M3D1: Design Experiment

Announcements

- Module 2 Report due **Wednesday at 5pm**
  - ★ submit to 20109.submit@gmail.com
  - ★ free late day if you visit writing center
  - ★ methods/results -- me ; all else -- Alan
  - ★ OH tonight 7-10pm
- Next time, two shifts (1pm or 3pm) **--** quiz before or after

Module 2 Reports

- ## Comments on Methods

  - concise statements, higher-level subsections

  - only report methods for experiments where you present the results

  - Design --> Experiments --> Analysis

  - remember kits -- did we deviate from those kits?

- ## Comments on Results

  - tell a story -- what did you ask, what did you find, how did that lead you to the next question?

  - concise statements! no interpretation

  - subheadings should be informative

  - SDS-PAGE vs. Bradford assay

Tissue culture environment

What is the 'cellular environment' *in vitro*?

1. Temperature = 37C

2. Oxygen level -- ambient, normally

3. CO_2 level (5%) -- $NaHCO_3$

4. pH 7.2-7.4

5. Humidity (>95%)

6. [salt] -- osmolarity

*Sterility -- absence of exogenous microorganisms

Bovine bone marrow stem cells in exracellular matrix
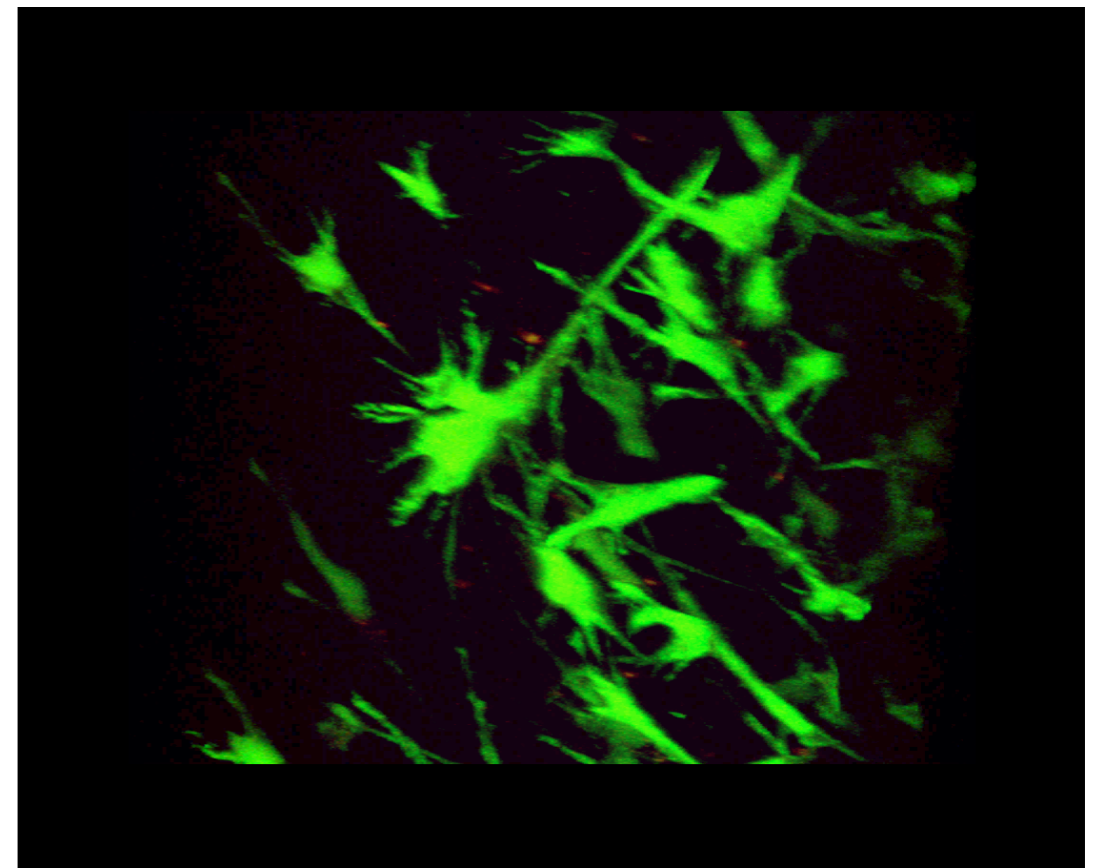
University of Oxford Bioengineering

Tissue culture medium

What do cells need to survive?

Cell culture media:
DMEM

Food(s):

Optimized:
[salt]
[vitamins]
[amino acids]
[glucose]

Non-essential
amino acids:

L-glutamine

Why?

**Components of
biochemical Rxns
for cell survival.**

Serum:

Non-food(s):

Growth factors,
lipids,
cytokines

Antibiotics (Pen/Strep)
Antimycotics (AmpB)
Phenol Red -- Why??
    pH indicator --
        something is wrong!

Tissue engineering considerations:

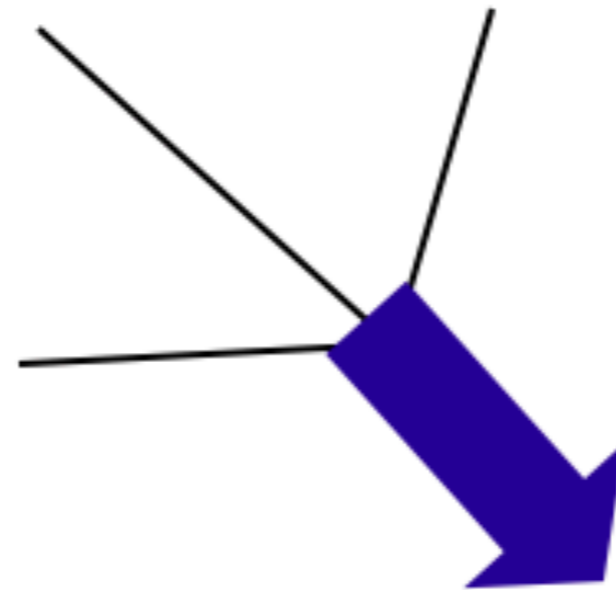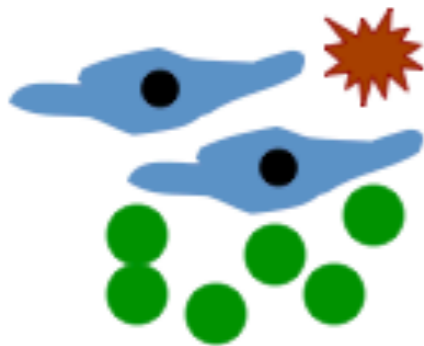**scaffold/matrix**
→ usually degradable, porous
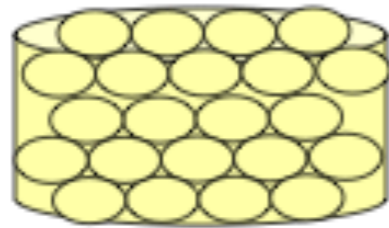
soluble factors
→ made by cells or synthetic
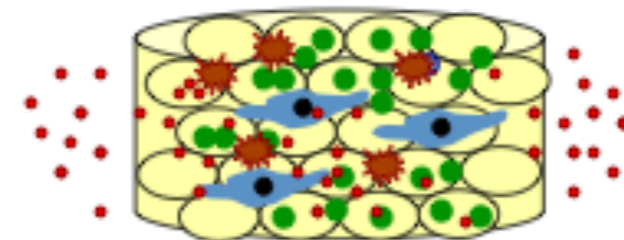→ various release profiles

cells
→ precursors and/or differentiated
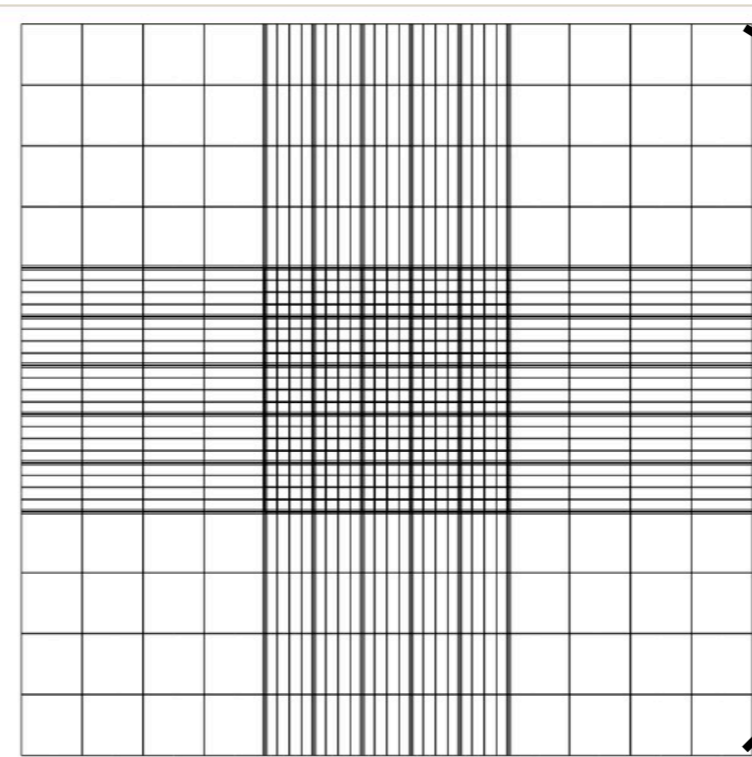→ usually autologous
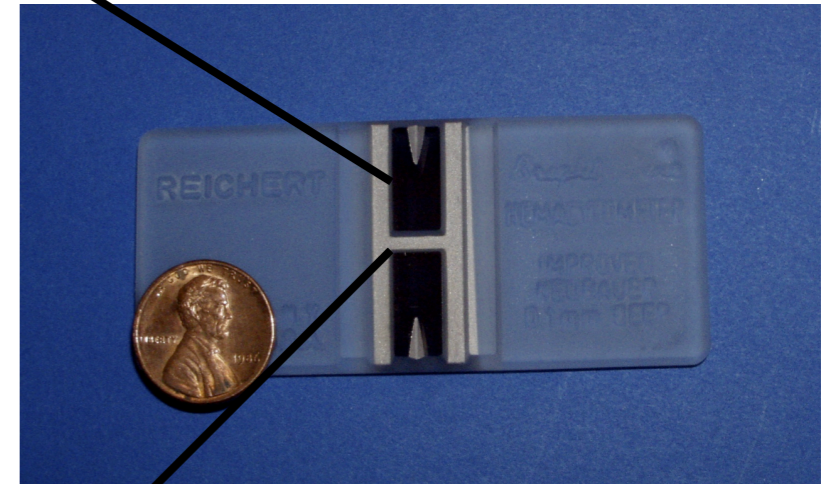
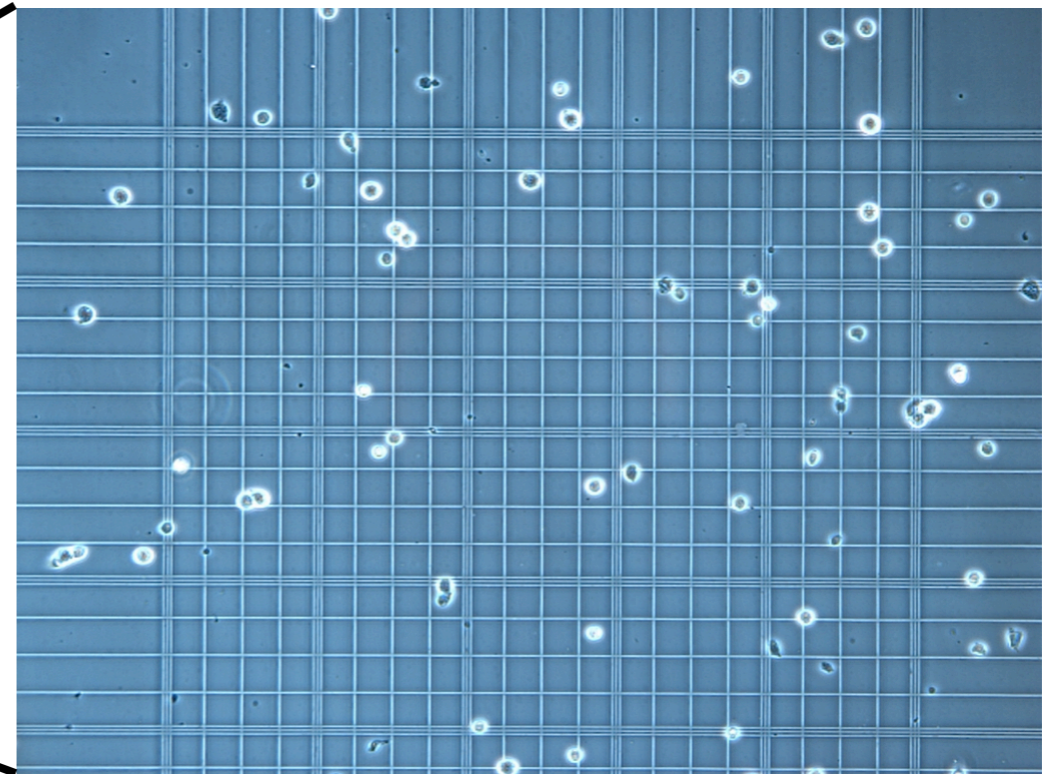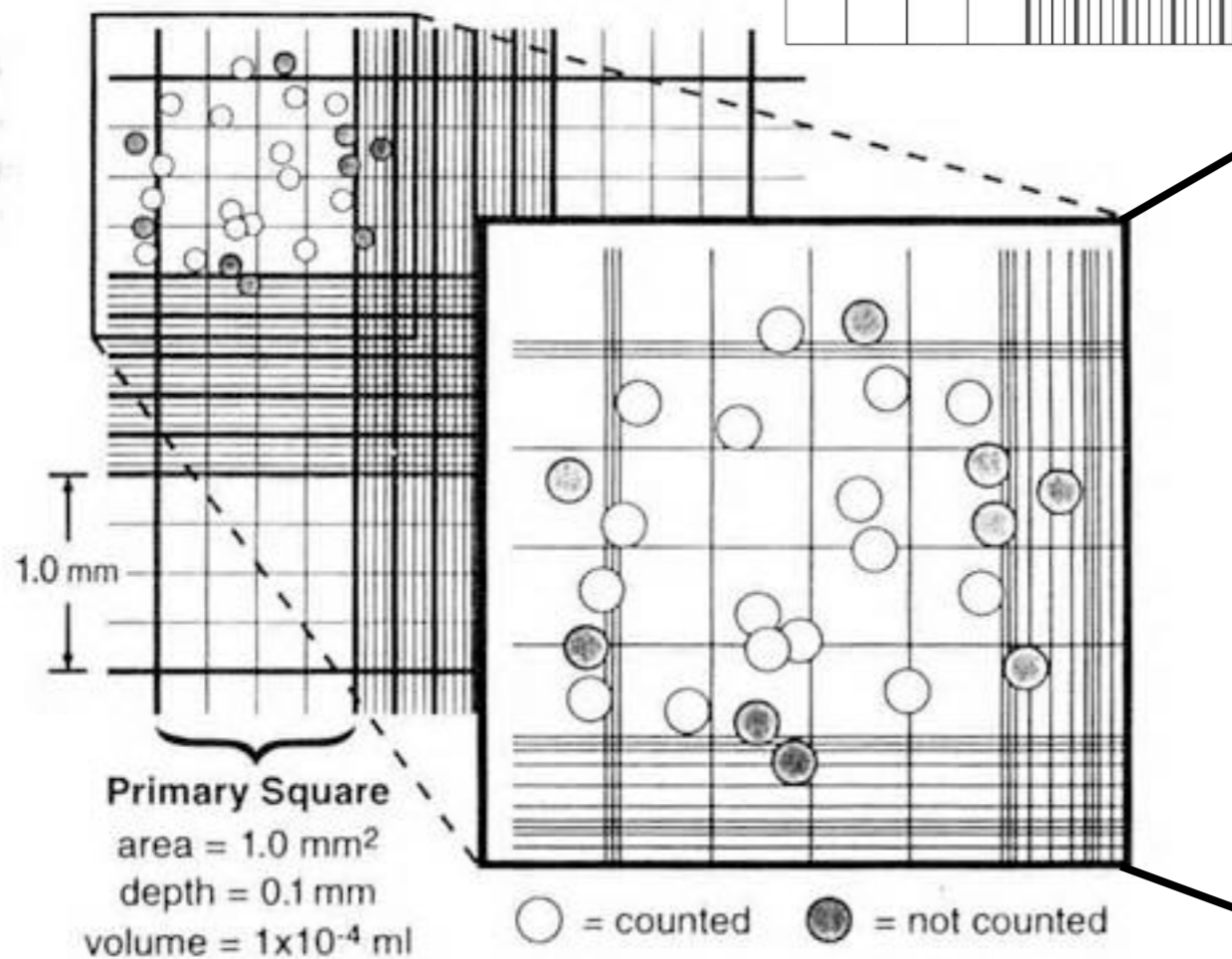**integrated implantable or injectable device**

Controlling design parameters:

How will we compare across conditions?

Hemocytometer

commons.wikimedia.org

1.0 mm

Primary Square
area = 1.0 mm^2
depth = 0.1 mm
volume = 1x10^{-4} ml

◯ = counted   ◉ = not counted

flexiblelearning.au.edu

Today in lab:

★Tissue culture primer: See me if you have not done mammalian tissue culture in the past.

★Experimental design:

1. Read/Skim 3+ papers to get ideas
   - Read abstract.
   - Skim methods.
   - Skim resuls/discussion -- summarize in 1-2 sentences.
   - Goal:

2. Make your plan!
   - Vary one parameter: simple or sophisticated
   - Check cell availability with teaching faculty
   - Request materials and/or equipment as needed
   - Goal: