Introduction to Cell-Biomaterial Engineering

Module 3, Lecture 1

20.109 Spring 2010
Topics for Lecture 1

• Introduction to tissue engineering
  – motivation
  – basic principles + examples

• Introduction to Module 3
  – background: cells and materials
  – experiment: purpose and structure
Ability to repair tissue is limited

- Severe trauma (acute or disease-challenges tissue repair capacity
- Donor tissue
  - scarcity, immune response (graft or
- Autologous tissue
  - availability, donor site morbidity
- Permanent synthetic substitute
  - inflammation, mis-match, failure
- A new approach: promote regeneration of ~native tissue
“TE... applies the principles of engineering and the life sciences toward the development of biological substitutes that restore, maintain, or improve tissue function.”


**What is in a tissue engineer’s toolkit?**

**How good are the outcomes?**

Image by MIT OpenCourseWare. After Langer and Vacanti (1993).
Scaffolds provide a framework

• Why a porous, degradable scaffold?
  – mechanical support
  – allow ingrowth, avoid inflammation
  – promote nutrient+oxygen diffusion

• How is the scaffold made degradable?
  – cross-links susceptible to cleavage
Cytokines promote cell functions

• Types of cytokines
  – growth factors (FGF, TGF, BMP)
  – angiogenic (VEGF)
  – chemokines (attract cells)

• Delivery of cytokines
  – release from scaffold or transplanted cells

• Example: CCL21 promotes T cell migration

Control

See supporting video, "Chemokinesis control."
See supporting video, "Chemokinesis +CCL21."

Courtesy of Darrell Irvine. Used with permission.
Cells make up tissues

- Progenitors vs. differentiated cells
  - scarcity, function
- Transplanted vs. *in situ* cells
  - scarcity, safety

- Example: tumor-infiltrating lymphocytes (TIL)
  - T cells lose function in tumors
  - expand TIL *ex vivo*, treat with cytokines, and transplant
  - tested in mice


Courtesy of Willem Overwijk, et al., and Rockefeller University Press.
Components of a TE construct

**scaffold/matrix**
- usually degradable, porous

**soluble factors**
- made by cells or synthetic
- various release profiles

**cells**
- precursors and/or differentiated
- often autologous

**integrated implantable or injectable device**
Putting it all together: *in vitro* construct

See supporting video, "Cells in Scaffold."

Interlude: Shmeat


2:24 – 4:32
Commercial success in TE

• Regenerating severely burned skin
    • top: protects wound, retains fluid
    • bottom: provides scaffold for growth
  – forms neotissue comparable to native skin
  – sold as Integra Dermal Regeneration template

www.integra-ls.com/products/?product=46

Courtesy of Integra LifeSciences Corporation. Used with permission.
Joint diseases: an unmet need

- Leading cause of physical disability in U.S.
- $100’s billion in in/direct costs
- Osteoarthritis
  - common in elderly population
  - acute injury (athletes) → susceptibility to early disease
- [http://www.youtube.com/watch?v=0dUSmaev5b0&feature=related](http://www.youtube.com/watch?v=0dUSmaev5b0&feature=related)
- Limited pharma solutions
  - pain management
  - targets unknown
  - cell therapies (Genzyme, Osiris)

Self-reported disease in U.S., 2005

<table>
<thead>
<tr>
<th>Condition</th>
<th>Rate per 100 Population</th>
</tr>
</thead>
<tbody>
<tr>
<td>Muscle/bone</td>
<td>0.7</td>
</tr>
<tr>
<td>Heart</td>
<td>1.7</td>
</tr>
<tr>
<td>Lung</td>
<td>0.5</td>
</tr>
</tbody>
</table>

Our focus: cartilage tissue

Water-swollen, heterogeneous, avascular tissue.
Alginate: material for 3D culture

- Seaweed-derived polysaccharide
- Co-polymer of M and G acids
- G-block polymer chains cross-linked by cations (e.g., Ca$^{2+}$)
- Forms water-swollen gel

- G/M content and MW influence
  - mechanical properties
  - swelling
  - degradability
  - viscosity of solution
# Cells for cartilage TE

<table>
<thead>
<tr>
<th></th>
<th><strong>Stem cells</strong></th>
<th><strong>Chondrocytes</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>Obtained from…</td>
<td>Bone marrow</td>
<td>Digested cartilage</td>
</tr>
<tr>
<td>Recovery</td>
<td>Difficult, initially very few cells</td>
<td>Easy, many cells</td>
</tr>
<tr>
<td>Expansion</td>
<td>Many-fold</td>
<td>Minimal</td>
</tr>
<tr>
<td>Upkeep</td>
<td>FGF to expand, TGF-β1 to differentiate</td>
<td>Multiple factors to maintain phenotype</td>
</tr>
</tbody>
</table>

---

*Images of stem cells, chondrocytes, and fibroblasts.*
Specific goal and experiments

- **Goal:** examine effect of specific culture conditions on chondrocyte phenotype

- Observe cell morphology and viability
- Measure collagen content
  - Gene (RT-PCR) and protein (ELISA) expression
  - Collagen II:I ratio reflects cell state

- Grander purpose: cartilage TE
  - conditions for *ex vivo* cell expansion
  - conditions for *in vitro* cartilage production
Module overview: lab

Day 1: design

Day 2: seed cultures

Day 3: viability assay

Day 4: prep RNA+cDNA

Day 5: transcript assay

Day 6: protein assay

Day 7: remaining analysis

Day 8: your research ideas!
Lecture 1: conclusions

• Tissue engineering is an emerging interdisciplinary field
• Maintaining cell function is a key part of TE
• Alginate beads provide a culture system for researching soft tissues such as cartilage

Next time… more about engineered and natural biomaterials.