IL-8 10ng/ml
IL-8 100ng/ml

12h

Han, Lab Chip, 2012
Overview of 2D and 3D platforms

Micropipette

Boyden

Micropatterning

Durotaxis

Wound healing

3D ECM

Microfluidics

Cytokine gradient

Microfluidics

Interstitial flow