# Cellular Environment Affects HDF Cell Proliferation, Proliferation Rate, and Attachment

2/13/08

# Objectives

- HDF Cells cultured in medium with higher concentrations of Fetal Bovine Serum (FBS) show increased proliferation and proliferation rates
- The presence of fibronectin(Fn) on surfaces increases the number of attached cells

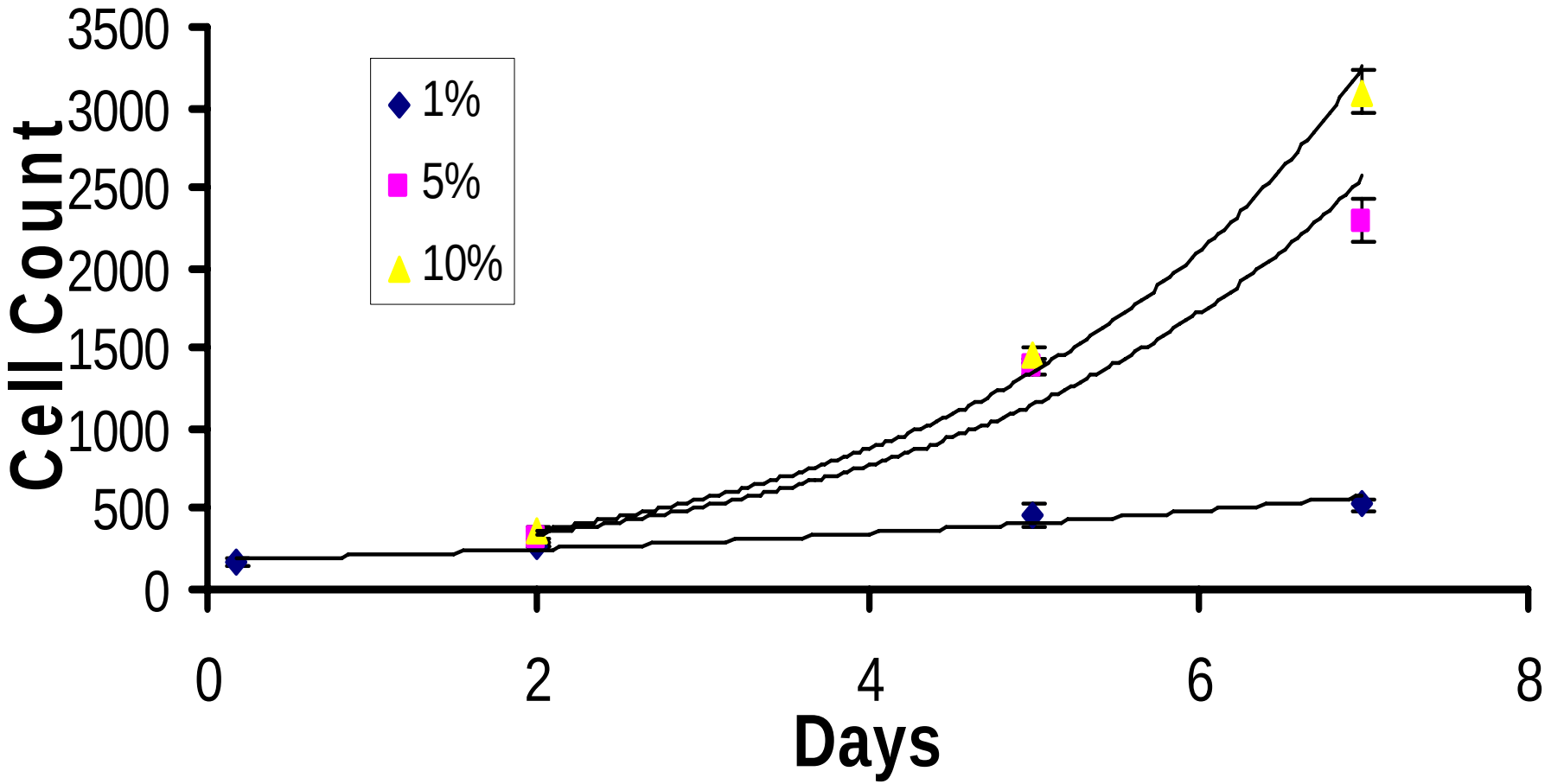
# Proliferation Assay

- Cells were seeded in wells containing media with 1%, 5%, and 10%  FBS
- At 4 hours, 2, 5, and 7 days, the cells were treated with trypsin and counted using a Coulter Counter
- The exponential fits to the proliferation curves can be modeled as: $N=N_0e^{kt,}$ and doubling time can be calculated as $t=.693/k$

# Anti-PCNA Staining and Fibronectin Attachment Assay

- Cells are seeded in media with 1%, 5% and 10% FBS for the Anti-PCNA
- Cells are treated with Hematoxylin to stain them blue and AEC, which stains nuclei cells in S1 phase red
- For the Fn attachment assay, cells were seeded on untreated surfaces or surfaces painted with Fn and incubated for 30 minutes
- Surfaces were then rinsed with PBS and viewed under a microscope to check for attachment

# 10% FBS Media Displays Highest Cell Proliferation Rate



# Cells Grown in 10% FBS Media doubled in 1.6 days

- Cell doubling times were calculated using the exponential fits:
  - 10% media: $y = 152e^{0.438x}$, $R^2 = 0.996$
    - Doubling Time: 1.6 days
  - 5%: $y = 155e^{0.4x}$, $R^2 = 0.975$
    - Doubling time: 1.7 Days
  - 1%: $y = 179e^{0.167}x$, $R^2 = 0.967$
    - Doubling time: 4.2 Days

# FBS Concentration increased percentage of cells in S1 phase

- 70% of cells in media with 10% FBS had red stained nuclei
- 40% in the 5% media had red nuclei
- 10% in 1% media stained red

# Fibronectin coating enhances cell attachment

- Wells that were uncoated with fibronectin showed no cell attachment

- Fn coated regions of wells showed an even distribution of flat, extended cells in every well.

# Proliferation

- The proliferation rate of cells in media with higher FBS concentrations is higher

- Cell populations in media with higher FBS concentrations had a higher percentage of cells in S1 Phase

# Attachment

- Cells do not attach to untreated wells
- Cells attach more rapidly to Fn treated wells
- More cells attach to Fn treated surfaces than untreated surfaces