Lecture #17: Microtechnology Tools I
Cell Spreading and Apoptosis

Chen+, Geometric Control of Cell Life and Death, Science 1997
Microcontact Printing

Kaufmann and Ravoo, Polym Chem, 2010
Non-Fouling Surfaces

Figure 3. Agarose and pluronic substrates fail by cell-independent mechanisms. (A) Phase contrast images of ECs on agarose substrates 1 day before (left) and 1 day after (right) agarose lifts off. The agarose film can be seen out of focus floating above the substrate (right). (B) Fluorescence images of fluorescent pluronic (green) and fibronectin (red) 1 day before and 1 day after L1s invade.

Culture on Patterned ‘Beads’

Fig. 1. Effect of cell spreading on apoptosis. (A) Combined phase contrast–fluorescence micrographs of capillary endothelial cells cultured in suspension in the absence or presence of different-sized microbeads or

Chen+, Geometric Control of Cell Life and Death, Science 1997
Apoptosis vs. Proliferation

Chen+, Geometric Control of Cell Life and Death, Science 1997
Well-Controlled Experiment

Chen+, Geometric Control of Cell Life and Death, Science 1997
Cell Shape Drives Differentiation

Rho/ROCK pathway

Rho A Induction

RV14- Active RhoA

RN19- Inactive/Blocks RhoA

Cell-Cell Contact vs. Growth

Observation:

What are the variables?

Rapid Growth

Slow Growth
Cell Spreading & Contact Are Convoluted

Decouple Cell Contact & Cell Spreading

Constrain Cell Spreading Precisely

Is It Contact or a Secreted Factor?

Brefeldin A & Monensin block protein secretion

LY294002 blocks PI3 kinase

Effects of Co-Culture

![Graph showing the effects of co-culture on albumin production over 14 days.](image)

- **Freshly Isolated Hepatocytes**
- **Pure Hepatocytes 2 weeks later**

**Albumin (µg/10^6 cells/hour)**

**Day**
Effects of Co-Culture

Albumin (µg/10^6 cells/hour)

Day

Freshly Isolated Hepatocytes

Co-Cultured Hepatocytes 2 weeks later
Photolithographic Micropatterning

Bhatia+, J Biomed Mat Res, 1997; Bhatia+, J of Biom Science, 1998; Bhatia+, Biotech Prog, 1998; Bhatia +, FASEB J, 1999; Khetani+, Hepatology,
Control of Cell-Cell Interactions

*Island size* 36 μm  100 μm  490 μm

Hepatocytes

Cocultures
Micropatterned Co-Cultures Reveal Role of Tissue Organization

Bhatia+ FASEB J, 1999; Khetani+ Hepatology, 2004
Reconfigurable Cell Culture

Hui and Bhatia, PNAS, 2007
DEP Cell Arraying

Immobilization in 3D by Photopolymerization

Immobilization in 3D by Photopolymerization

light

100 µm
3D Microenvironment

Effect of 3D cluster size on chondrocytes