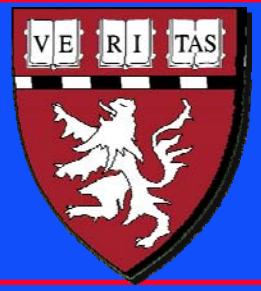

Massachusetts Institute of Technology
Harvard Medical School
Brigham and Women's Hospital
VA Boston Healthcare System




2.79J/3.96J/20.441/HST522J

# BIOMATERIALS-TISSUE INTERACTIONS:

## "Tools for Understanding the Molecular, Cellular, and Physiological Bases of the Tissue Response to Implants

M. Spector, Ph.D.

# BIOMATERIALS-TISSUE INTERACTIONS

**Tissue* + Biomaterial****

**Cell + Matrix****

* Structure comprising cells of the same type
** Solid surface

# CELL-MATRIX INTERACTIONS

**In Tissue**

**Cell + Extracellular Matrix**

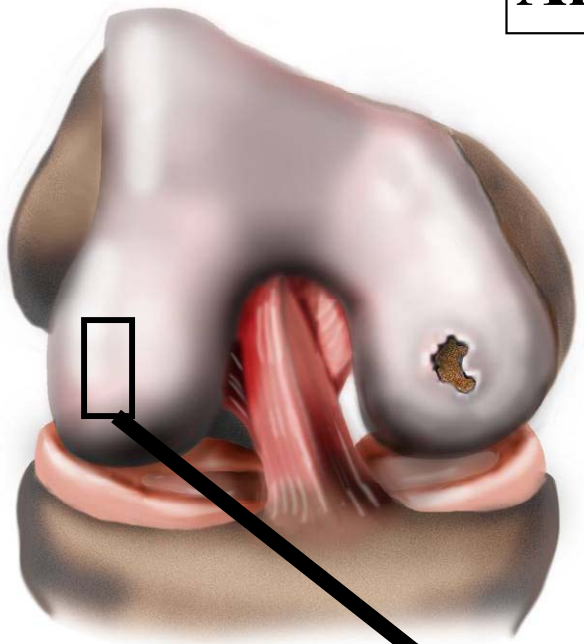
**In Tissue Engineering Scaffolds**

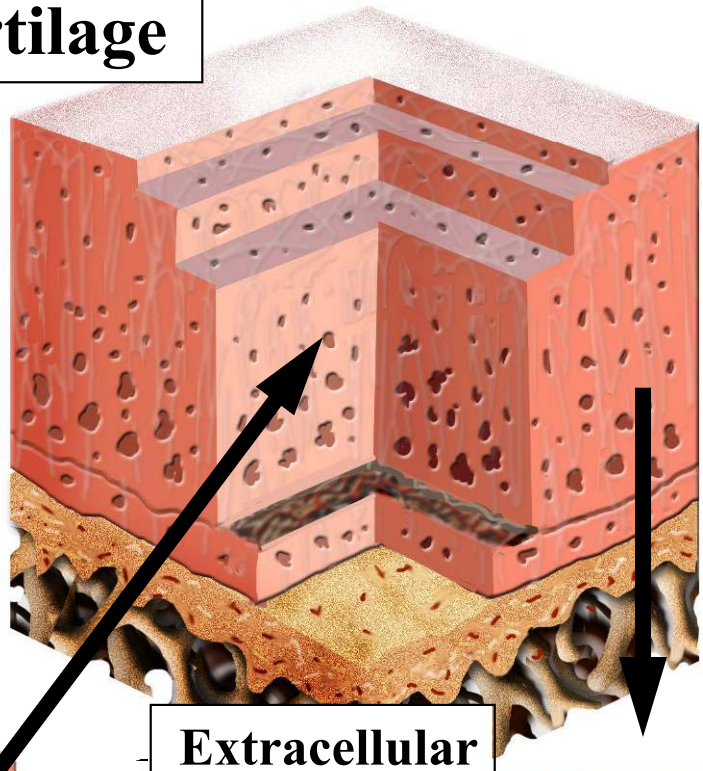
**Cell + Biomaterial Scaffold**

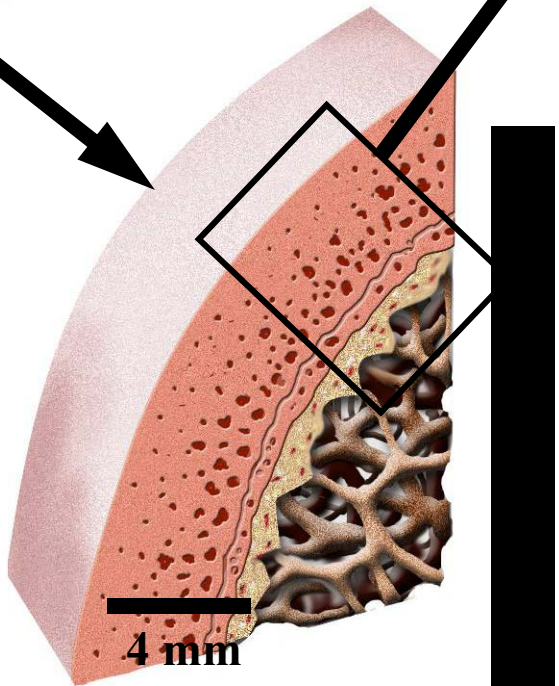
# CONCEPTS FOR UNDERSTANDING BIOMATERIALS-TISSUE INTERACTIONS

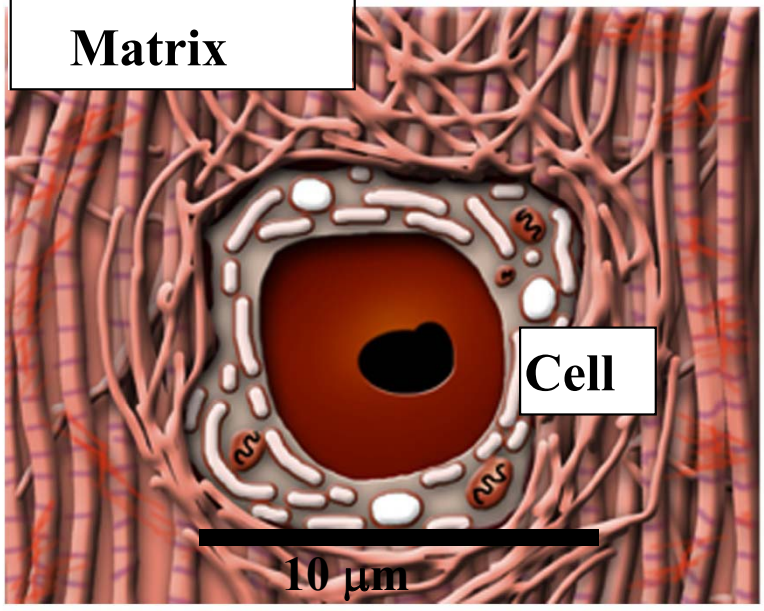
- **Control Volume**
- **Unit Cell Processes**
- **Types of Tissues**
- **Tissue Formation and Remodeling *In Vitro***
- **Wound Healing *In Vivo***

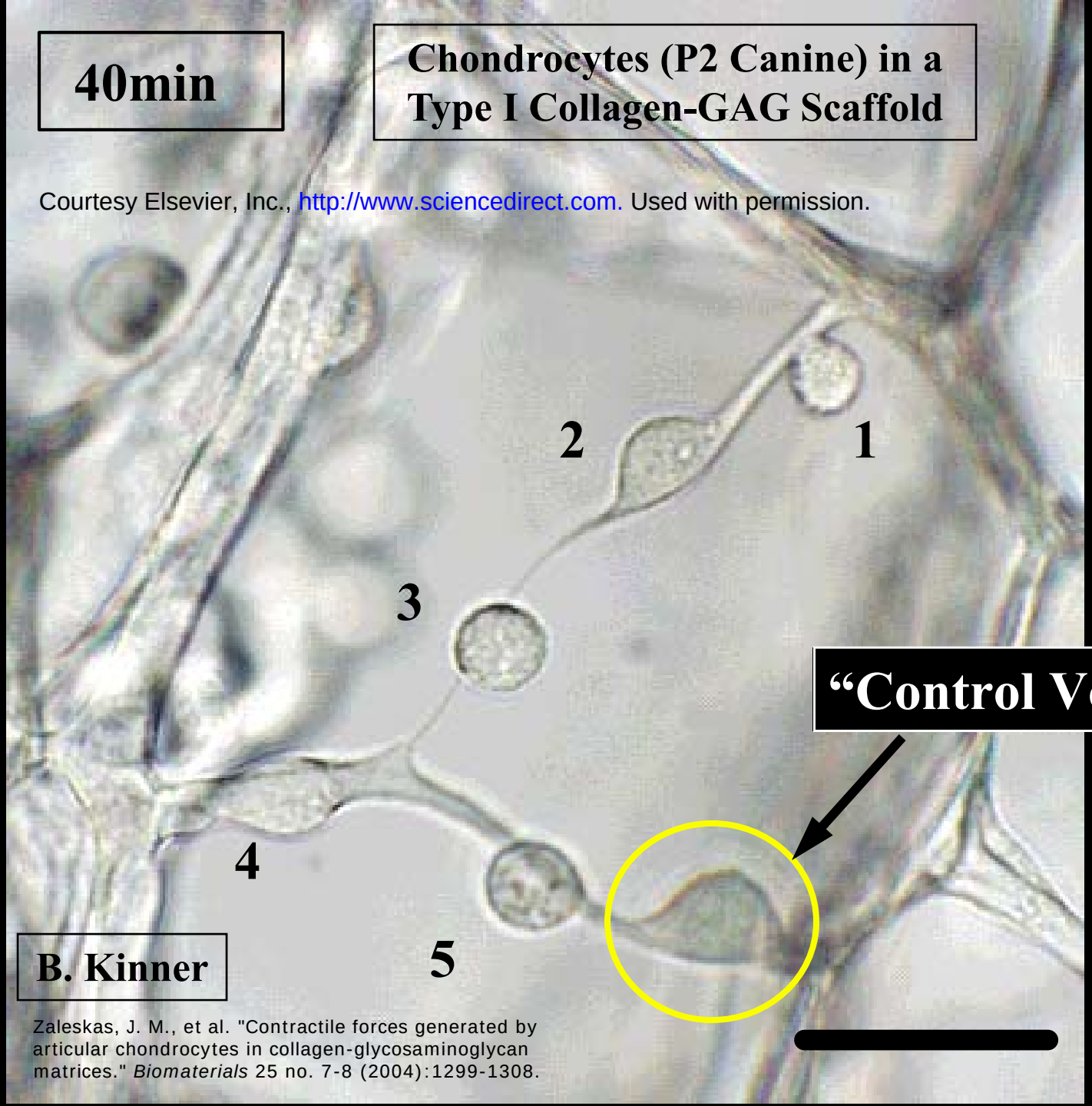
# Articular Cartilage



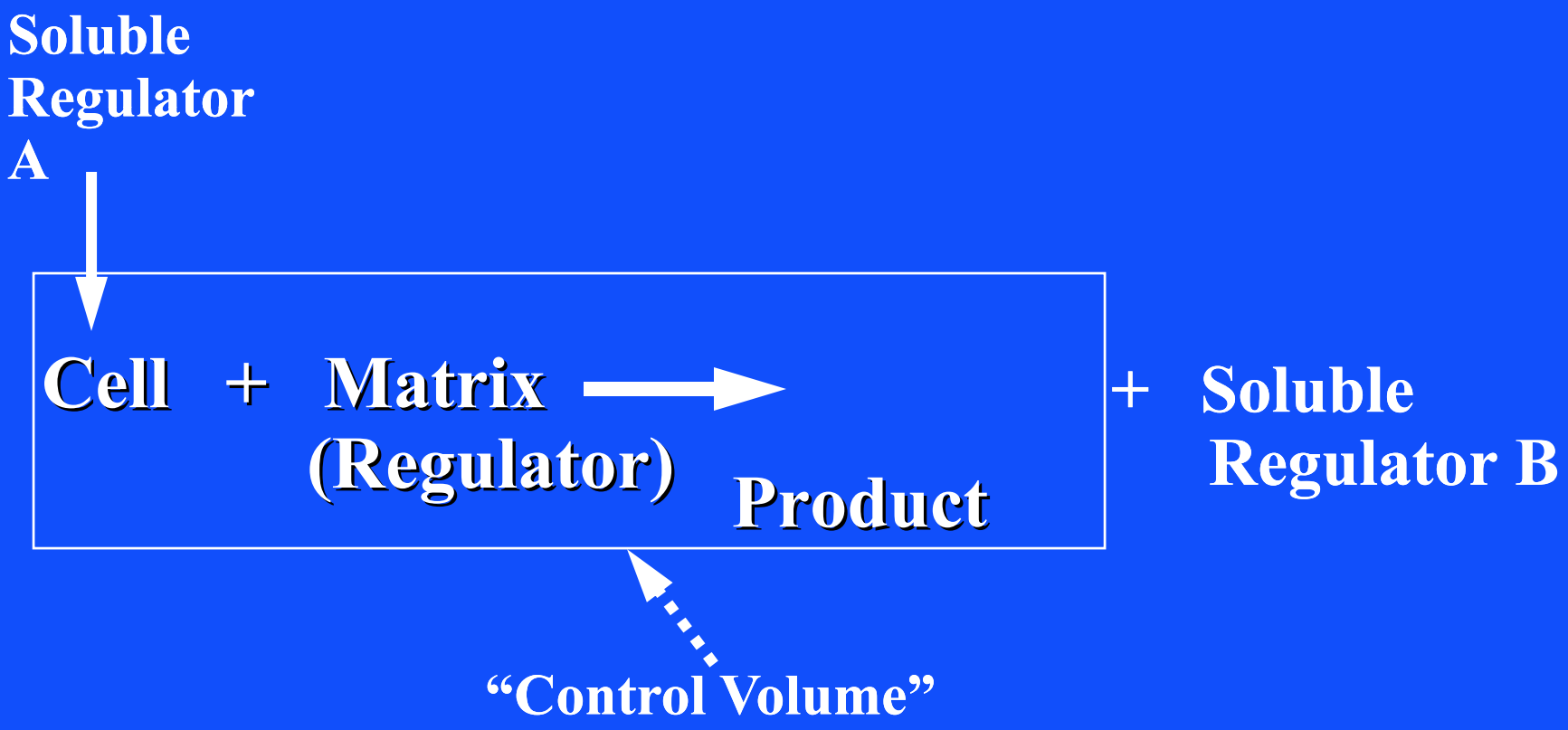





40min
Chondrocytes (P2 Canine) in a Type I Collagen-GAG Scaffold
Courtesy Elsevier, Inc., http://www.sciencedirect.com. Used with permission.
2
1
3
"Control Volume"
4
5
B. Kinner
Zaleskas, J. M., et al. "Contractile forces generated by articular chondrocytes in collagen-glycosaminoglycan matrices." *Biomaterials* 25 no. 7-8 (2004):1299-1308.

# UNIT CELL PROCESSES
## Concept of a "Control Volume" around a Cell



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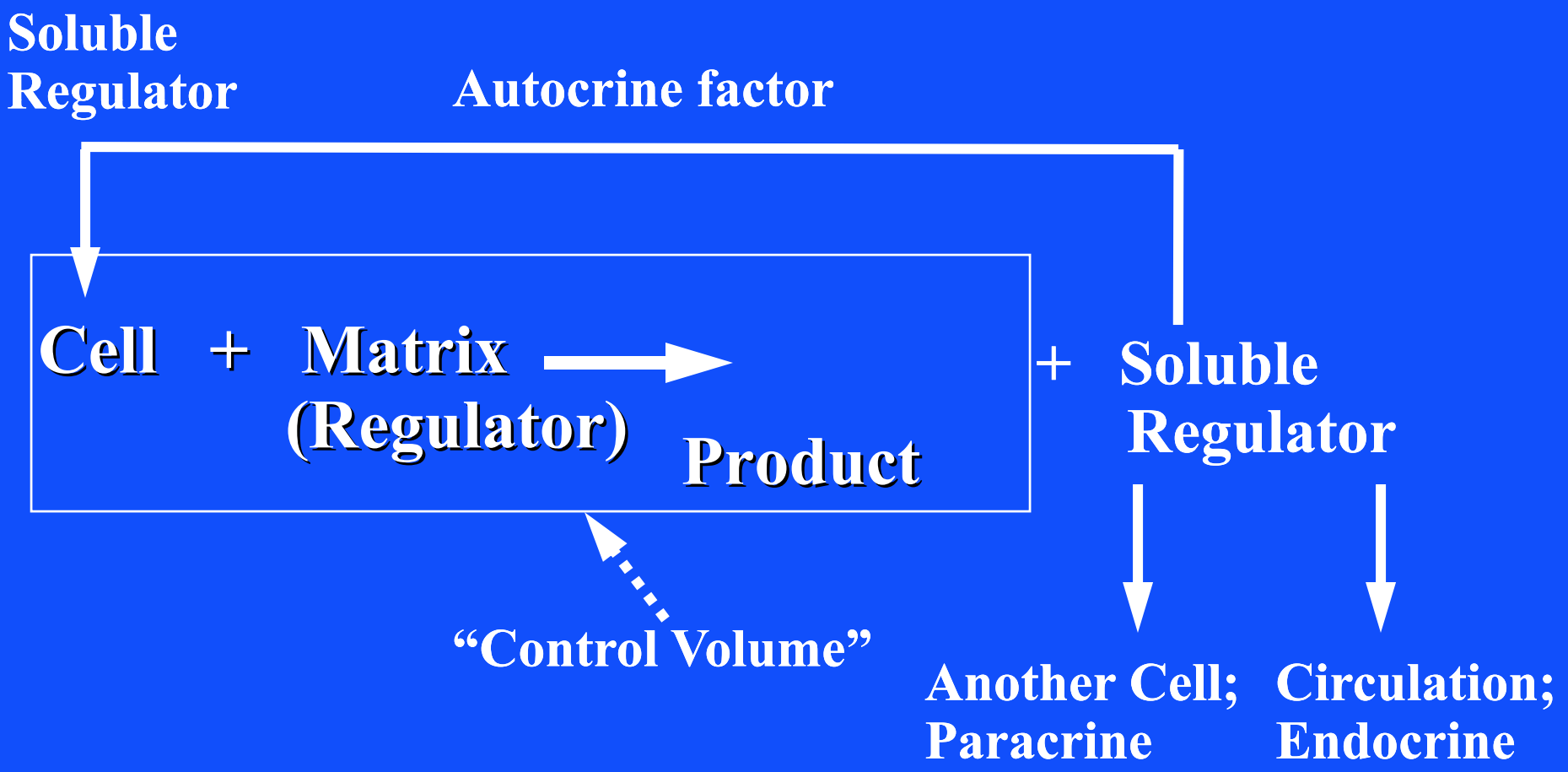

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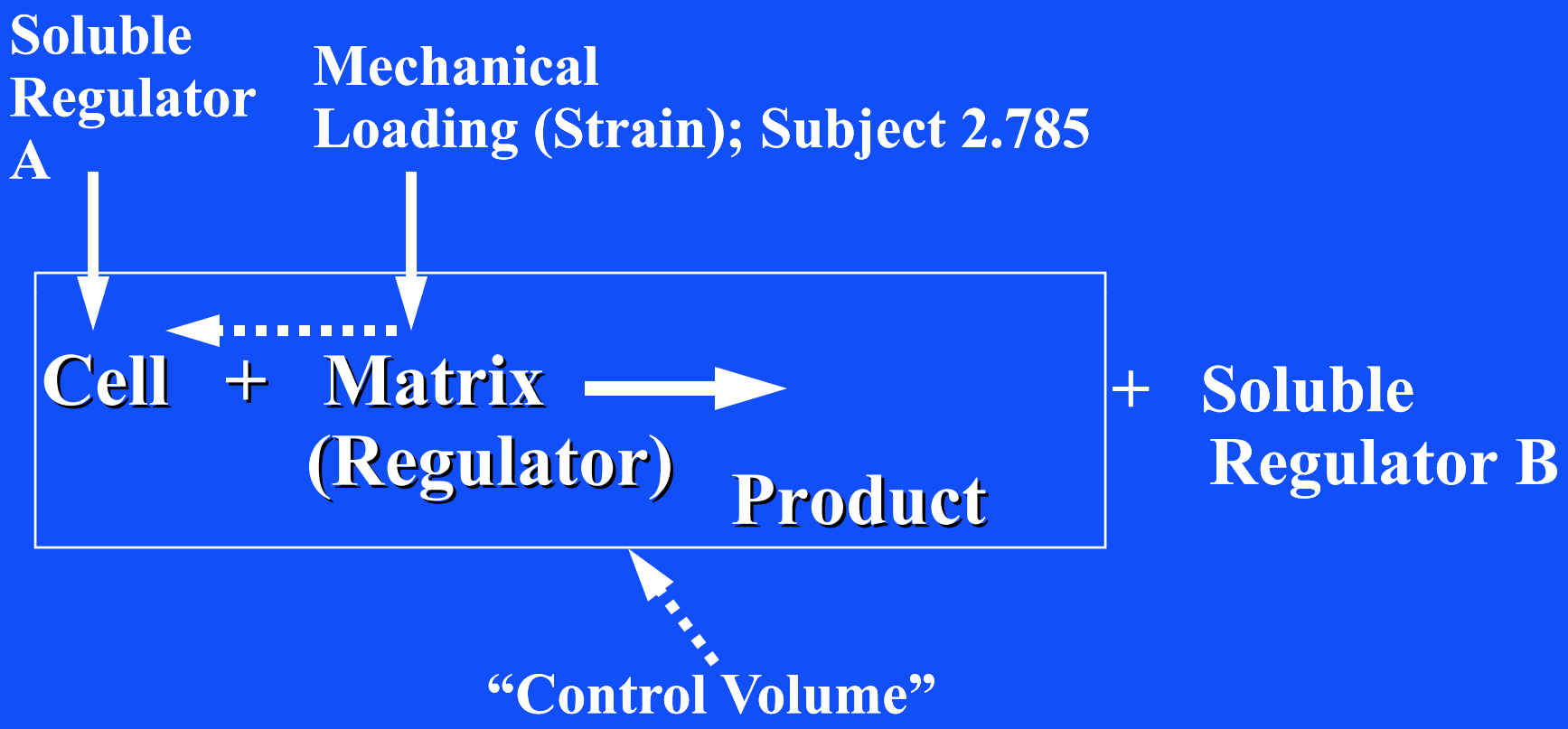
## Concept of a "Control Volume" around a Cell



# CONCEPTS FOR UNDERSTANDING BIOMATERIALS-TISSUE INTERACTIONS

- Control Volume
- Unit Cell Processes
- Types of Tissues
- Tissue Formation and Remodeling *In Vitro*
- Wound Healing *In Vivo*

# UNIT CELL PROCESSES

- Mitosis
- Migration
- Synthesis
- Contraction
- Endocytosis
- Exocytosis

# UNIT CELL PROCESSES

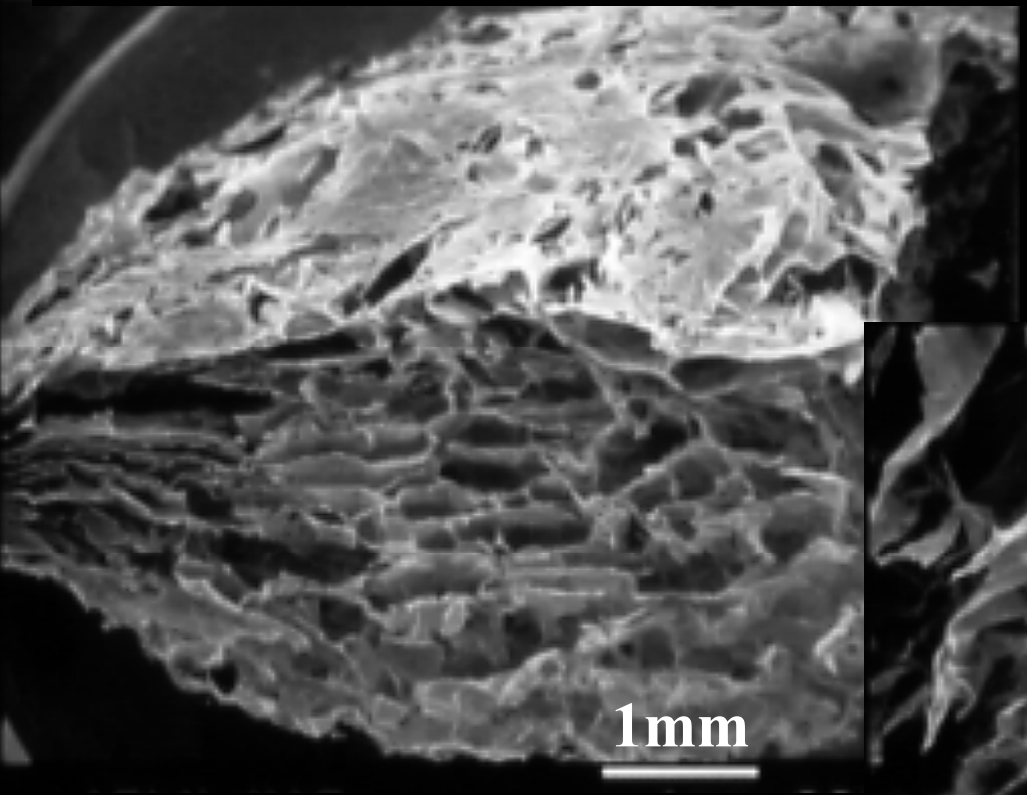
- Mitosis
- Migration
- Synthesis
- Contraction
- Endocytosis
- Exocytosis
- ?
- ?

# UNIT CELL PROCESSES

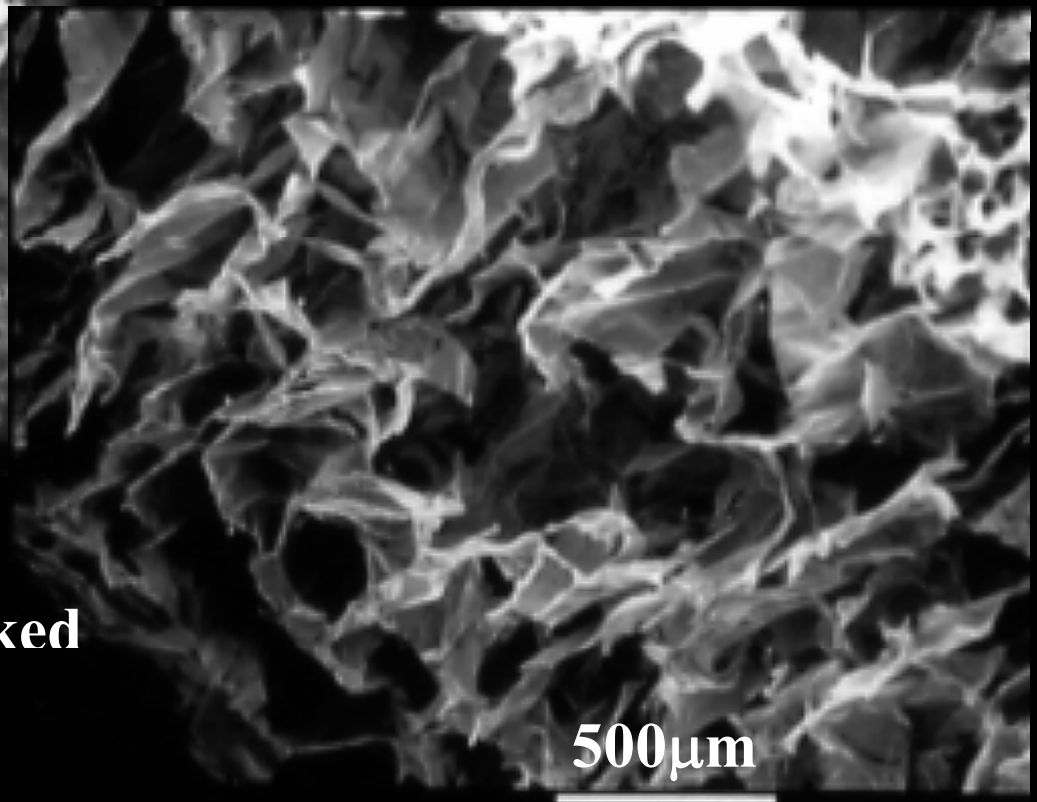
- Mitosis
- Migration
- Synthesis
- Contraction
- Endocytosis
- Exocytosis
- Apoptosis
- Differentiation

# COLLAGEN-GAG MATRICES: MODEL BIOMATERIALS (ANALOGS OF EXTRACELLULAR MATRIX)

## Investigation of cell interactions (UCPs) *in vitro*




- Type I (bovine and porcine)
- Type II (porcine)
- Chondroitin 6-sulfate



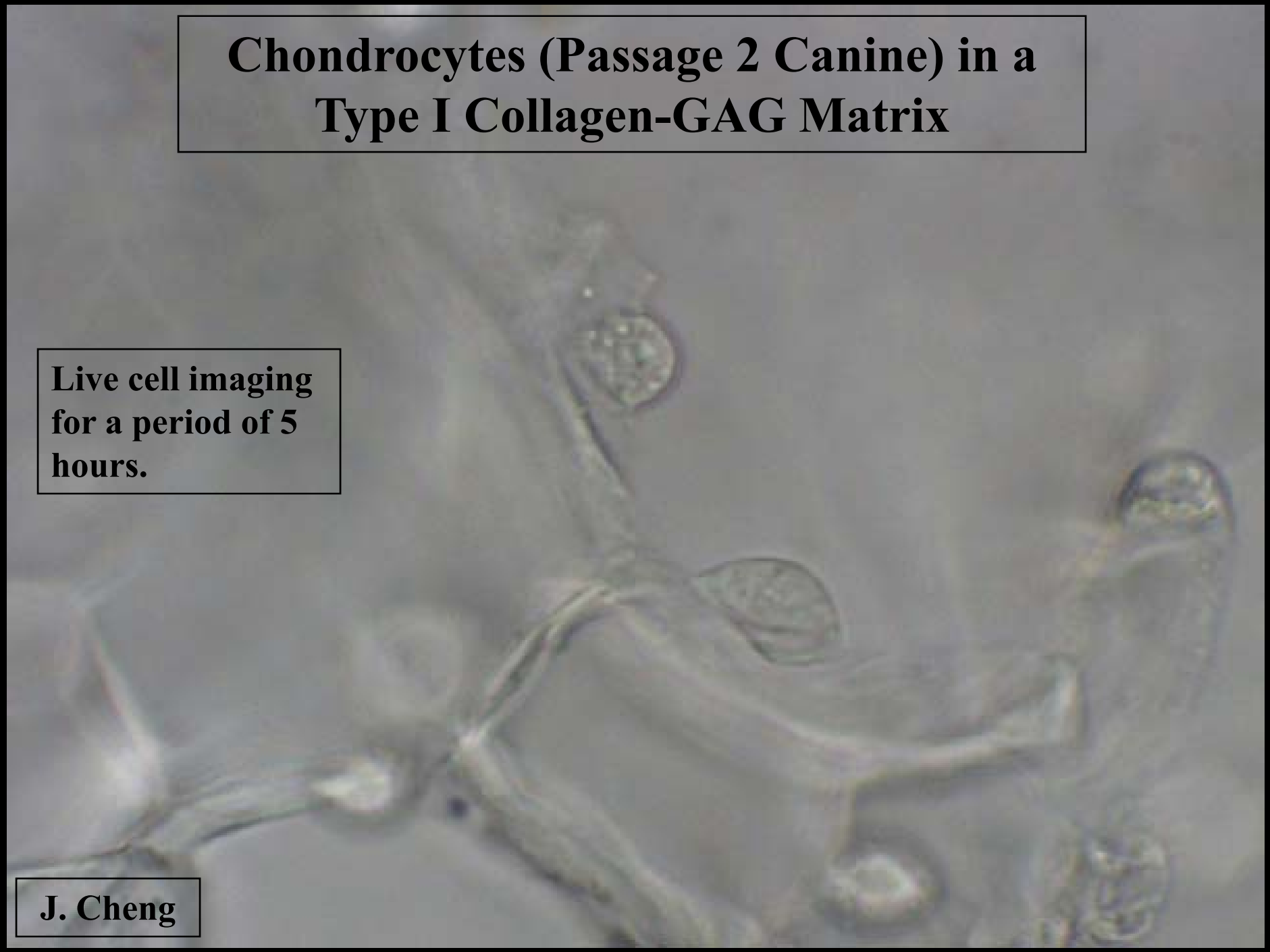

- Freeze-dried
- Dehydrothermally cross-linked
- Additional cross-linking

See IV Yannas, *et al*. PNAS, 1989

# CELL –MATRIX INTERACTIONS WITH COLLAGEN-GAG MATRICES *IN VITRO*

- **Can provide insights into interrelationships among cell processes.**
  - **How do mitosis and synthesis interrelate?**
  - **How do mitosis and synthesis relate to contraction?**
  - **How does migration relate to contraction?**
- **Can provide insights into cell behavior *in vivo*.**
- **Can provide insights into scaffold composition and structure for improved performance in regenerative medicine.**

# Chondrocytes (Passage 2 Canine) in a Type I Collagen-GAG Matrix



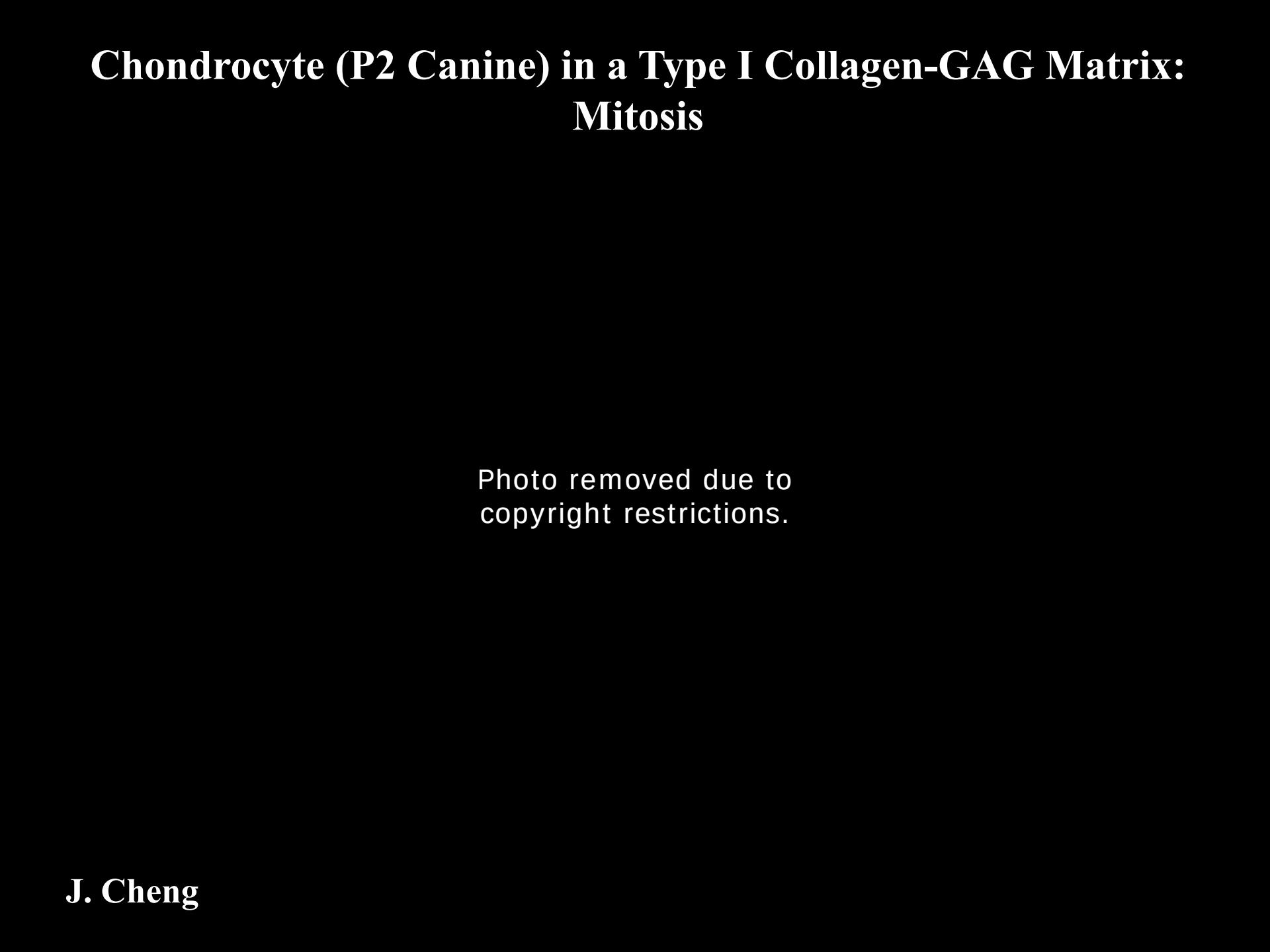
Live cell imaging for a period of 5 hours.

J. Cheng

# CELL –MATRIX INTERACTIONS

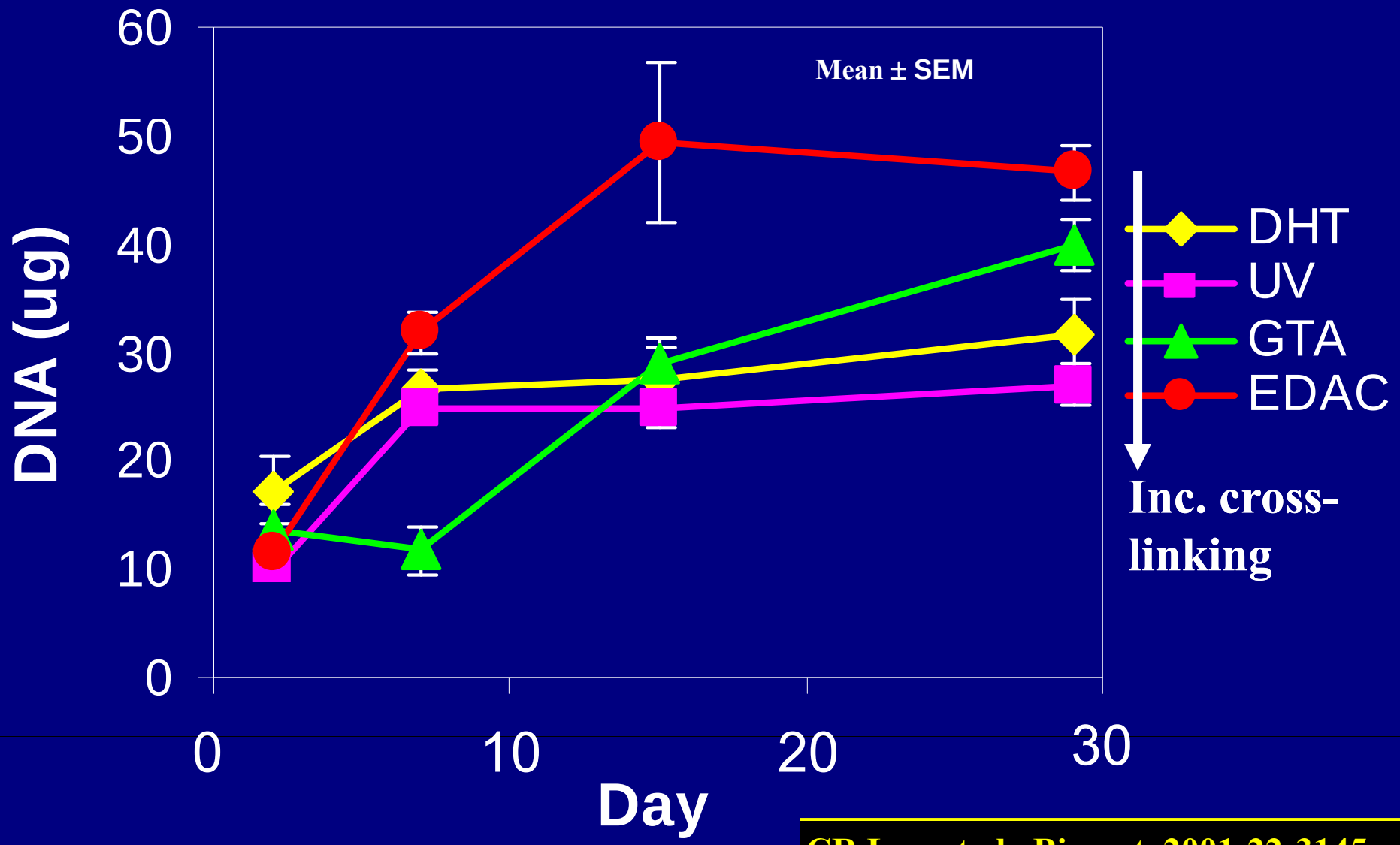
- Mitosis
- Migration
- Synthesis
- Contraction

# Chondrocyte (P2 Canine) in a Type I Collagen-GAG Matrix: Mitosis




J. Cheng

# Effects of Cross-Linking on Chondrocyte Proliferation in Collagen-GAG Matrices




CR Lee, et al., Biomat. 2001;22:3145

# CELL –MATRIX INTERACTIONS

- Mitosis
- Migration
- Synthesis
- Contraction

# Fibroblasts Migrate Away from Soft Substrates

**NIH 3T3 cells are plated on polyacrylamide substrates with a transition in flexibility. The soft side is marked with fluorescent beads (to the left). Cells turn to avoid the soft substrate as they approach the boundary from the stiff side, by retracting the leading lamellipodium that crossed the boundary.**



C.-M. Lo, *et al*., Biophys. J. 79:144 (2000)

# Fibroblasts Migrate Toward Stiff Substrates

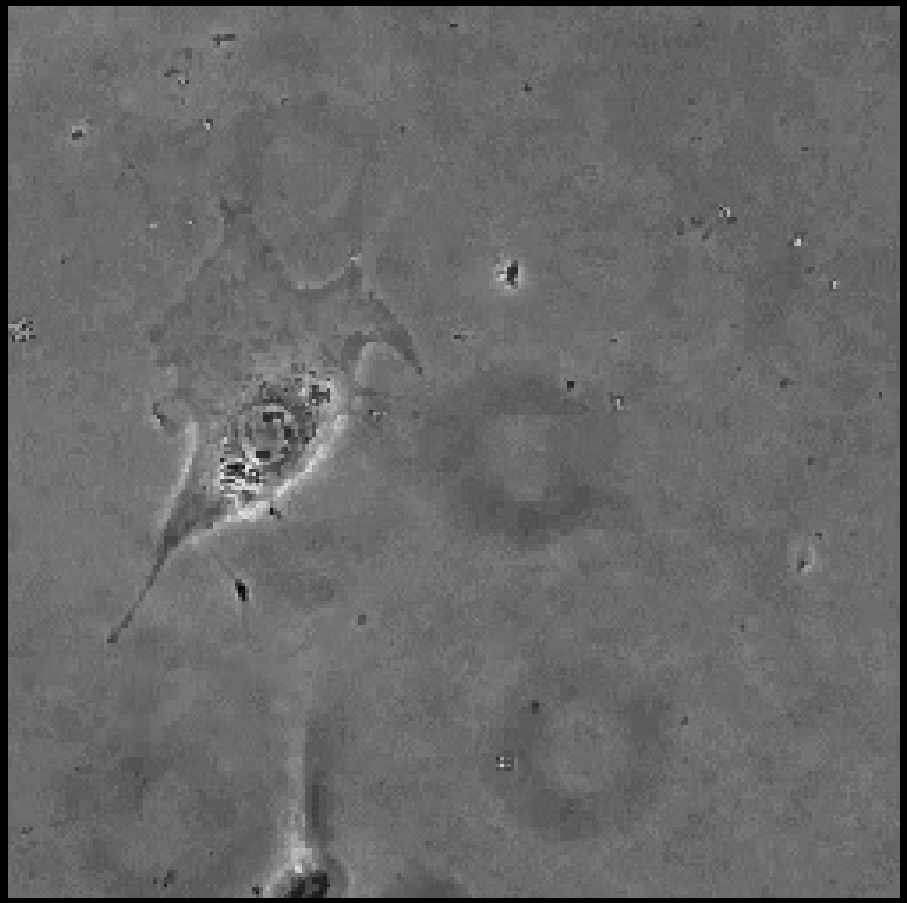
**NIH 3T3 cells are plated on polyacrylamide substrates with a transition in flexibility. The soft side is marked with fluorescent beads (to the left). Cells turn toward and enter the stiff side as they approach the boundary from the soft side, by expanding protrusions toward the boundary into a leading lamellipodium.**



C.-M. Lo, *et al*., Biophys. J. 79:144 (2000)

# Fibroblasts Migrate Toward Stretching Forces

**NIH 3T3 cells are plated on polyacrylamide substrates. Pulling forces are exerted by inserting a blunted needle in the substrate near the trailing end of the cell and dragging the needle away from the cell. Cells switch the direction of migration by expanding secondary protrusions toward the needle into a leading lamellipodium.**

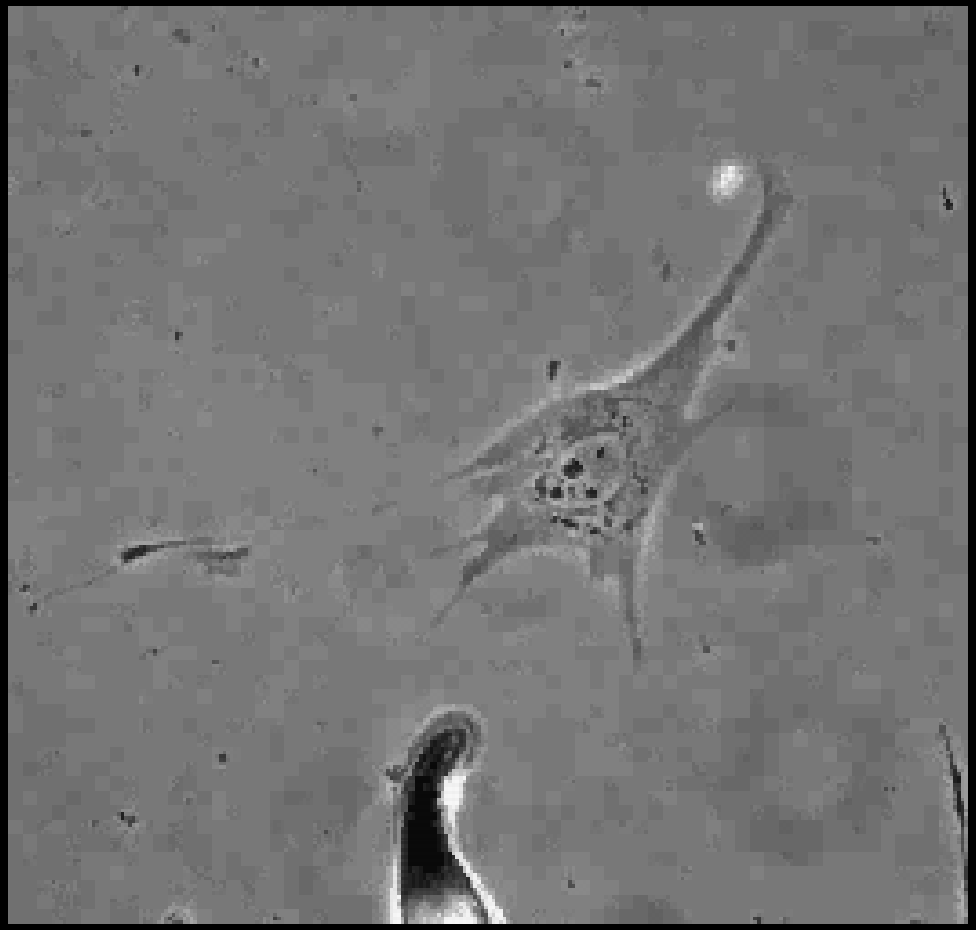




**C.-M. Lo, *et al*., Biophys. J. 79:144 (2000)**

# Fibroblasts Migrate Away from Compressing Forces

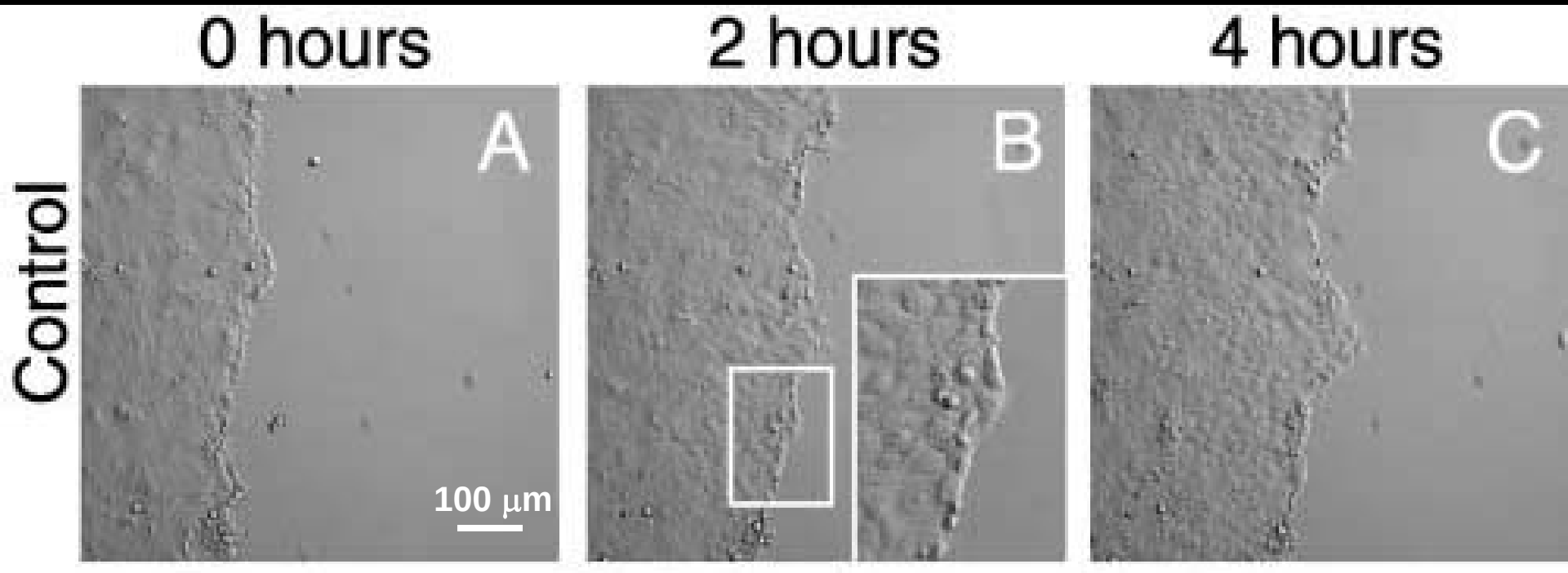
**NIH 3T3 cells are plated on polyacrylamide substrates. Pushing forces are exerted by inserting a blunted needle in the substrate near the leading edge of an approaching cell and moving the needle toward the cell. Cells switch the direction of migration by retracting the leading lamellipodium.**





**C.-M. Lo, *et al*., Biophys. J. 79:144 (2000)**

# Migration of Epithelial Cells *In Vitro* in a Wound Healing Assay





Published by The Rockefeller University Press.
http://dx.doi.org/10.1083/jcb.200104019

**Monolayers were ““wounded” by scraping.**

**LC Santy, J. Cell Biol. 154:599 (2001)**

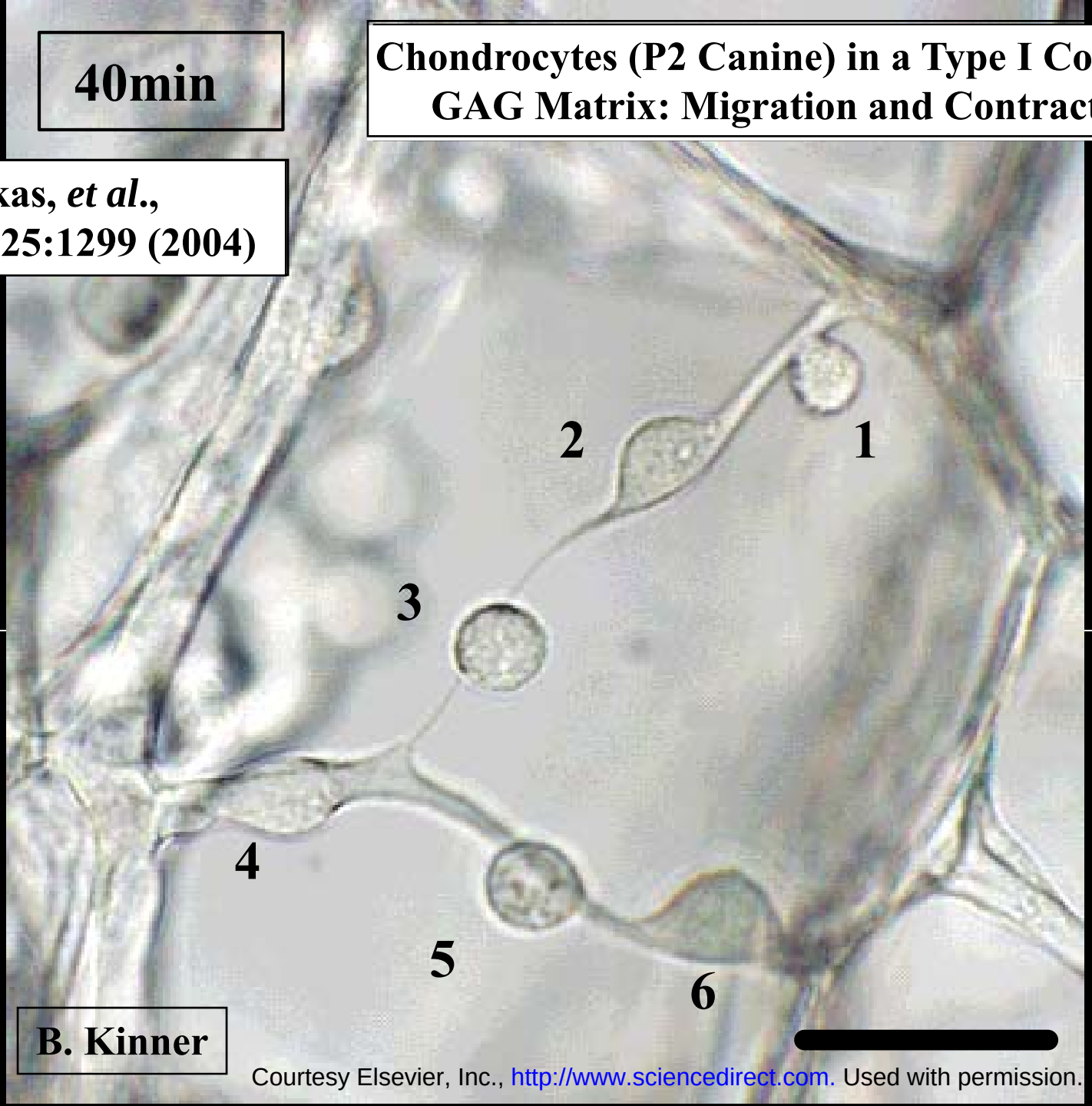


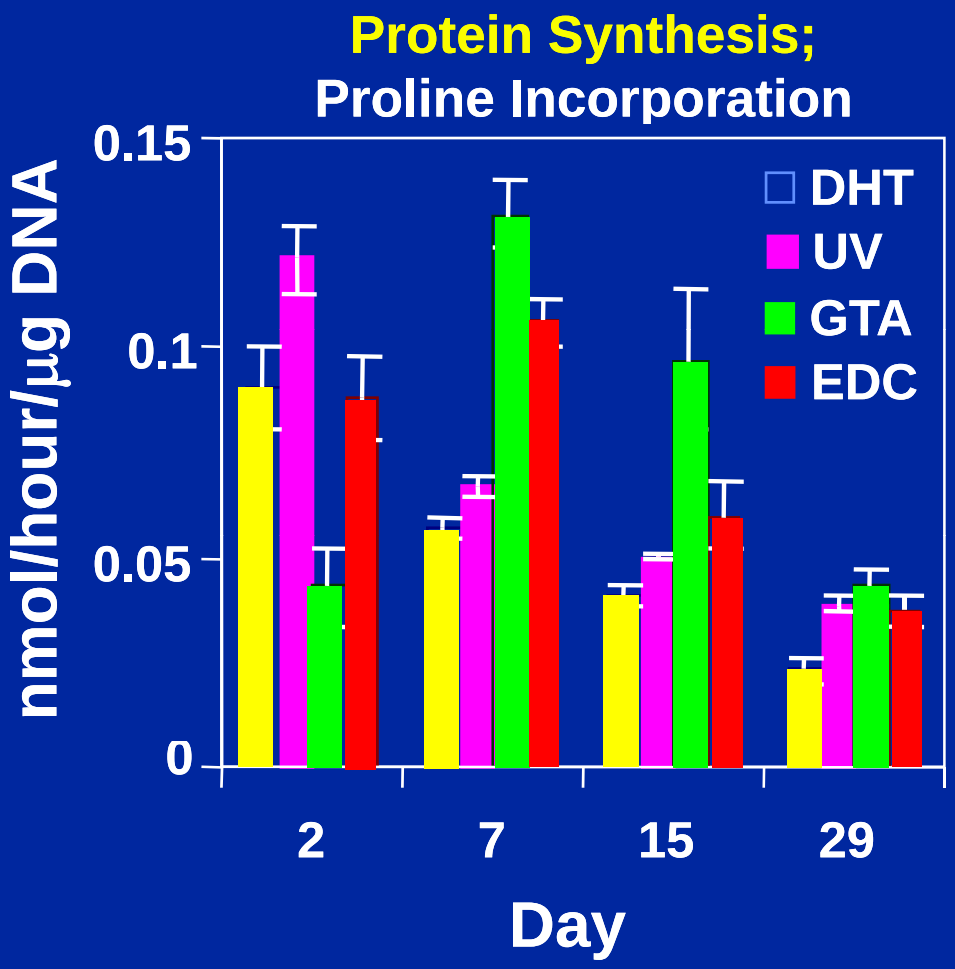
Diagram removed due to copyright restrictions.
Fig. 2 in Madri, Kidney Int. 41 (1992): 562.
Schematic of the modulation of microvascular endothelial cell phenotype during angiogenesis.

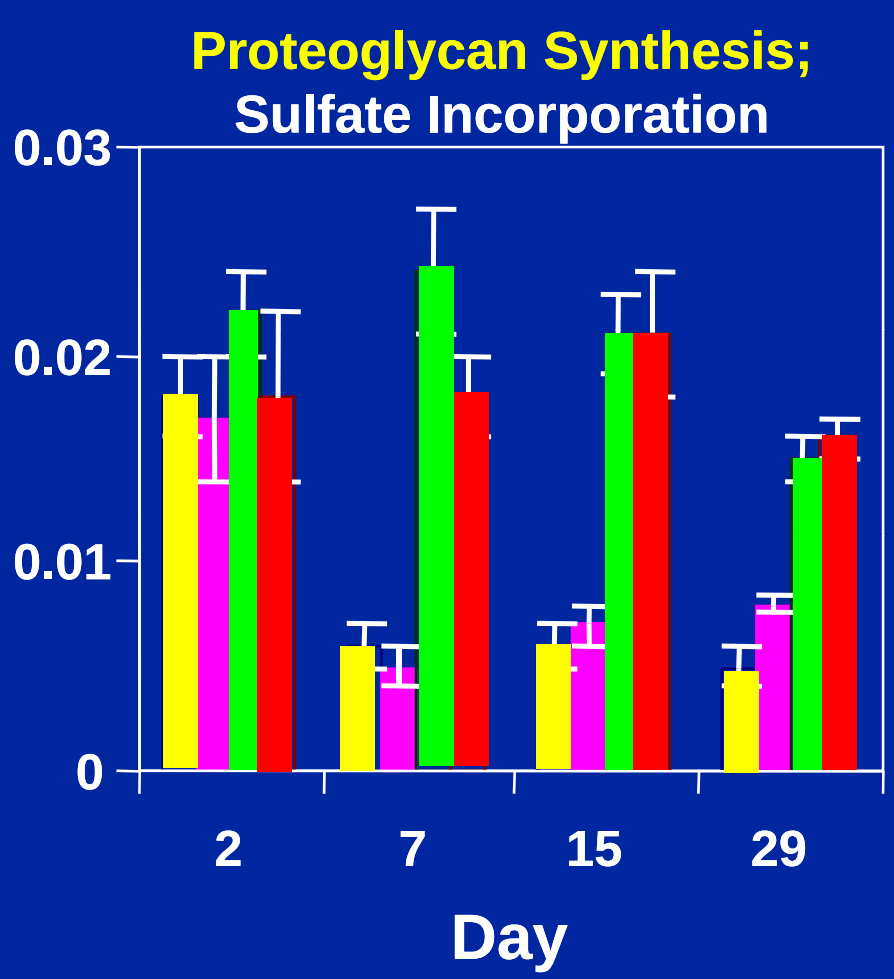
**Madri, Kidney Int. 41:562 (1992)**

# CELL –MATRIX INTERACTIONS

- Mitosis
- Migration
- Synthesis
- Contraction

# Effects of Cross-Linking on Chondrocyte Biosynthesis in Collagen-GAG Matrices




CR Lee, *et al*., Biomat. 2001;22:3145

# CELL –MATRIX INTERACTIONS

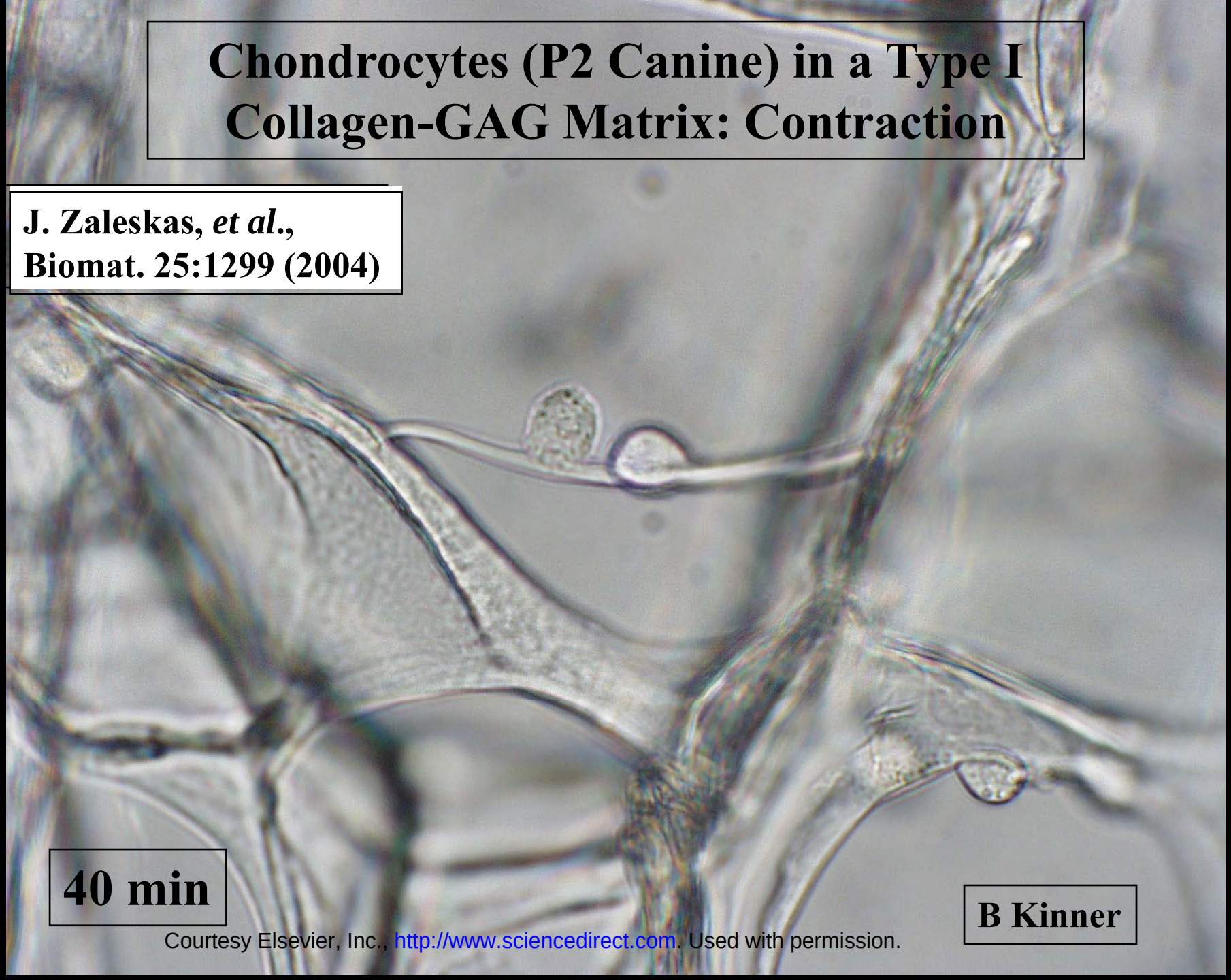
- Mitosis
- Migration
- Synthesis
- Contraction

**α-smooth muscle actin-fusion peptide (SMA-FP) inhibits the tension exerted by lung fibroblasts on silicone substrates. After washing our of the FP, cells contract again.**

See video at
http://jcb.rupress.org/content/suppl/2002/05/03/jcb.200201049.DC1/1.html

**Hinz B, *et al*., J Cell Biol 157:657 (2002)**

# Chondrocytes (P2 Canine) in a Type I Collagen-GAG Matrix: Contraction

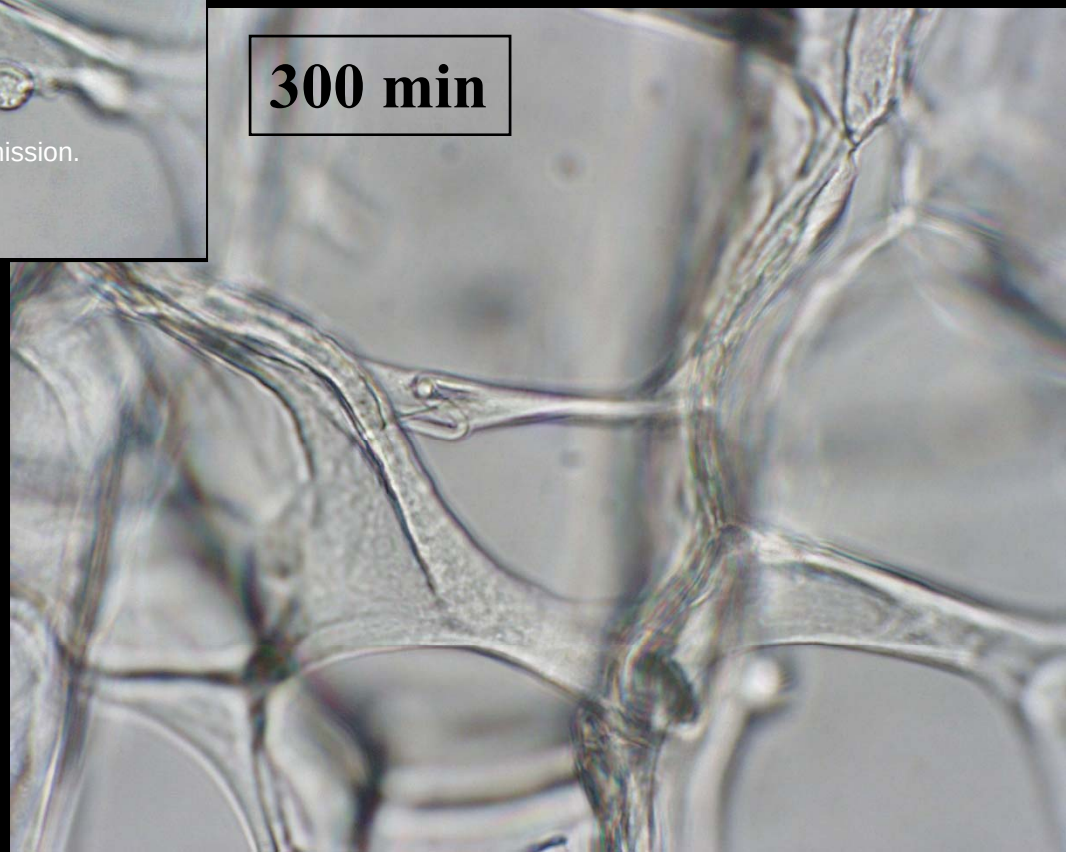


**J. Zaleskas, *et al*., Biomat. 25:1299 (2004)**

**40 min**

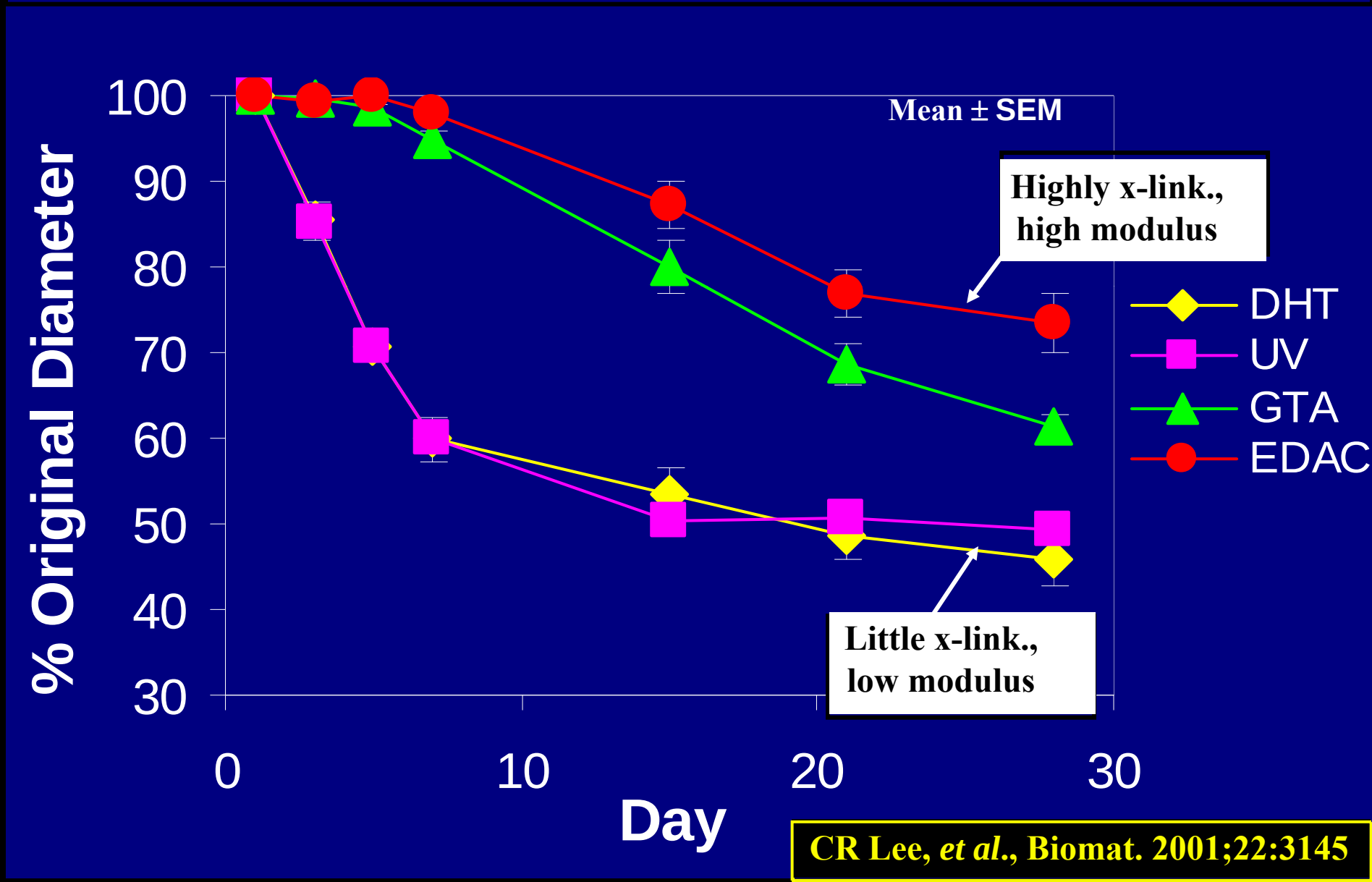
**B Kinner**




J. Zaleskas, *et al.*,
Biomat. 25:1299 (2004)

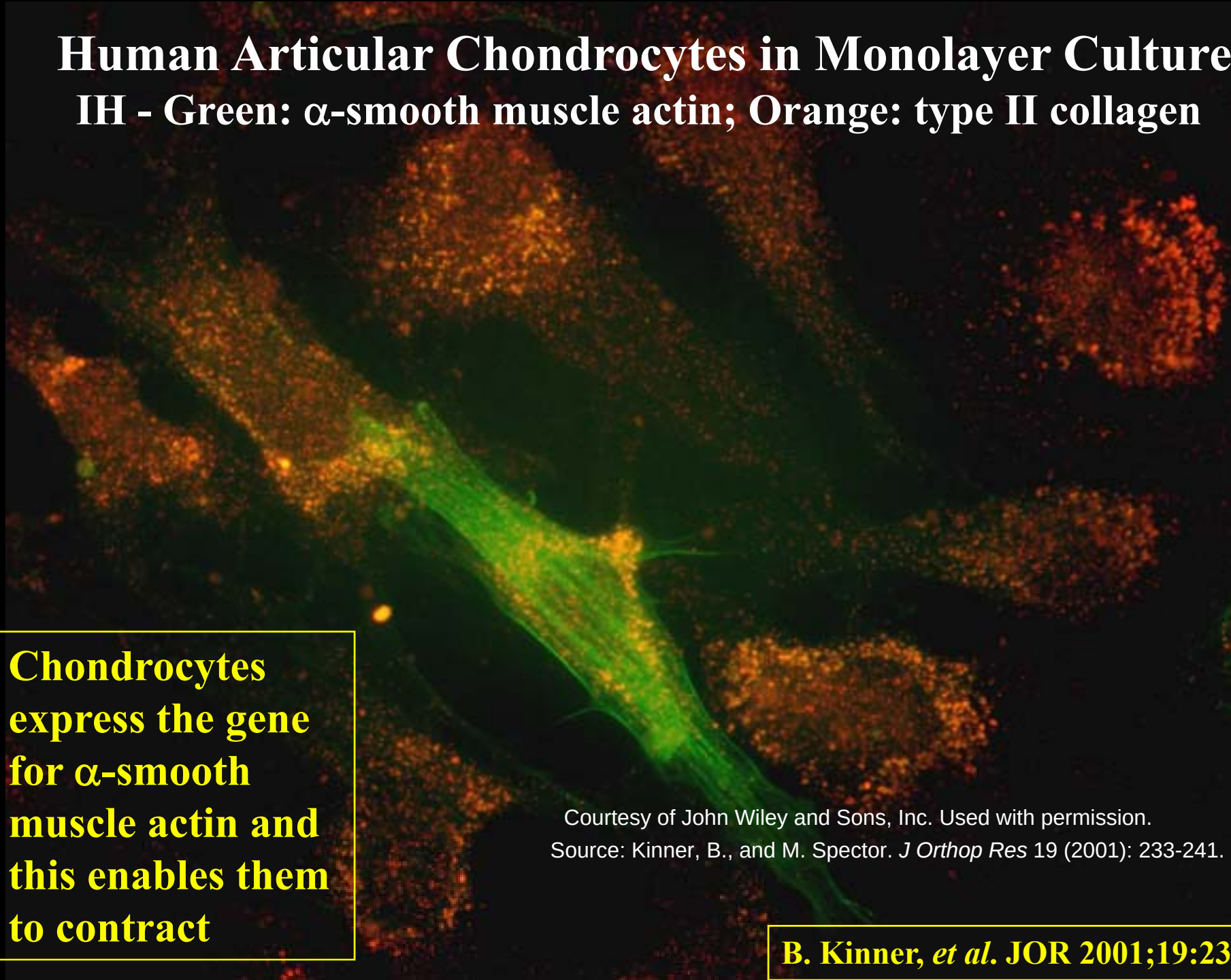
# Adult canine articular chondrocytes (passage 3) contract a type I collagen-GAG matrix, reflected in the decrease in diameter




CR Lee, *et al.*, Biomat. 2001;22:3145

# Human Articular Chondrocytes in Monolayer Culture

## IH - Green: α-smooth muscle actin; Orange: type II collagen

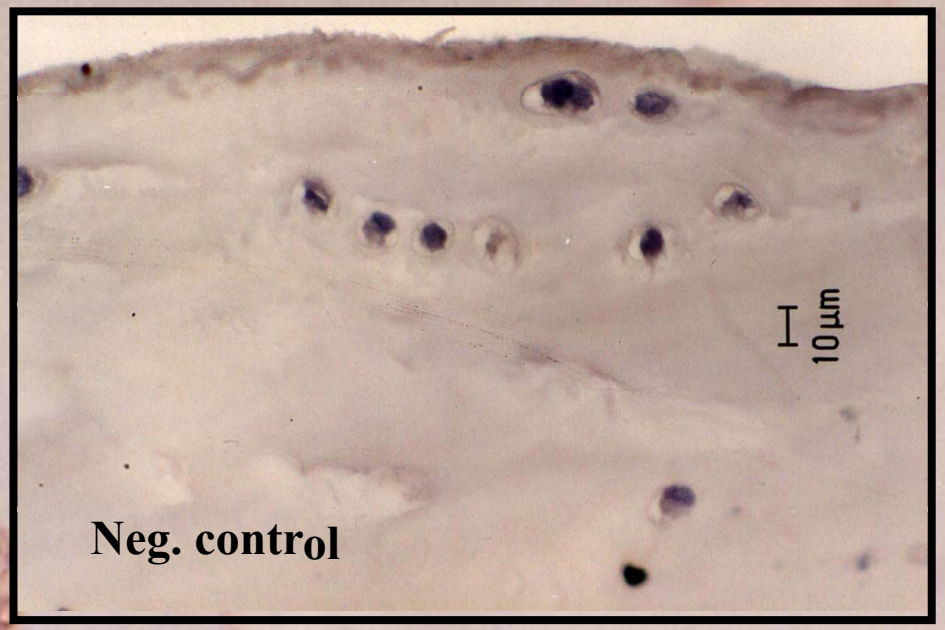


**Chondrocytes express the gene for α-smooth muscle actin and this enables them to contract**



**B. Kinner, *et al*. JOR 2001;19:233**

# α-Smooth Muscle Actin Immunohistochemistry of Human Articular Cartilage






**Kim and Spector, JOR 2000;18:749**

# MUSCULOSKELETAL CELLS THAT CAN EXPRESS α-SMOOTH MUSCLE ACTIN AND CAN CONTRACT

- Articular chondrocyte
- Osteoblast
- Meniscus fibroblast and fibrochondrocyte
- Intervertebral disc fibroblast and fibrochondrocyte
- Ligament fibroblast
- Tendon fibroblast
- Synovial cell
- Mesenchymal stem cell

M. Spector,
Wound Repair Regen.
9:11-18 (2001)

# POSSIBLE ROLES FOR α-SMOOTH MUSCLE ACTIN-ENABLED CONTRACTION

**Musculoskeletal Connective Tissue Cells**

| | |
|---|---|
| • Tissue engineering | Contracture of scaffolds |
| • Healing | Closure of wounds (skin wounds and bone fractures) |
| • Disease processes | Contracture (Dupuytren's) |
| • Tissue formation and remodeling | Modeling of ECM architecture (*e.g.*, crimp in ligament/tendon?) |

# CONCEPTS FOR UNDERSTANDING BIOMATERIALS-TISSUE INTERACTIONS

- Control Volume
- Unit Cell Processes
- Types of Tissues
- Tissue Formation and Remodeling *In Vitro*
- Wound Healing *In Vivo*

# TYPES OF TISSUES
## Which Tissues Can Regenerate Spontaneously?

| | Yes | No |
|---|---|---|
| **Connective Tissues** | | |
| • Bone | √ | |
| • Articular Cartilage, Ligament, Intervertebral Disc, Others | | √ |
| **Epithelia (e.g., epidermis)** | √ | |
| **Muscle** | | |
| • Cardiac, Skeletal | | √ |
| • Smooth | √ | |
| **Nerve** | | √ |

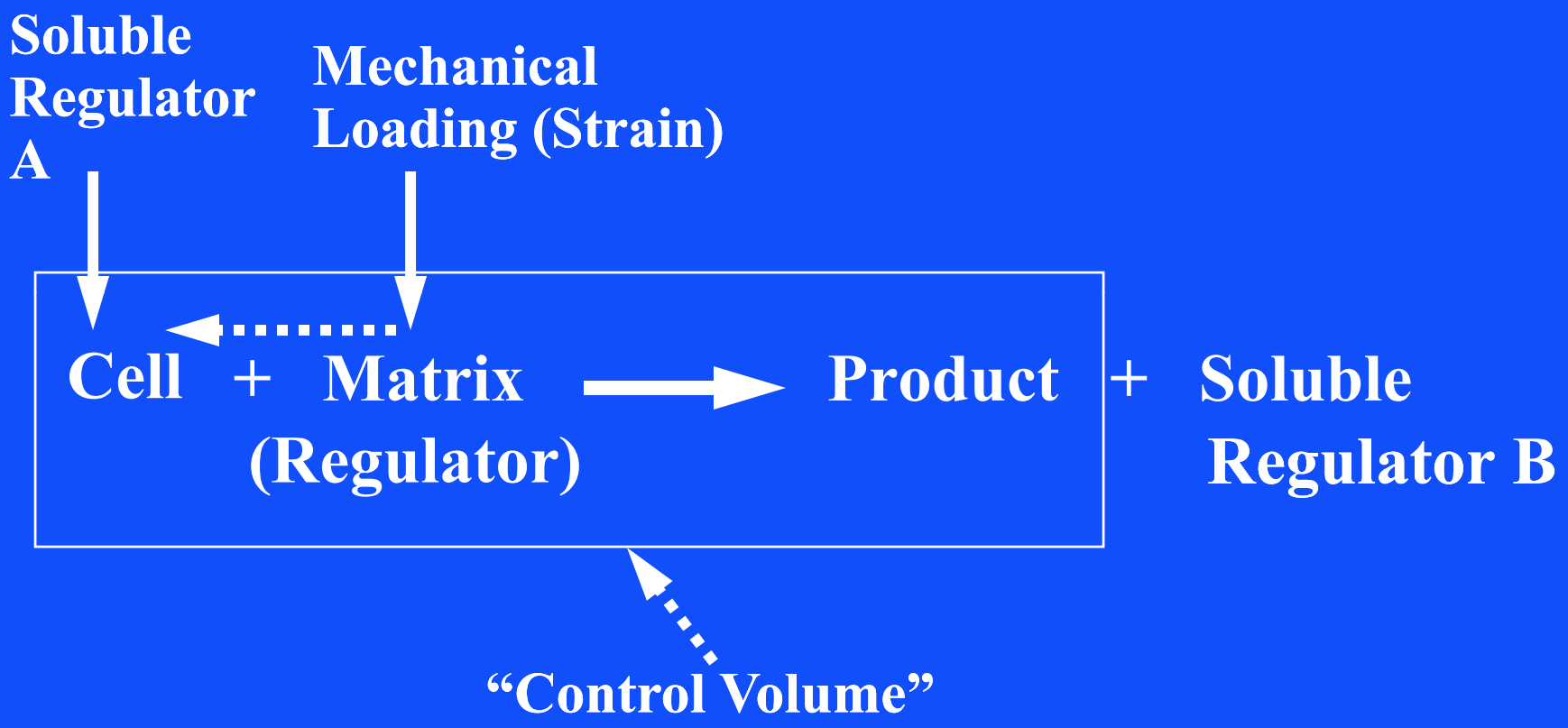
# BIOMATERIALS-TISSUE INTERACTIONS

## Cell + Matrix

Connective
Tissue
Epithelia
Muscle
Nerve

# UNIT CELL PROCESSES

## Concept of a "Control Volume" around a Cell



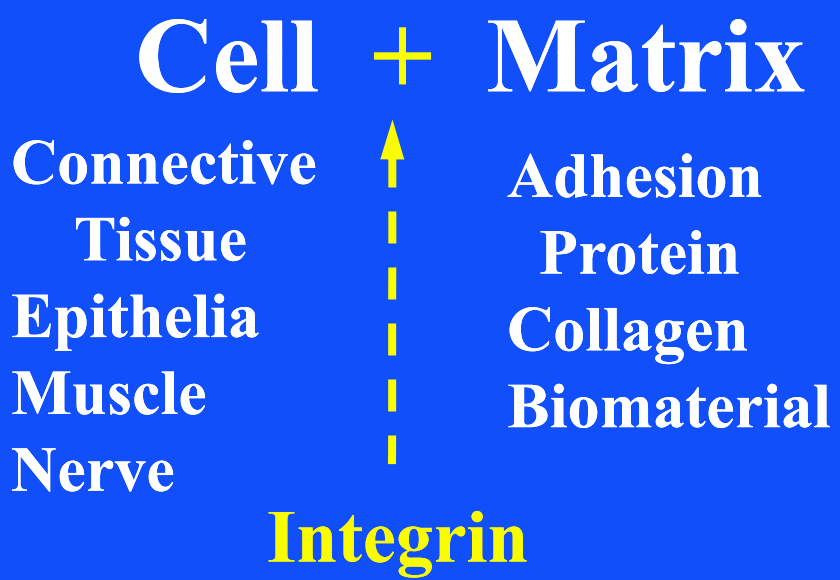
# BIOMATERIALS-TISSUE INTERACTIONS

## Cell + Matrix

Connective Tissue
Epithelia
Muscle
Nerve

Adhesion Protein
Collagen
Biomaterial

# BIOMATERIALS-TISSUE INTERACTIONS



# Chinese hamster ovary cell migration in a wound-healing assay

**CHO cells express the $\alpha 5\beta 1$ but not $\alpha 4\beta 1$ integrin**

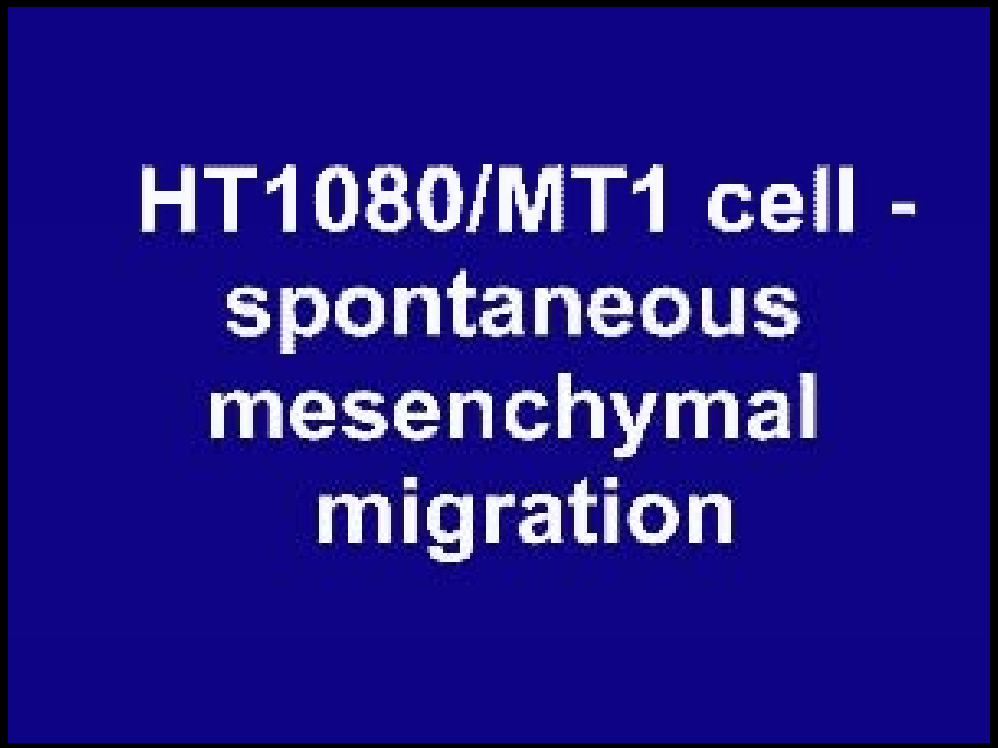
**C. Watters, Cell Biol. Ed. 2:210 (2003)**
**K.A. Pinco, Mol. Biol. Cell 13:3203 (2002)**

Photos removed due to copyright restrictions.
Figure 3 in Watters, C. *Cell Biol. Ed*. 2:210 (2003)

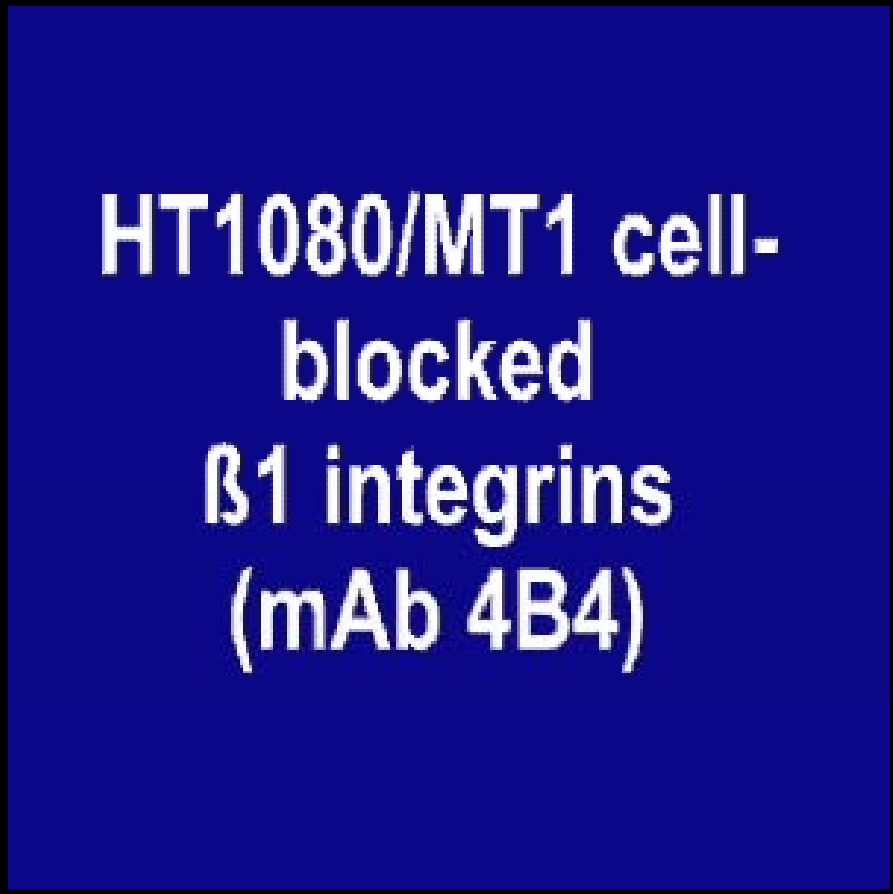
Image and links to associated videos at
http://www.ncbi.nlm.nih.gov/pmc/articles/PMC256980/figure/F3/

**Cells transfected with plasmid DNAs for $\alpha 4$ and $\alpha 4$/GFP**

# Migration of fibroblast-like fibrosarcoma cells in a 3-D collagen lattice




K. Wolf, *et al*., J. Cell Biol. 160:267 (2003)




**Reduction of migration speed and induction of detached, nonmobile spherical morphology by adhesion perturbing anti–α1 integrin antibody, as a consequence of impaired collagen fibril binding.**

# "UNIT CELL PROCESSES"

**Cell + Matrix** —**UCP**→

**Connective Tissue**
**Epithelia**
**Muscle**
**Nerve**

**Mitosis**
**Synthesis**
**Migration**
**Contraction**
**Endocytosis**
**Exocytosis**

# "UNIT CELL PROCESSES"

**Cell + Matrix** —UCP→ **Product**

| | | |
|---|---|---|
| Connective Tissue | Mitosis | Cell proliferation |
| Epithelia | Synthesis | Matrix molecules, enzymes, cytokines |
| Muscle | Migration | Translocation |
| Nerve | Contraction | Strain |
| | Endocytosis | Solubilized fragments |
| | Exocytosis | Regulators |

# "UNIT CELL PROCESSES"

**Regulator**

**UCP**

**Cell + Matrix → Product + Regulator**

**Connective Tissue**
**Epithelia**
**Muscle**
**Nerve**

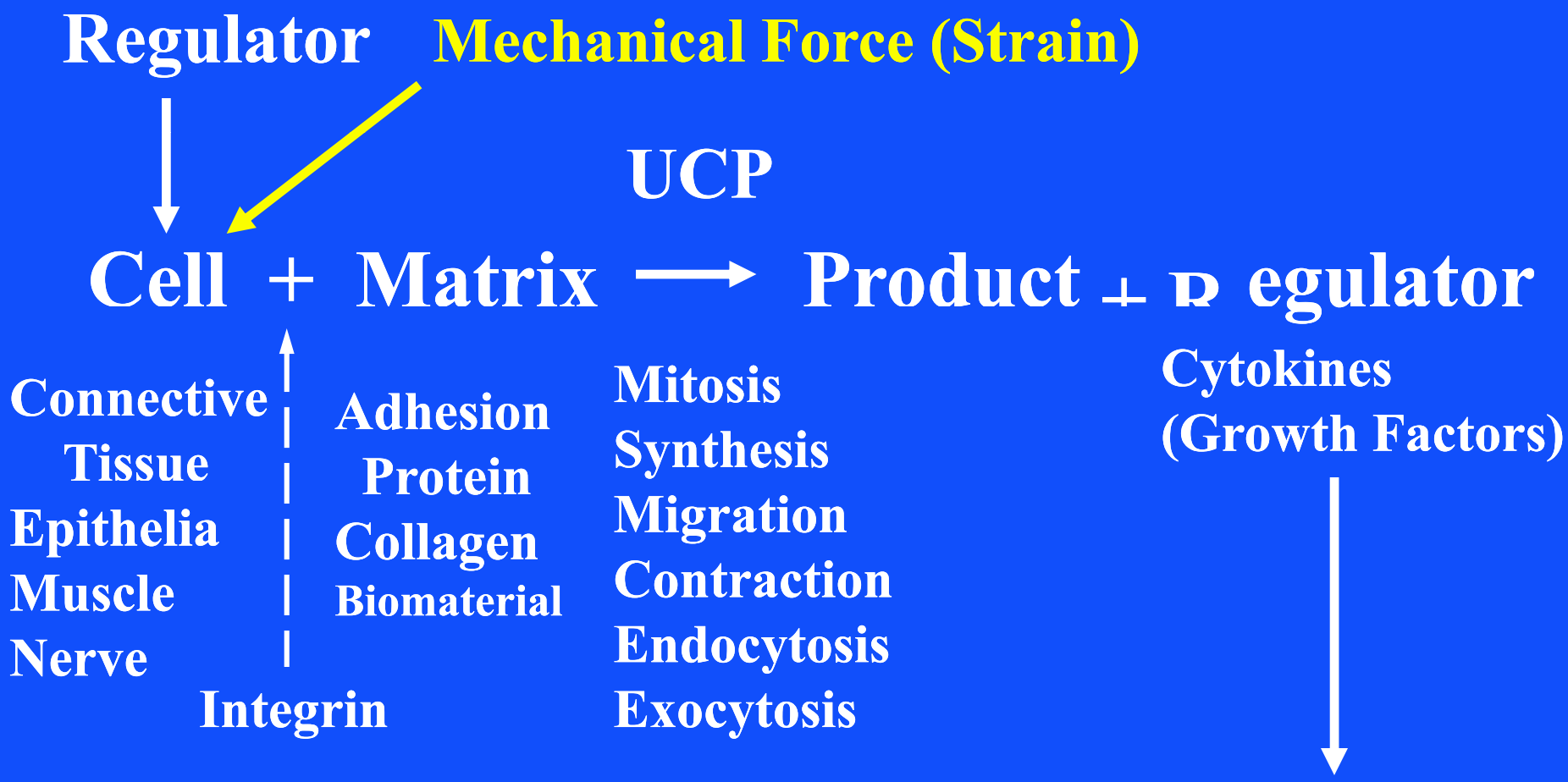
**Mitosis**
**Synthesis**
**Migration**
**Contraction**
**Endocytosis**
**Exocytosis**

**Cytokines (Growth Factors)**

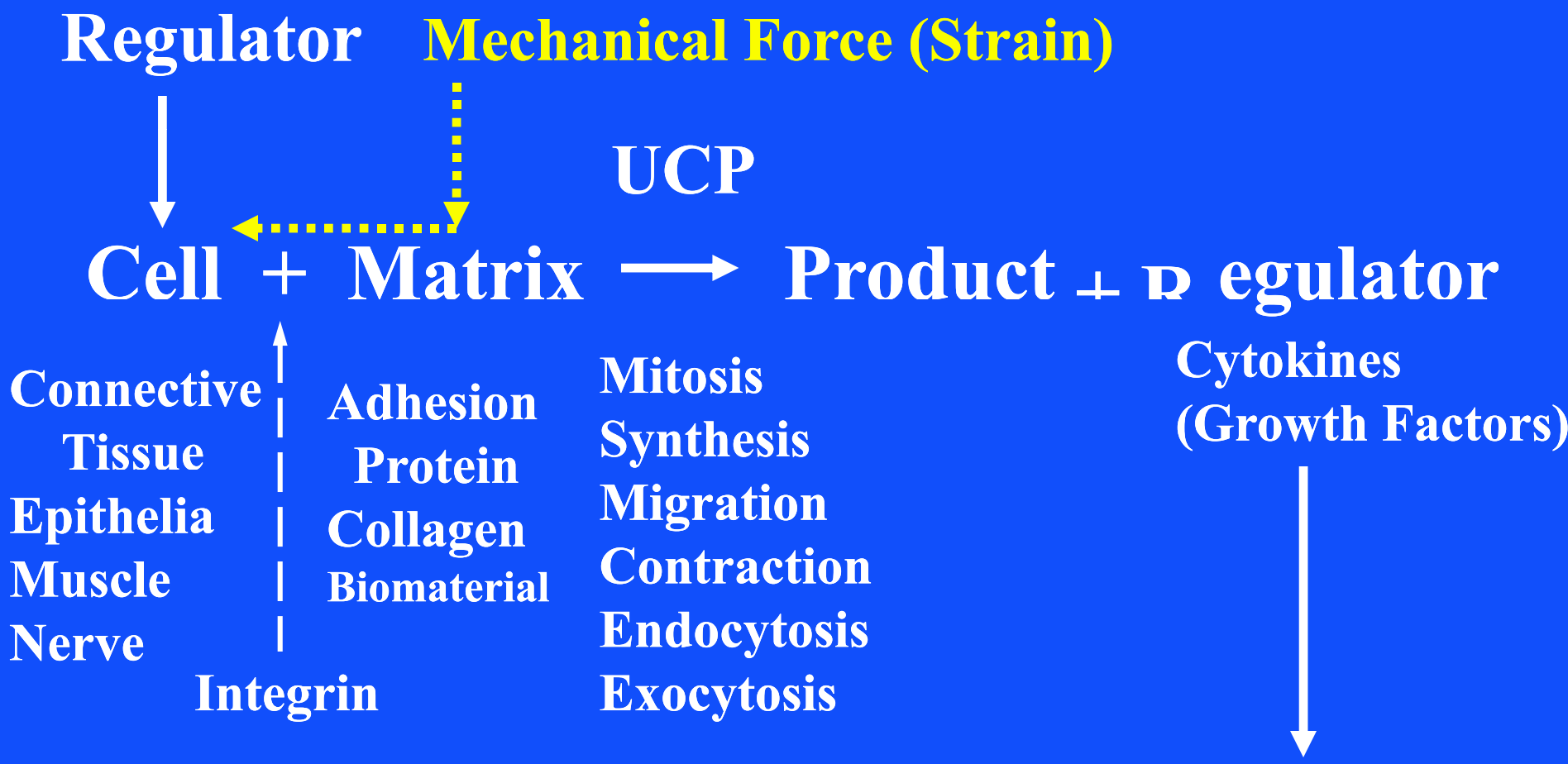
# REGULATORS

- Cytokines/Growth Factors
  - http://themedicalbiochemistrypage.org/growth-factors.html
  - (previously: http://web.indstate.edu/thcme/mwking/growth-factors.html
- http://www.copewithcytokines.de/

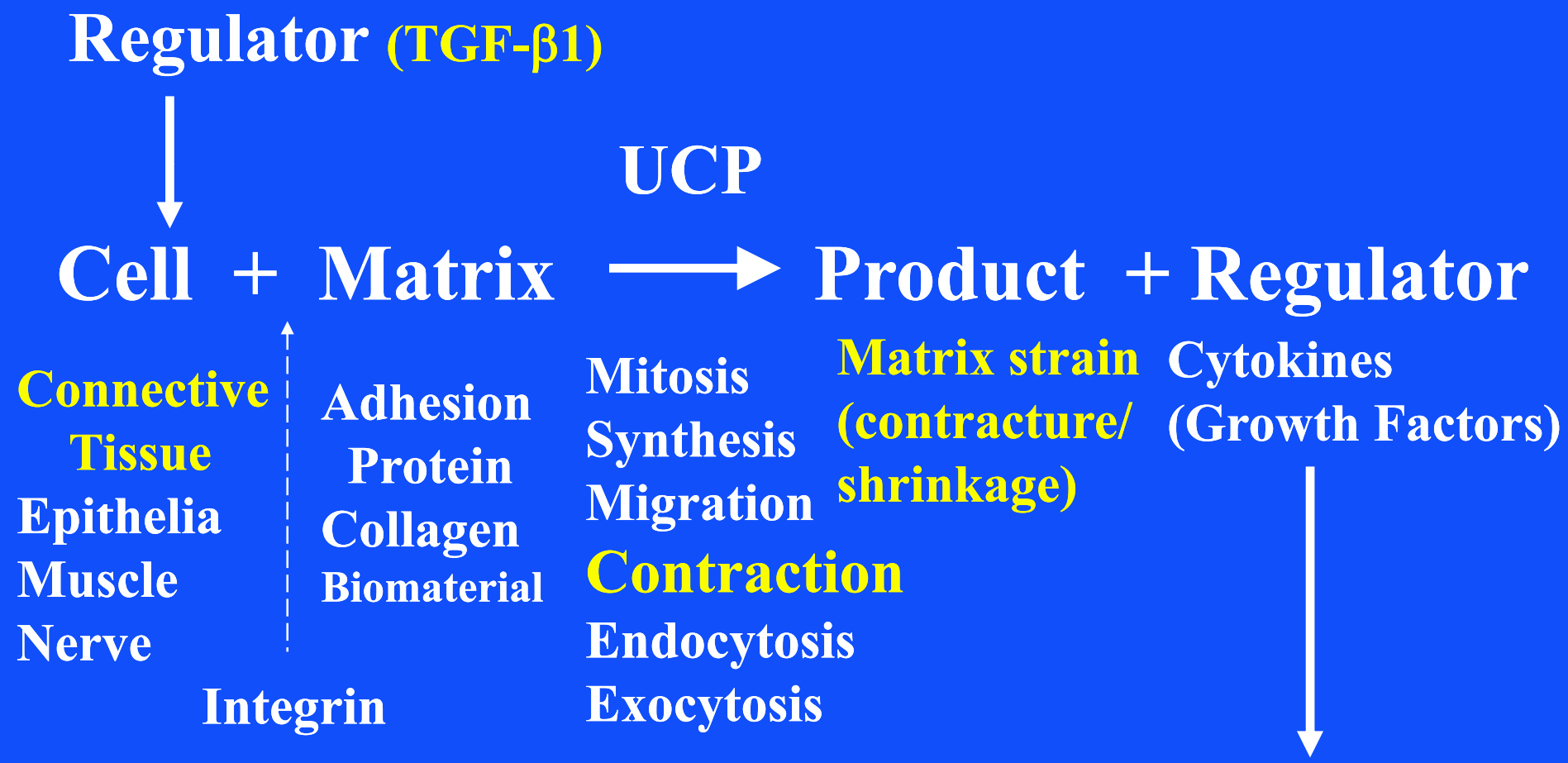
# "UNIT CELL PROCESSES"



# "UNIT CELL PROCESSES"



# "UNIT CELL PROCESSES"



# "UNIT CELL PROCESSES"

TGF-β1

↓

Contraction

Fibroblast + Collagen ⟶ Contracture + Reg.

# CONCEPTS FOR UNDERSTANDING BIOMATERIALS - TISSUE INTERACTIONS

- Control Volume
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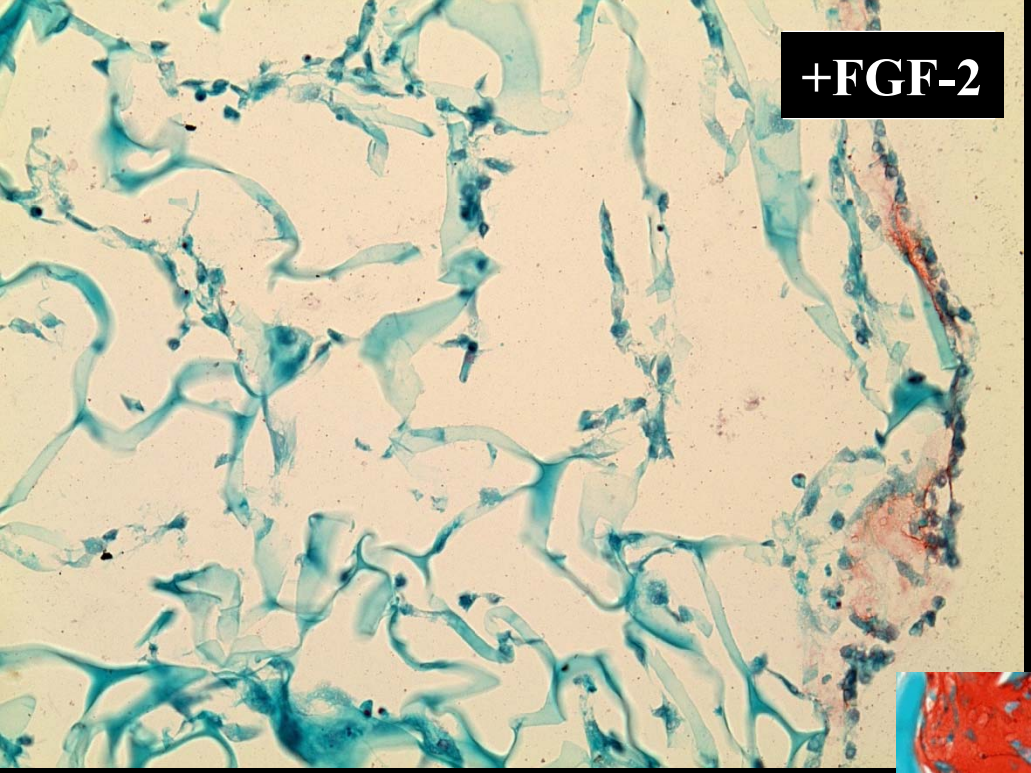
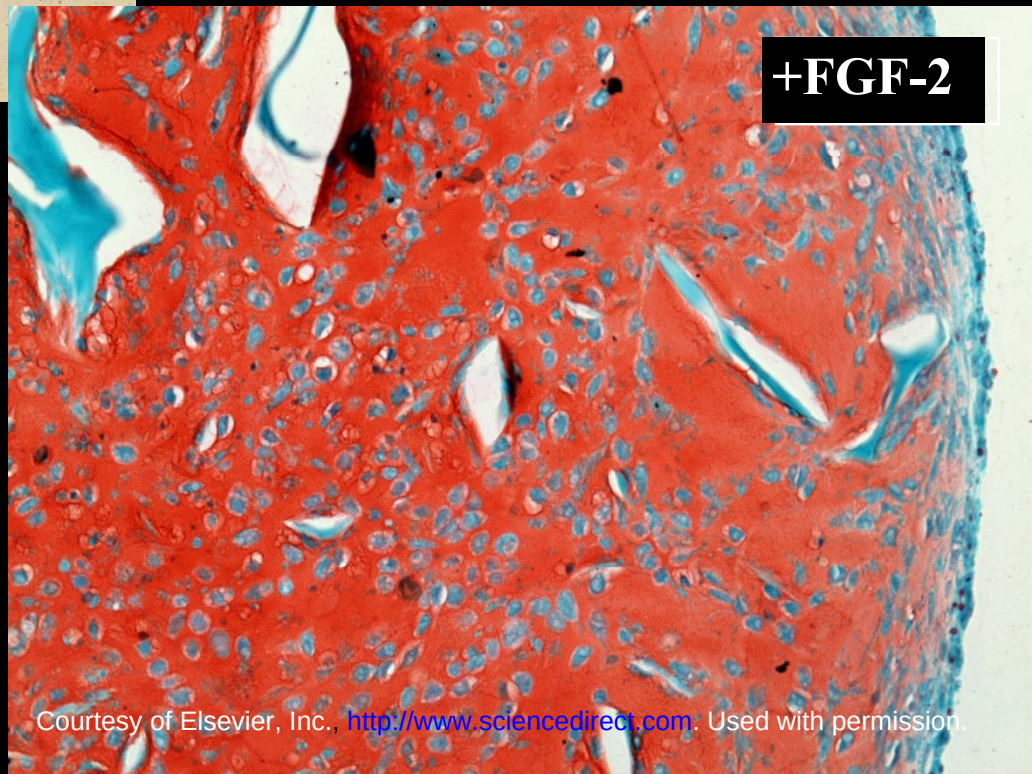
- Tissue Formation and Remodeling *In Vitro*
- Wound Healing *In Vivo*

# TISSUE FORMATION AND REMODELING *IN VITRO*




Canine chondrocytes grown in a type II collagen-GAG scaffold for 2 weeks. (Safranin O stain for GAGs)

N. Veilleux, *et al*., Osteoart. & Cart. 2005;13:278



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- Control Volume
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# WOUND HEALING
# Roots of Tissue Engineering

Injury

↓

Inflammation
(Vascularized tissue)

↓

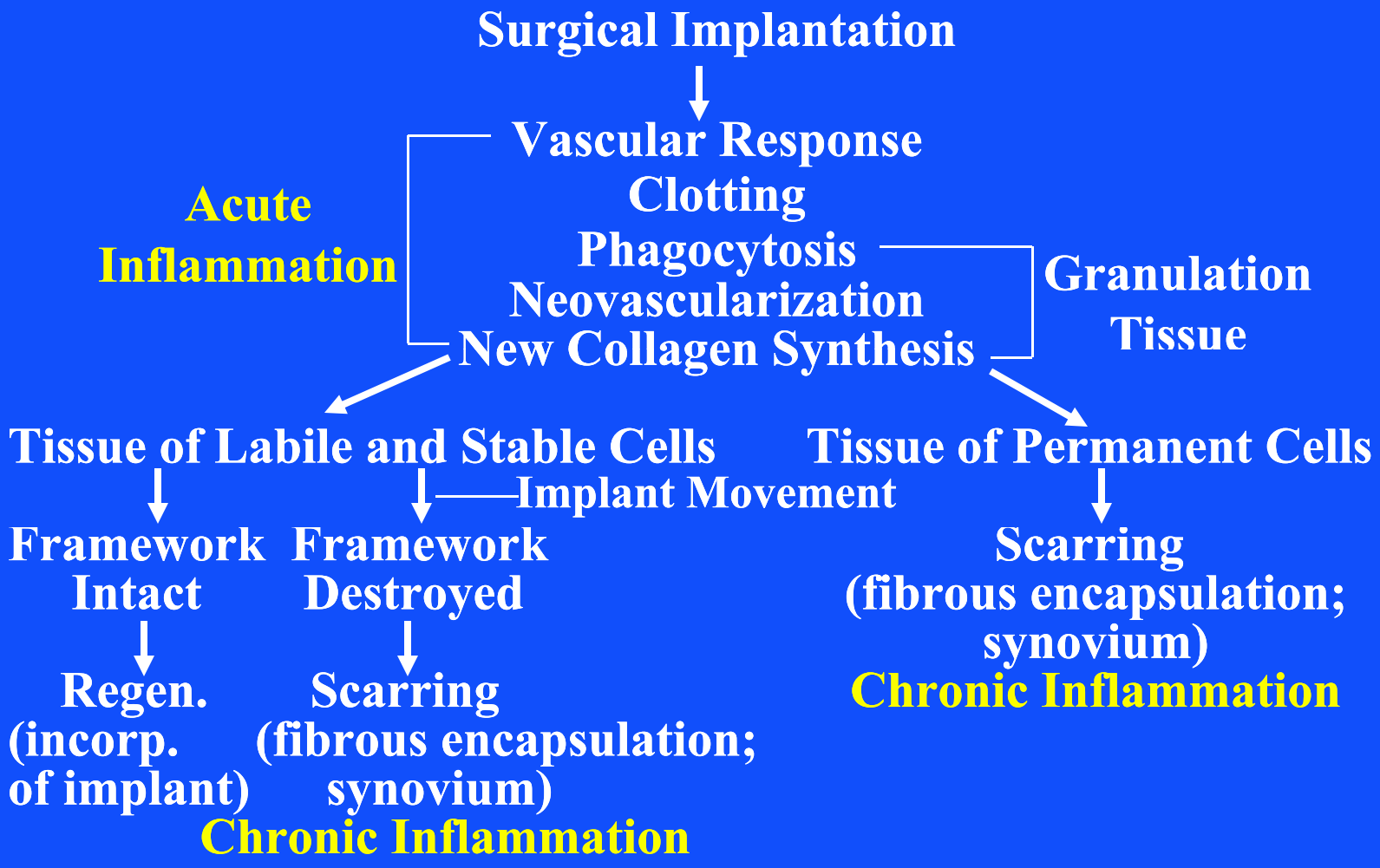
Reparative
Process

**4 Tissue Categories**
Connective Tissue
Epithelium
Nerve
Muscle

**Regeneration***
CT: bone
Ep: epidermis
Muscle: smooth
*spontaneous

**Repair (Scar)**
CT: cartilage
Nerve
Muscle: cardiac, skel.

# RESPONSE TO IMPLANTS: WOUND HEALING



20.441J / 2.79J / 3.96J / HST.522J Biomaterials-Tissue Interactions

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