Materials with Biological Recognition (continued)

TODAY: Using materials to mimic cell-cell contacts
start new section: inorganic biomaterials

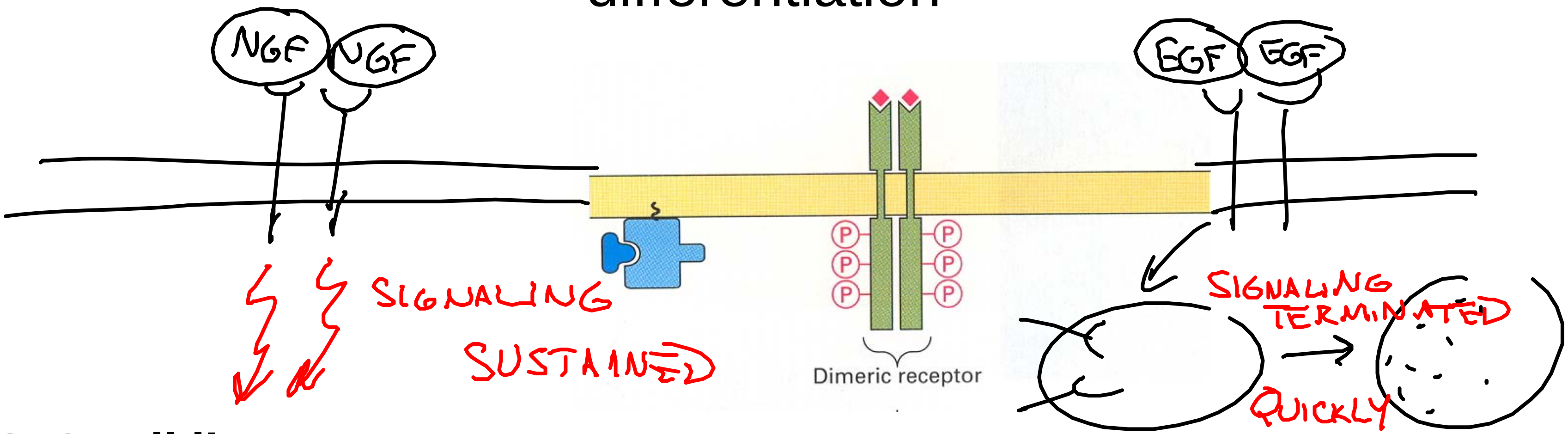
READING: CELL ADHESION / MECH. PROPS OF SUBSTRATES
ENZYMATIC RECOGNITION OF BIOMATERIALS
IMMOBILIZED PROTEINS

ANNOUNCEMENTS: NO CLASS NEXT TUES., 4/11
PS POSTED THIS AFTERNOON, DUE NEXT THURS.

Changes in signaling achieved by cytokine immobilization on surfaces

Image and figure text removed due to copyright reasons.
Please see: Figure 1 in Ito, Y. “Tissue Engineering by Immobilized Growth Factors.” *Materials Science and Engineering* C6 (1998): 267-274.

Surface immobilization can induce new function in cytokines: case of tethered EGF-triggered neuronal cell differentiation



PC12 cell line:

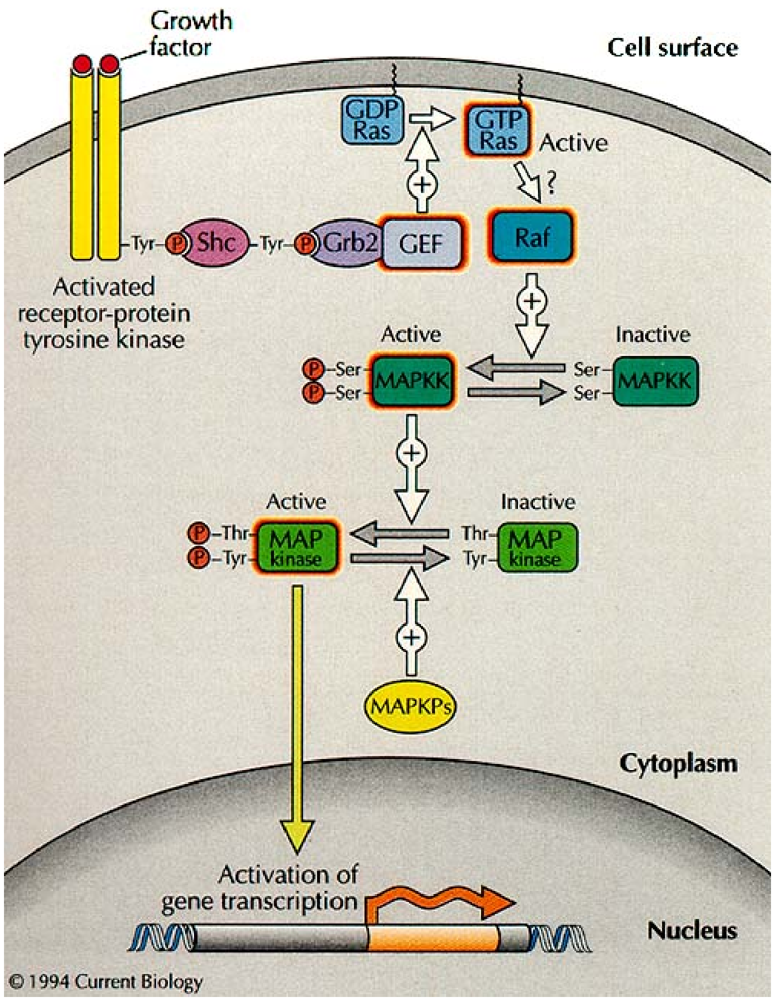
- induced to differentiate and extend axons under stimulation of **NGF** (nerve growth factor)

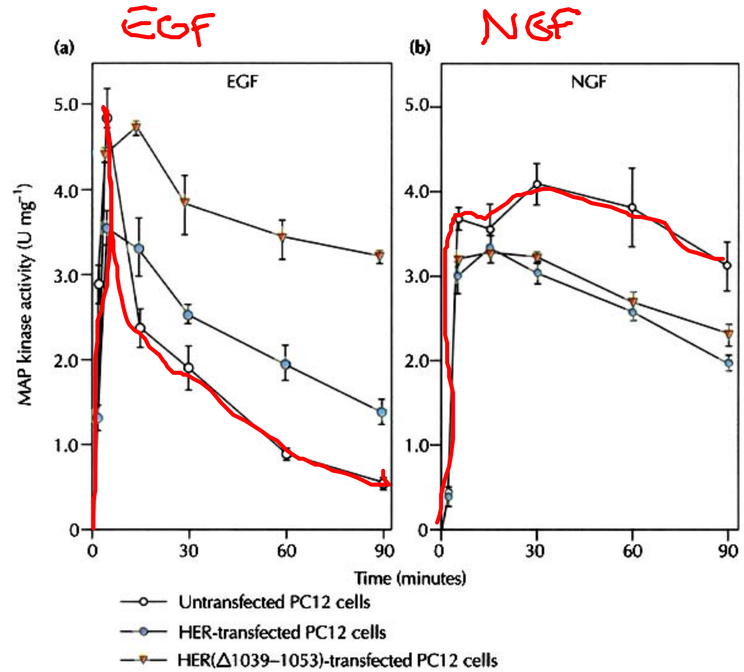
Signal doesn't trigger internalization of receptor; thus signal lasts longer and triggers differentiation

- induced to proliferate by **EGF**

Signal triggers internalization of receptor; short signal triggers proliferation

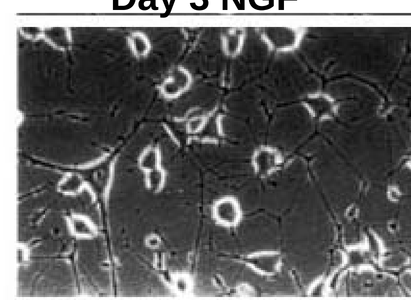
NGF vs. EGF signaling in PC12 neuronal cells





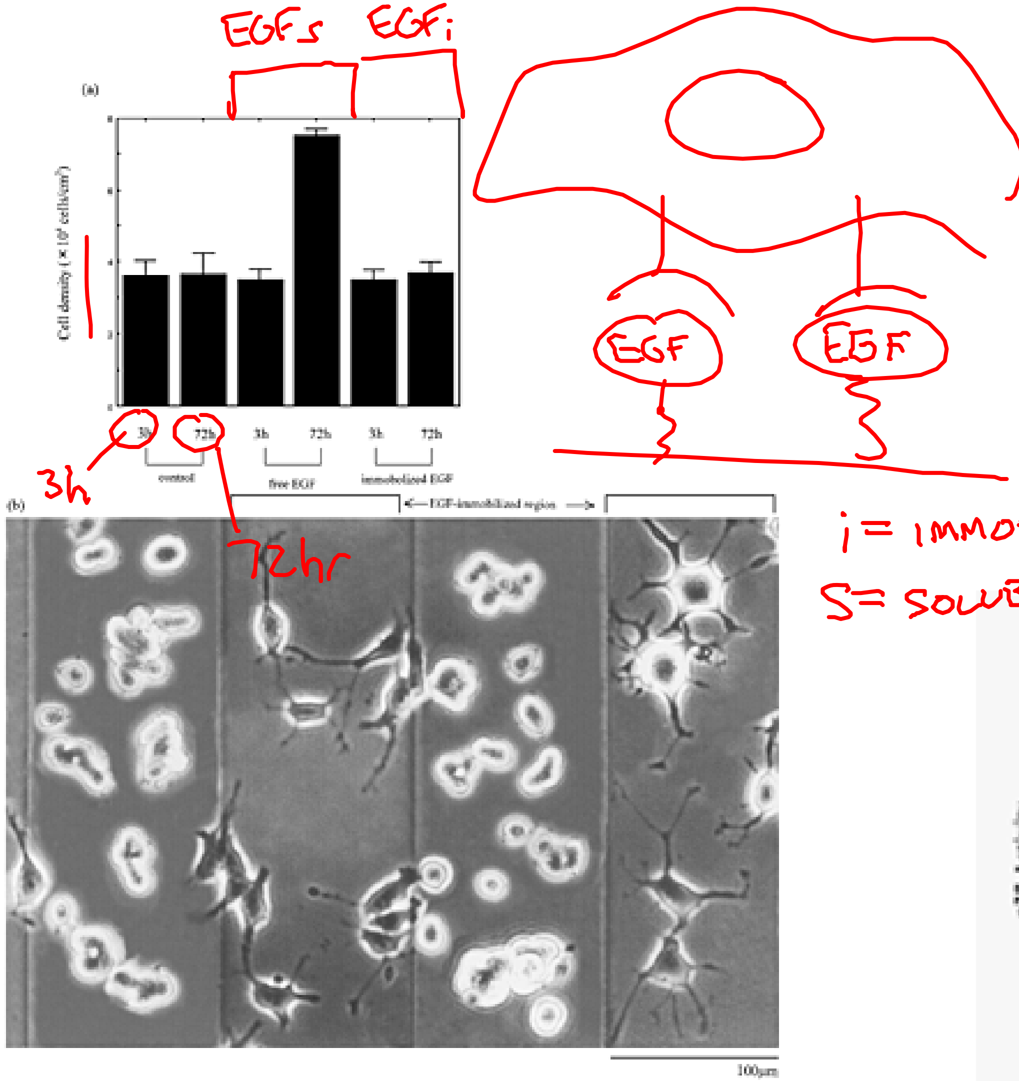
Day 3 EGF

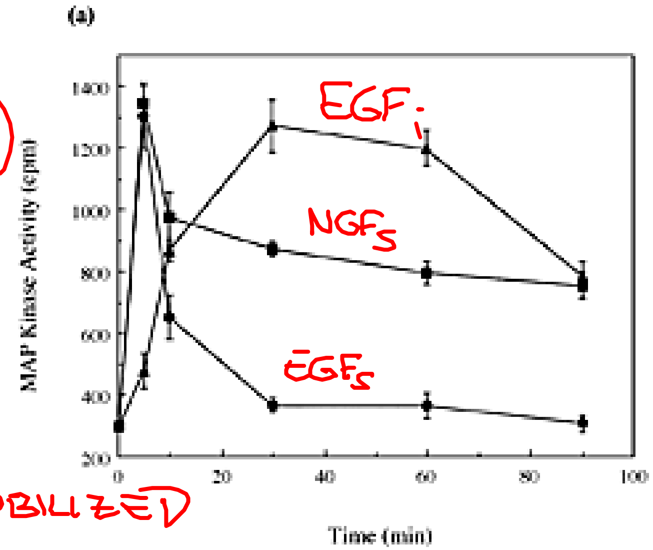
Day 3 NGF

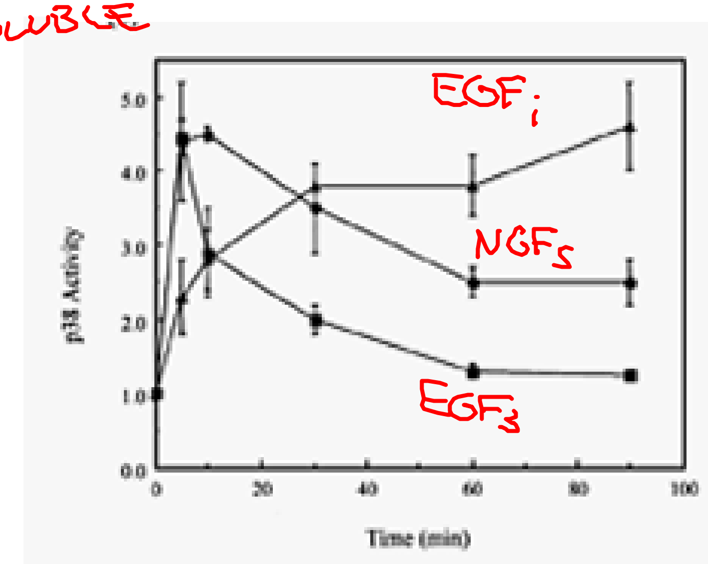


(Traverse et al. 1994)

Changing the biological activity of cytokines by surface immobilization:



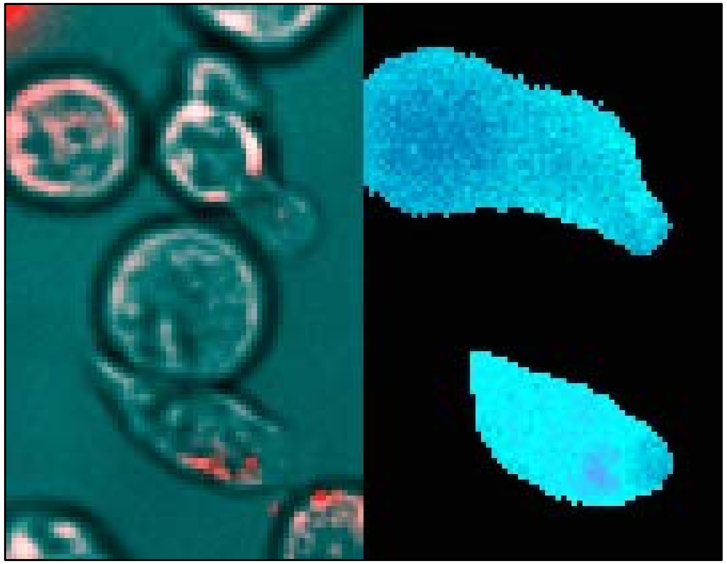


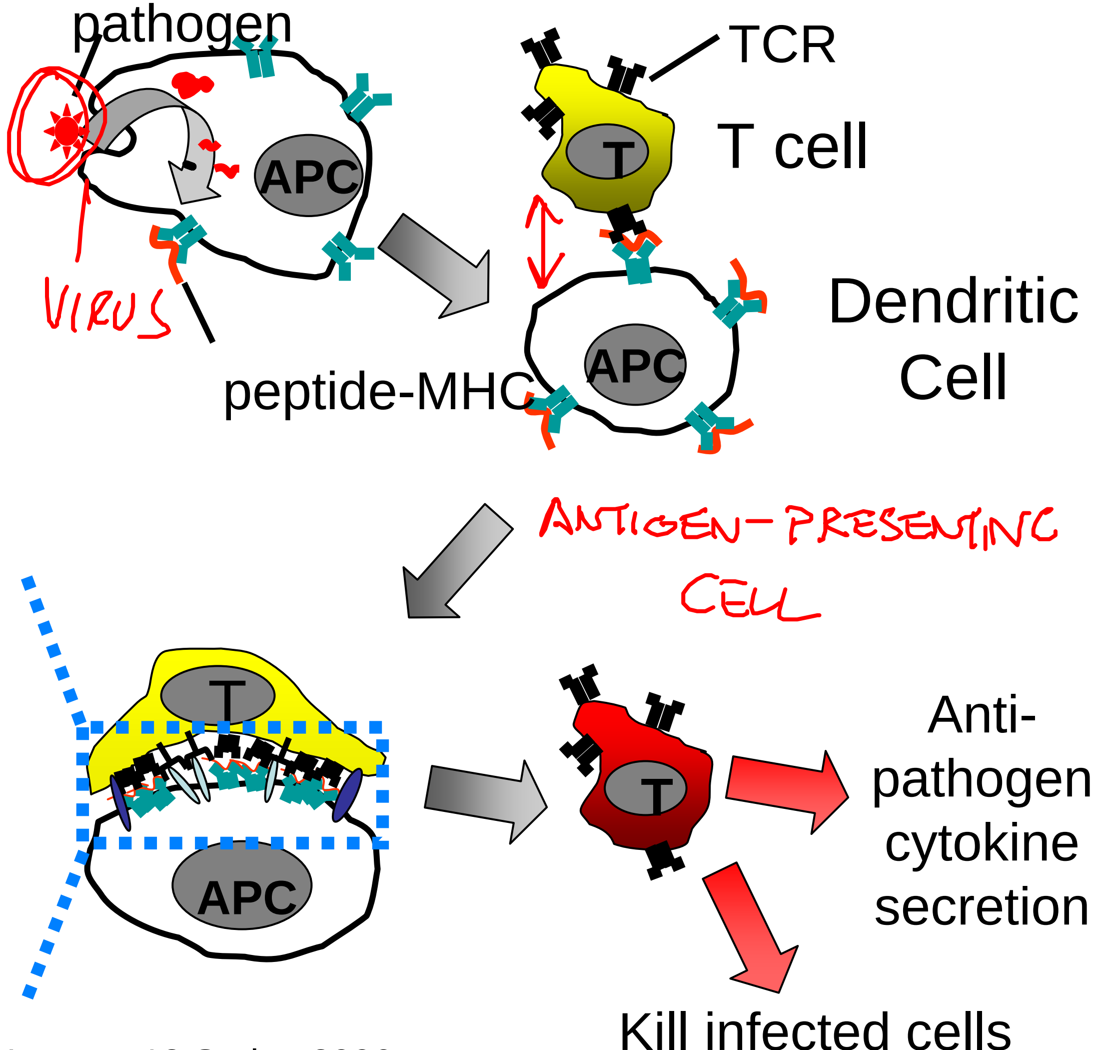


(Ito 2001)

Materials that mimic cell-cell contacts

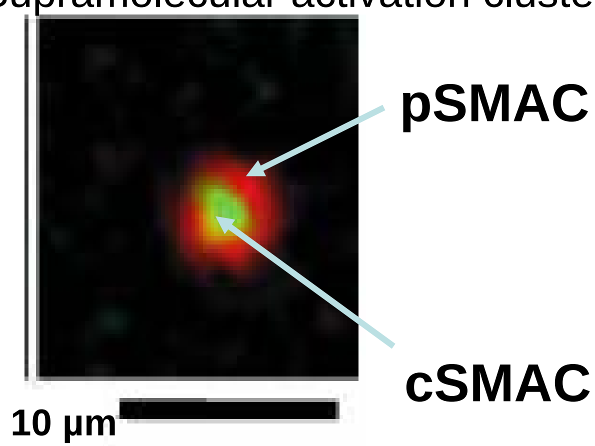
Physiology of the immune response: cellular level





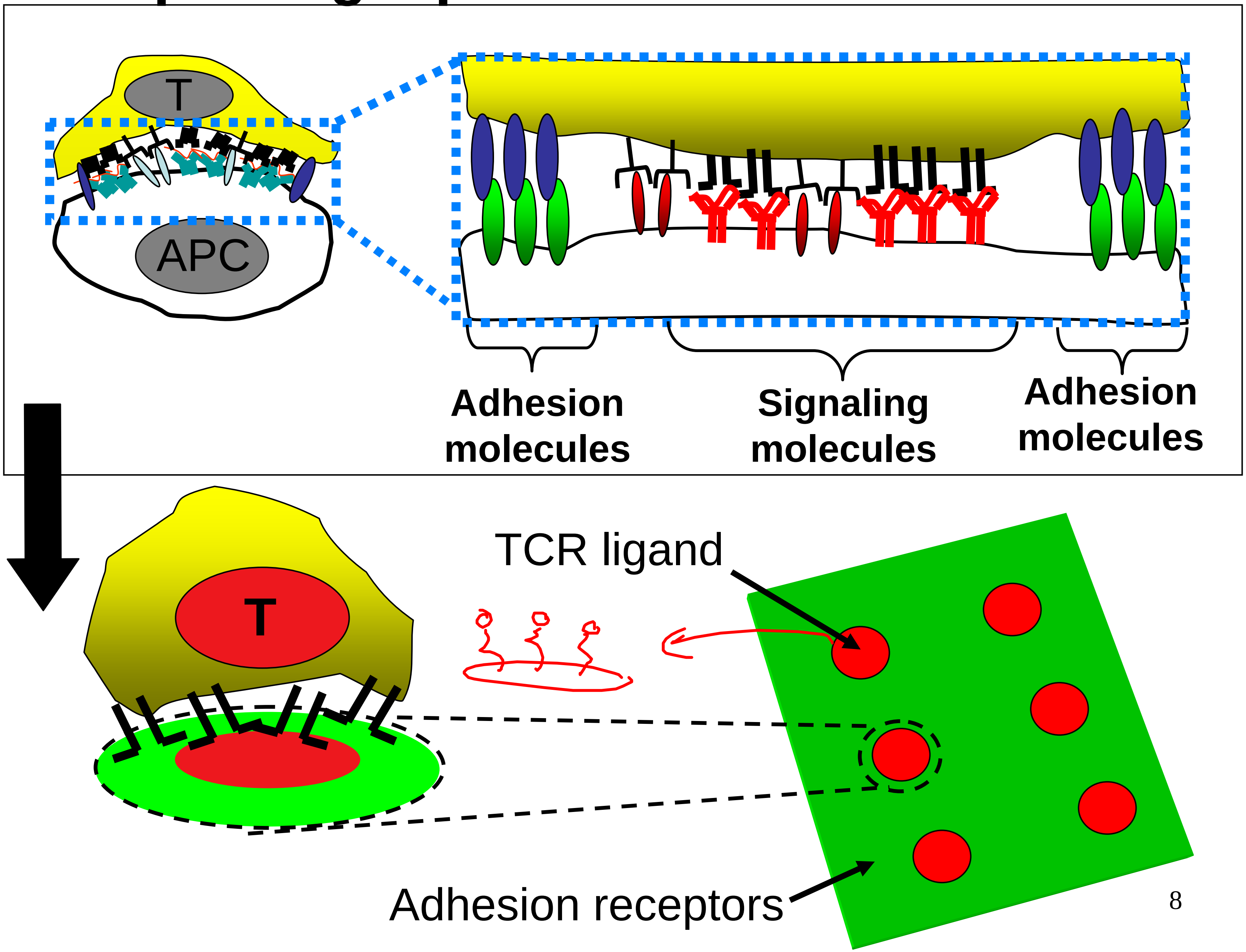
Immunological synapse (IS)

‘Supramolecular activation clusters’

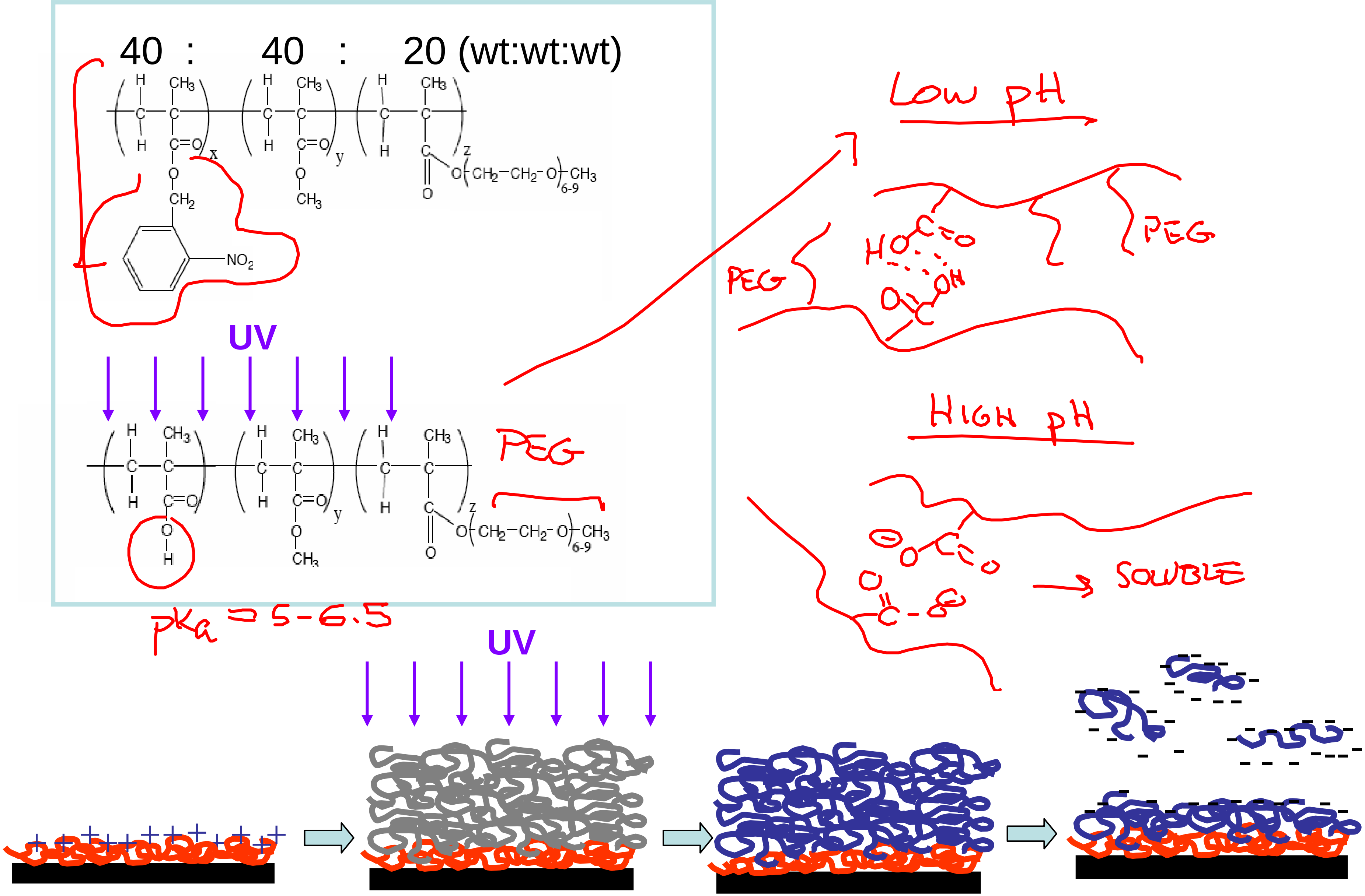


Grakoui et al. *Science* **285**, 221 (1999)

Replacing a partner cell with a surface:



PNMP photoresist



Cationic substrate

Spincoat photoresist

develop with PBS, pH 7.4

Methylene blue staining of patterned surface:

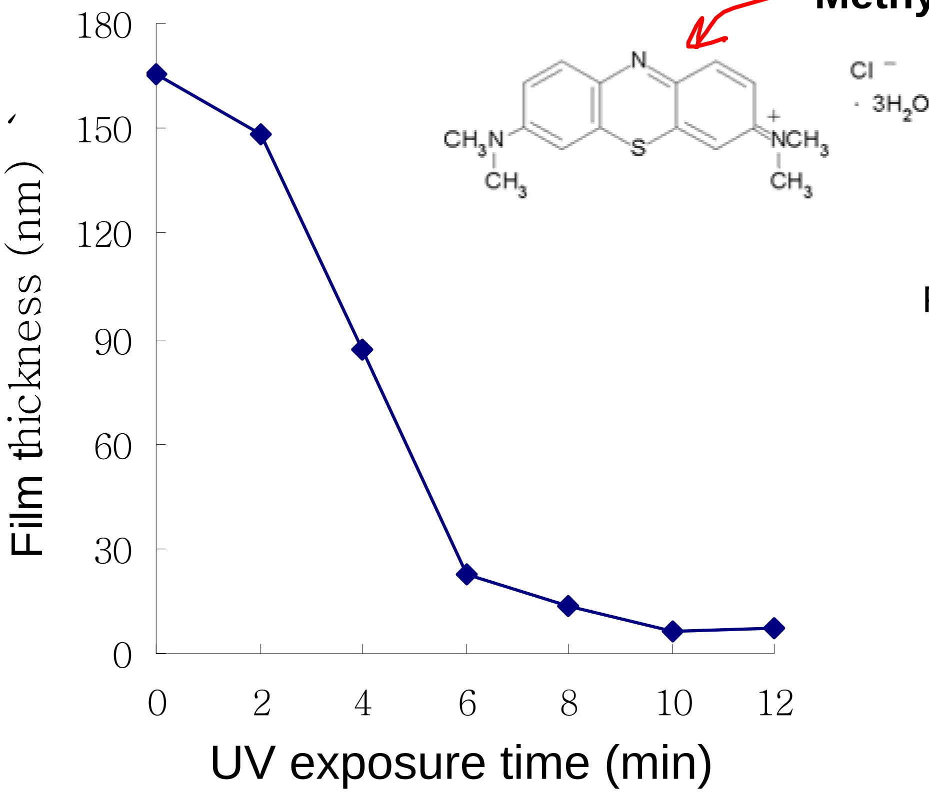
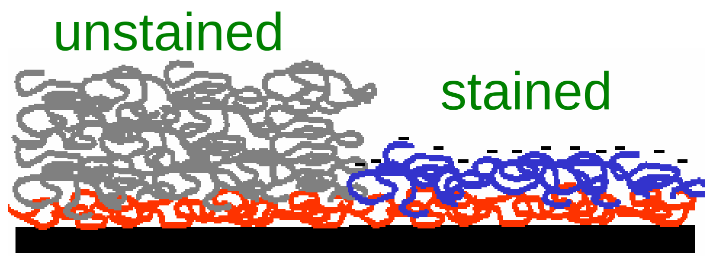
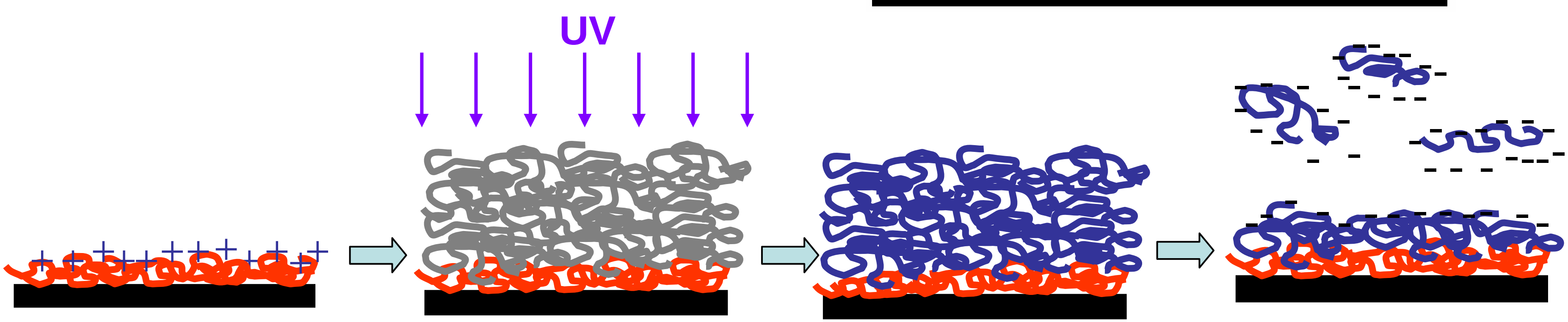


Image removed due to copyright reasons. Please see: Doh, J., and D. J. Irvine. *Journal of the Americal Chemical Society* 126, no. 30 (2004): 9170-9171.



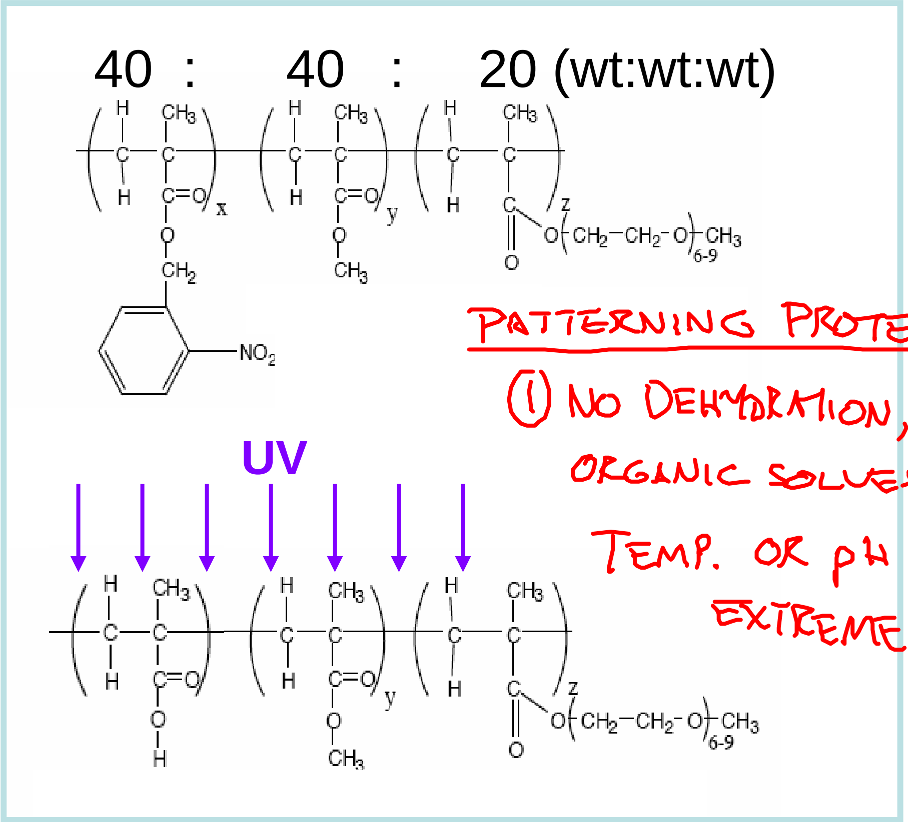


Cationic substrate

Spincoat photoresist

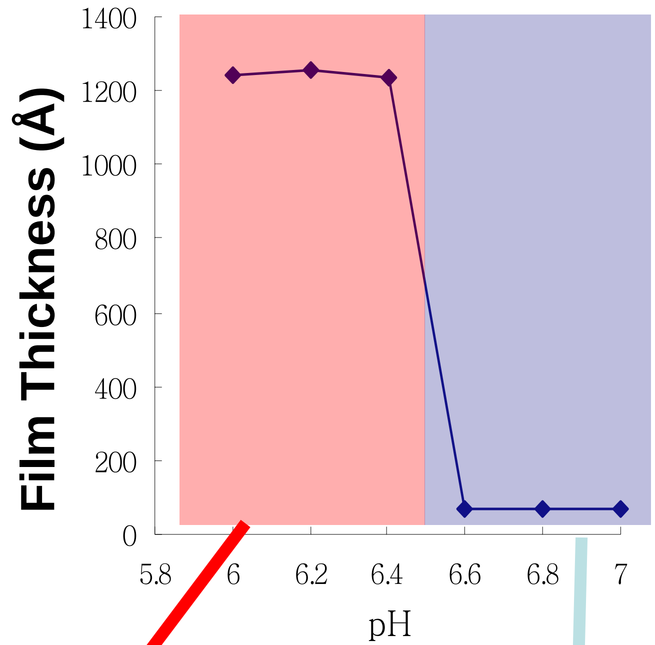
develop with PBS

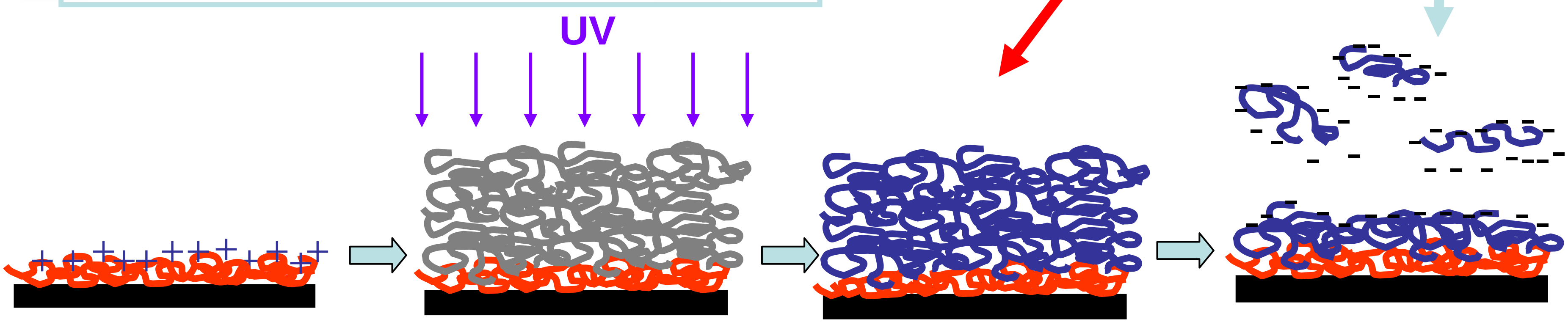
PNMP photoresist



PATTERNING PROTEINS:
(1) NO DEHYDRATION, ORGANIC SOLVENTS, TEMP. OR pH EXTREMES...

pH-dependent solubility of UV exposed photoresist:





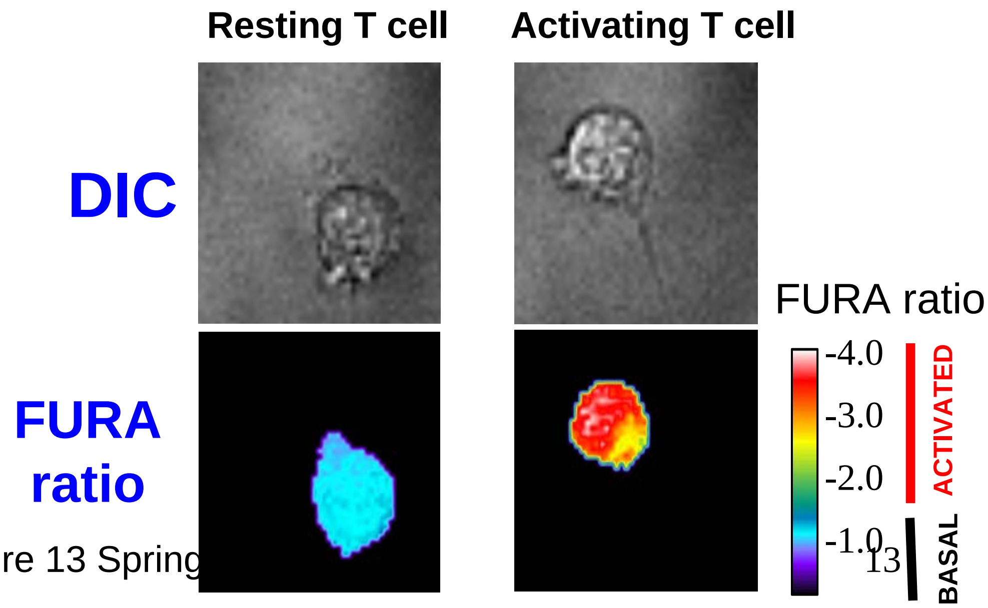
(J. Doh and D.J. Irvine. *JACS* (2004))

Images removed due to copyright reasons.
Please see: Doh, J., and D. J. Irvine. *PNAS* 103, no. 15 (2006): 5700-5705.

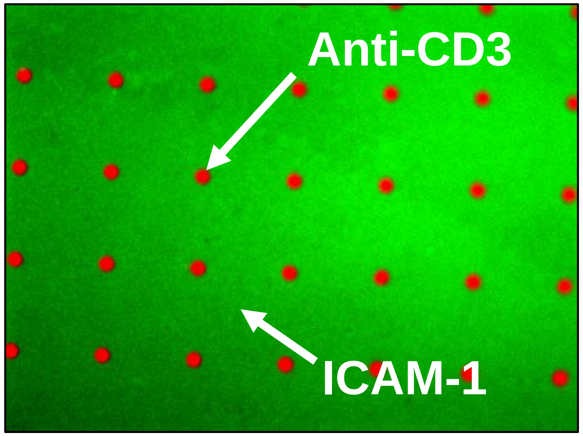
In situ tracking of T Cell Receptor triggering

Image removed due to copyright reasons.
Please see: Molecular probes web site - http://probes.invitrogen.com/

Image removed due to copyright reasons.
Please see: Abraham, and Weiss. *Nat Rev Immunol* 4 (2004): 301-308.



T cell migration on surfaces modulated by activation signals



Images removed due to copyright reasons.
Please see: Doh, J., and D. J. Irvine. *PNAS* 103, no. 15 (2006): 5700-5705.

T cells self-organize in response to synapse arrays

Graph and images removed due to copyright reasons.

Please see: Doh, J., and D. J. Irvine. *PNAS* 103, no. 15 (2006): 5700-5705.

Do surface-patterned ligands lead to full T cell activation?

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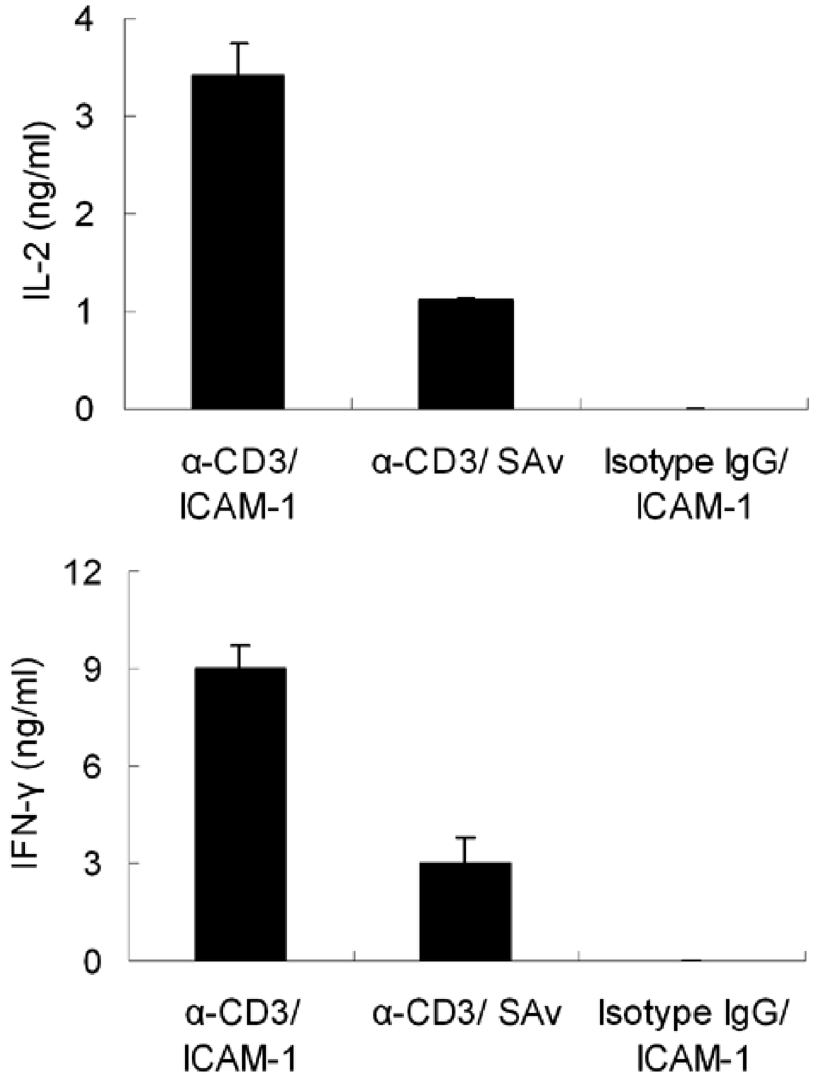
Please see: Doh, J., and D. J. Irvine. *PNAS* 103, no. 15 (2006): 5700-5705.

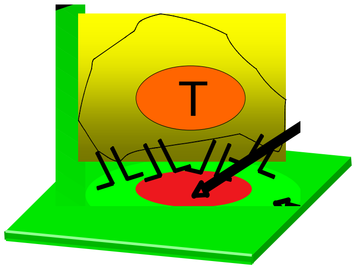


T cells assemble immunological synapses on ‘synapse array’ surfaces

Images removed due to copyright reasons.

Please see: Doh, J., and D. J. Irvine. *PNAS* 103, no. 15 (2006): 5700-5705.

Using protein micropatterned surfaces to direct immune cells:



Images removed due to copyright reasons.

Please see: Doh, J., and D. J. Irvine. *PNAS* 103, no. 15 (2006): 5700-5705.

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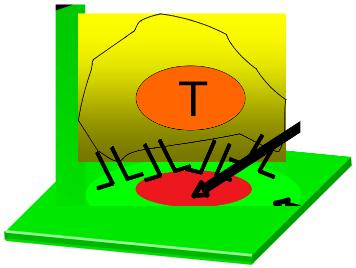


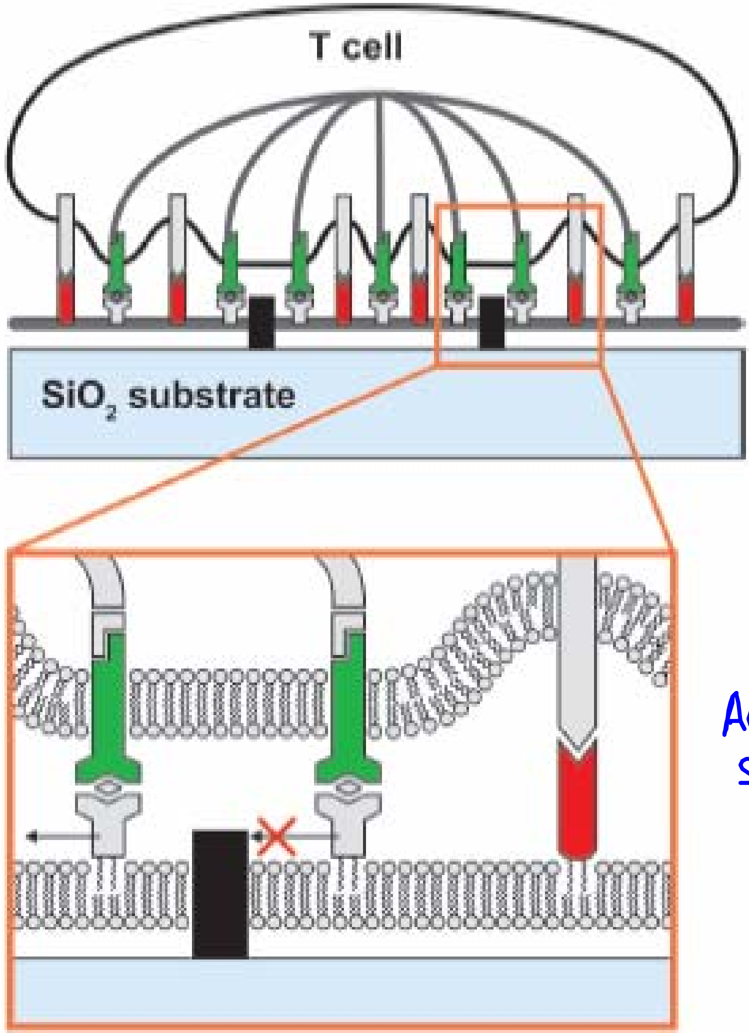
TCR ligands

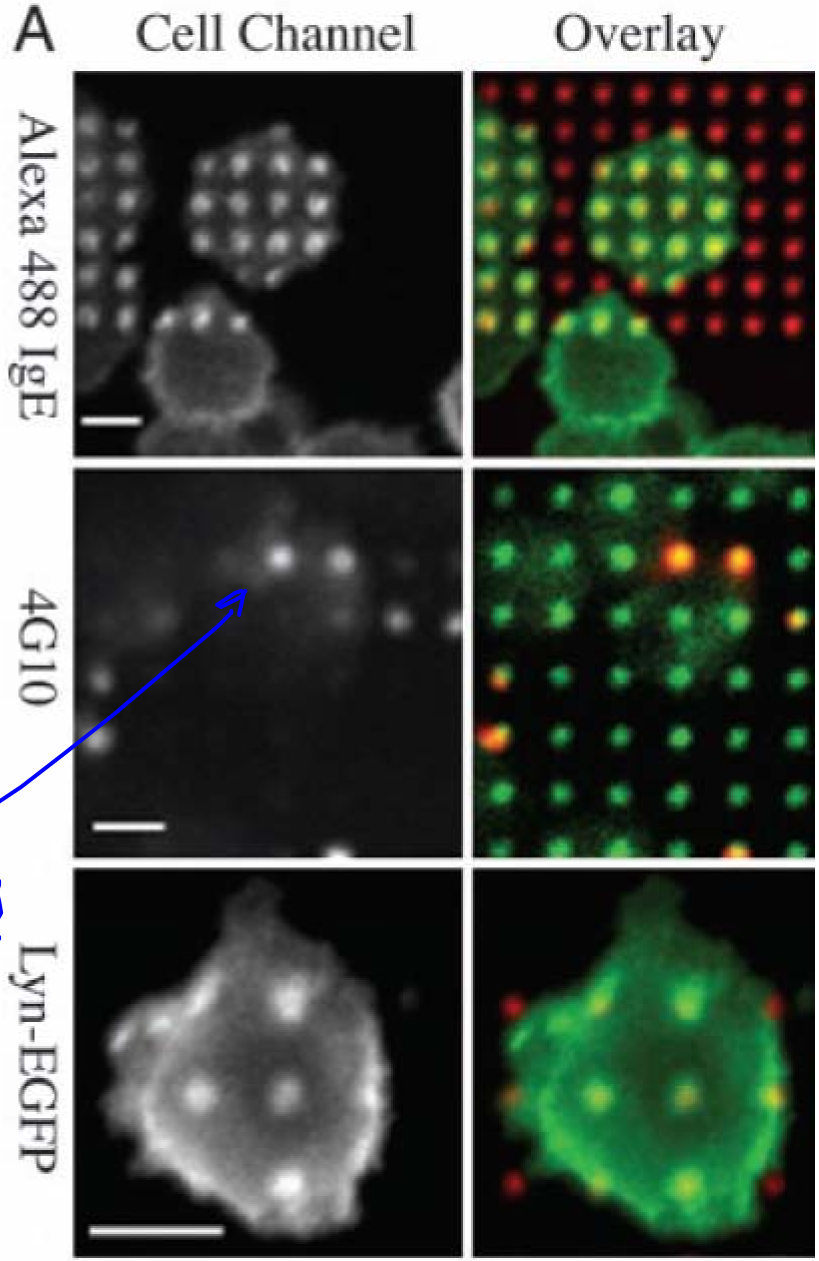
Adhesion ligands

Images removed due to copyright reasons.

Please see: Doh, J., and D. J. Irvine. PNAS 103, no. 15 (2006): 5700-5705.

Imparting mobility: patterned supported lipid bilayers





(Mossman et al *Science* **310** 1191-1193 (2005))
(Wu et al *PNAS* **101** 13798-13803 (2004))

Inorganic biomaterials

Last time: enzymatic recognition of biomaterials
Cytokine signaling from biomaterials

Today: introduction to biomineralization and biomimectic inorganic/organic composites
Interfacial biomineralization

Reading: Stephen Mann, 'Biomineralization: Principles and Concepts in Bioinorganic Materials Chemistry,' Ch. 3 pp. 24-37, Oxford Univ. Press (2001)
HANDOUTS

Supplementary Reading: -

ANNOUNCEMENTS:

Inorganic building blocks used by nature

Images removed due to copyright reasons.

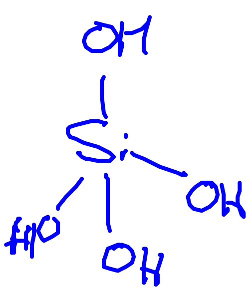
Please see: http://ruby.colorado.edu/~smyth/min/minerals.html

Inorganic building blocks used by nature

AMORPHOUS SILICA (SiO_2) → UNICELLULAR ORGANISMS: DIATOMS, RADIOLARIANS
PLANTS

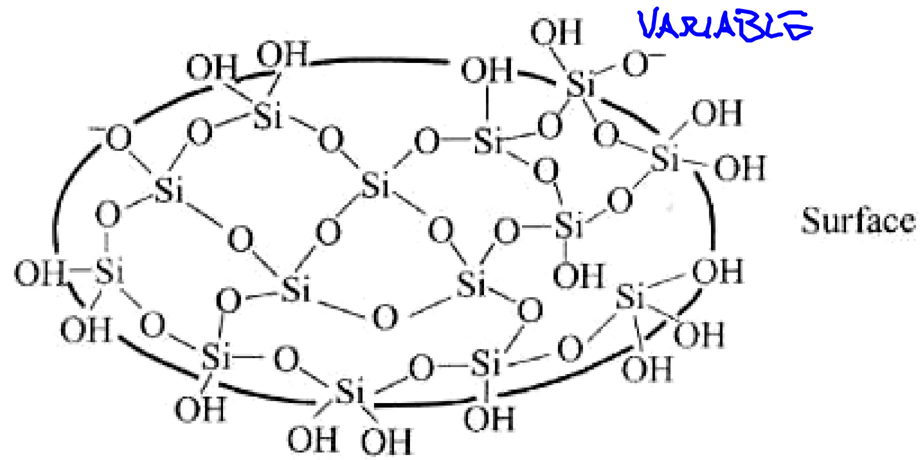
– LACKS FRACTURE/CLEAVAGE PLANES INHERENT IN CRYSTALS ALLOWS TOUGH, COMPLEX SHAPES TO BE SYNTHESIZED

DEGREE OF –OH CONVERSION IS VARIABLE



SILICIC ACID
($pK_a = 9.8$)

[SA] > 1 mM →



(Mann, 2001)

Inorganic building blocks used by nature

HYDROXYAPATITE:

Ca^{++}: OR Sr, Mg, Na, H_2O

PO_4^{3-} OR: HPO_4, CO_3 P_2O_7

OH OR: F, Cl, H_2O, O

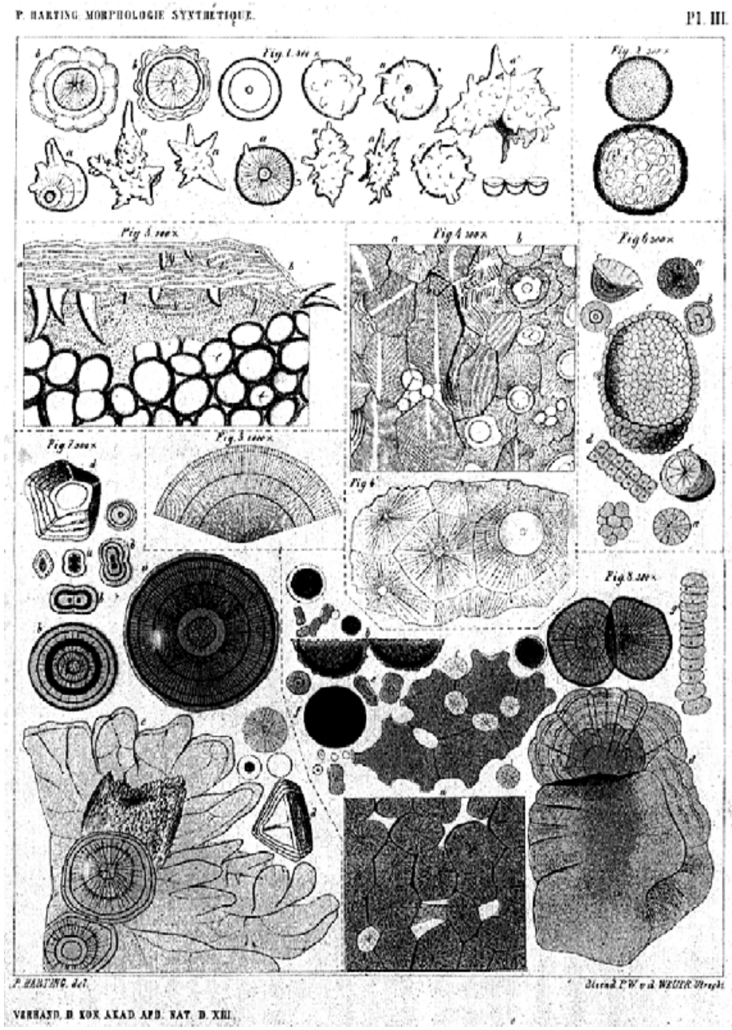
UNIVERSALLY FORMED AS A COMPOSITE W/ORGANIC MOLECULES (PROTEINS + POLYSACCHARIDES) AS A COMPONENT OF BONES AND TEETH

Table 2.2 Calcium phosphate biominerals

Mineral	Formula	Organism	Location	Function
Hydroxyapatite	$Ca_{10}(PO_4)_6(OH)_2$	Vertebrates	Bone	Endoskeleton
		Mammals	Teeth	Cutting/grinding
		Fish	Scales	Protection
Octacalcium phosphate	$Ca_8H_2(PO_4)_6$	Vertebrates	Bone/teeth	Precursor phase
Amorphous	variable	Chitons	Teeth	Precursor phase
		Gastropods	Gizzard plates	Crushing
		Bivalves	Gills	Ion store
		Mammals	Mitochondria	Ion store
		Mammals	Milk	Ion store

(Mann, 2001)

Bioceramics: motivation for studying and mimicking biomineralization



Pieter Harting's original hand drawings of calcareous microstructures (1872)

WHY SEEK TO MIMIC BIOMINERALIZATION PROCESSES?

BIOLOGY	LABORATORY METHODS
– PRECISE CONTROL OF MORPHOLOGY, STRUCTURES, (INCLUDING THOSE THAT DEFY CLASSICAL 230 SPACE GROUPS OF CRYSTALS) CRYSTAL ORIENTATION	– OBTAIN ONLY SIMPLE STRUCTURES
	– TYPICALLY REQUIRE HIGH TEMP. AND PRESSURES
– OCCUR AT NEAR-NEUTRAL pH, 37°C, AND 1atm	– RELY ON EXTREME pHs TO FORM CERTAIN STRUCTURES

Bioceramics: motivation for studying and mimicking biomineralization

APPLICATIONS:

BIOMATERIALS:

- REPLICATE TRABECULAR BONE STRUCTURE AND IT'S MECH. PROPS → THIS IS STILL ELUSIVE
- LOW-COST, REPRODUCIBLE, HIGH-VOLUME BONE GRAFT MATERIALS

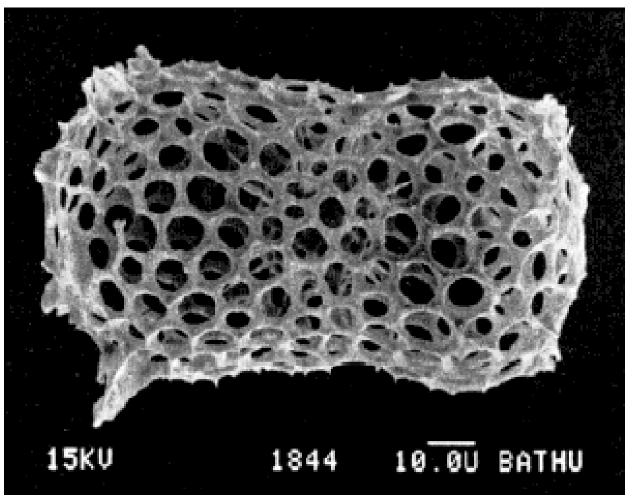
STRUCTURAL MATERIALS:

INORGANIC-ORGANIC COMPOSITES HAVE UP TO 3000X GREATER STRENGTHS THAN PURE INORGANIC CRYSTALS

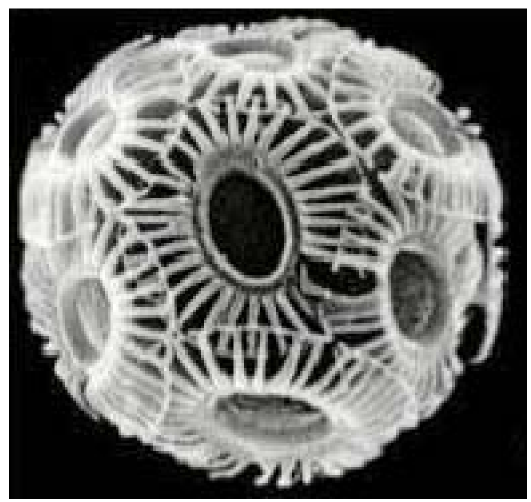
Complex macro- and microstructures of biological inorganic materials

CENTRAL TENETS OF BIOMINERALIZATION!

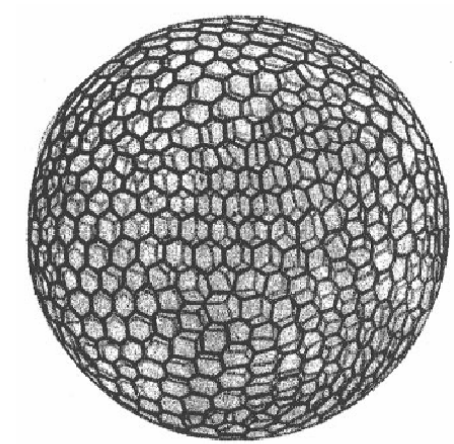
- ORGANIC MOLECULES REGULATE NUCLEATION, GROWTH, MORPHOLOGY, AND ASSEMBLY OF INORGANIC MATLS
- OFTEN EMPLOY MOLECULAR RECOGNITION AT ORGANIC-INORGANIC INTERFACES TO CONTROL SYNTHESES



Radiolarian: Microskeleton of amorphous silica



Coccolith: $CaCO_3$ microskeleton



A. hexagona: Microskeleton of amorphous silica

Complex macro- and microstructures of biological inorganic materials

Organized at macro, micro, meso, nano length scales

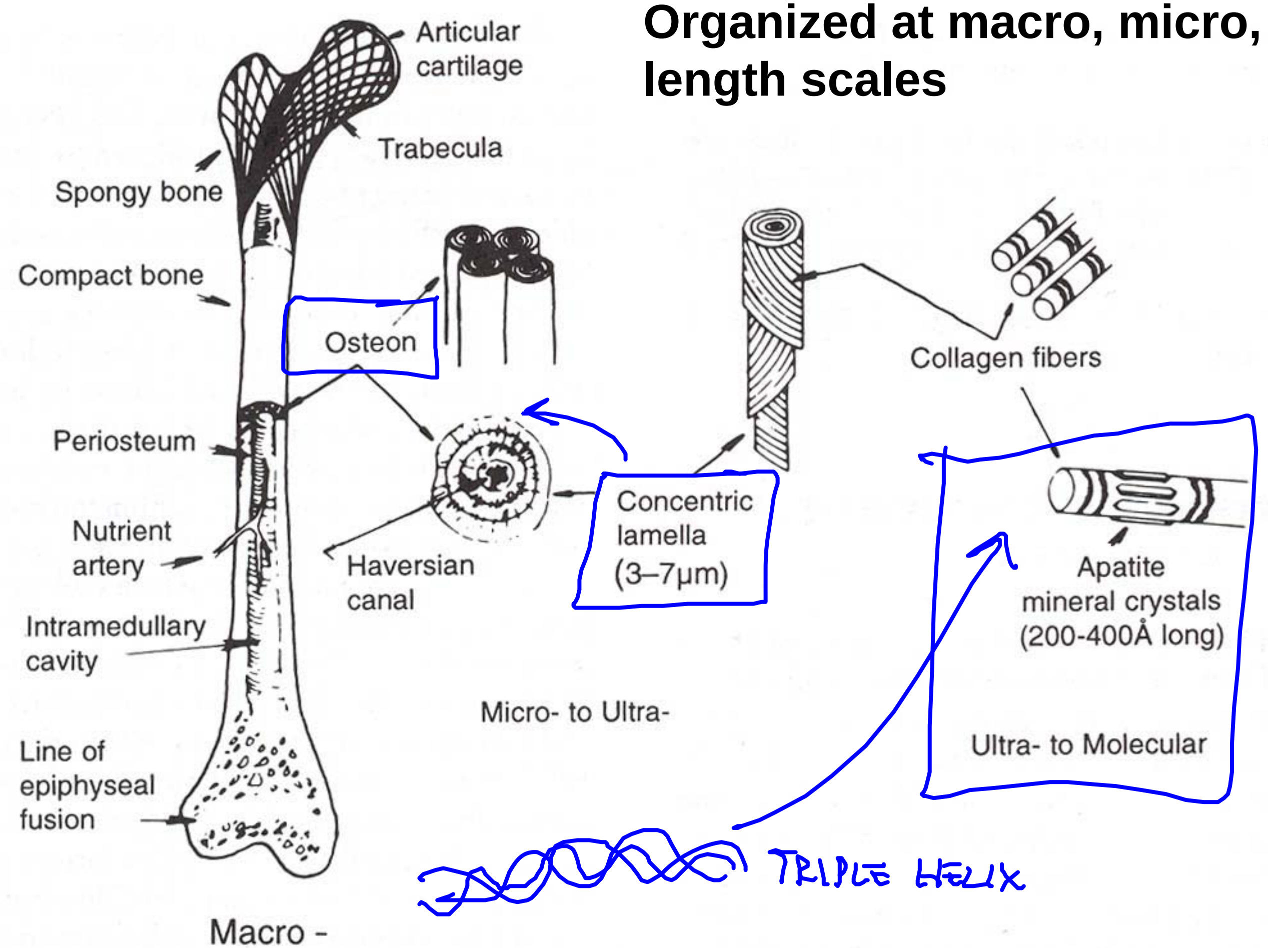


FIG. 1. Hierarchical levels of structural organization in a human long femur. (Adapted with permission from J. B. Park, *Biomaterials: An Introduction*, Plenum Publ., 1979, p. 105.)

Paradigms in biomineralization

Further Reading

1. Voet & Voet. in *Biochemistry*.
2. Paredes, N., Rodriguez, G. A. & Puiggali, J. Synthesis and characterization of a family of biodegradable poly(ester amide)s derived from glycine. *Journal of Polymer Science, Part A: Polymer Chemistry* **36**, 1271-1282 (1998).
3. Fan, Y., Kobayashi, M. & Kise, H. Synthesis and biodegradability of new polyesteramides containing peptide linkages. *Polymer Journal* **32**, 817-822 (2000).
4. O, S. C. & Birkinshaw, C. Hydrolysis of poly (n-butylcyanoacrylate) nanoparticles using esterase. *Polymer Degradation and Stability* **78**, 7-15 (2002).
5. Ekblom, P. & Timpl, R. Cell-to-cell contact and extracellular matrix. A multifaceted approach emerging. *Curr Opin Cell Biol* **8**, 599-601 (1996).
6. Chapman, H. A. Plasminogen activators, integrins, and the coordinated regulation of cell adhesion and migration. *Curr Opin Cell Biol* **9**, 714-24 (1997).
7. Mann, B. K., Gobin, A. S., Tsai, A. T., Schmedlen, R. H. & West, J. L. Smooth muscle cell growth in photopolymerized hydrogels with cell adhesive and proteolytically degradable domains: synthetic ECM analogs for tissue engineering. *Biomaterials* **22**, 3045-51 (2001).
8. West, J. L. & Hubbell, J. A. Polymeric biomaterials with degradation sites for proteases involved in cell migration. *Macromolecules* **32**, 241-244 (1999).
9. Gobin, A. S. & West, J. L. Cell migration through defined, synthetic ECM analogs. *Faseb J* **16**, 751-3 (2002).
10. Sperinde, J. J. & Griffith, L. G. Control and prediction of gelation kinetics in enzymatically cross-linked poly(ethylene glycol) hydrogels. *Macromolecules* **33**, 5476-5480 (2000).
11. Sperinde, J. J. & Griffith, L. G. Synthesis and characterization of enzymatically-cross-linked poly(ethylene glycol) hydrogels. *Macromolecules* **30**, 5255-5264 (1997).
12. Zhang, Z. Y., Shum, P., Yates, M., Messersmith, P. B. & Thompson, D. H. Formation of fibrinogen-based hydrogels using phototriggerable diplasmalogen liposomes. *Bioconjug Chem* **13**, 640-6 (2002).
13. Sanborn, T. J., Messersmith, P. B. & Barron, A. E. In situ crosslinking of a biomimetic peptide-PEG hydrogel via thermally triggered activation of factor XIII. *Biomaterials* **23**, 2703-10 (2002).
14. Collier, J. H. et al. Thermally and photochemically triggered self-assembly of peptide hydrogels. *J Am Chem Soc* **123**, 9463-4 (2001).
15. Collier, J. H. & Messersmith, P. B. Enzymatic modification of self-assembled peptide structures with tissue transglutaminase. *Bioconjug Chem* **14**, 748-55 (2003).
16. Schense, J. C., Bloch, J., Aebischer, P. & Hubbell, J. A. Enzymatic incorporation of bioactive peptides into fibrin matrices enhances neurite extension. *Nat Biotechnol* **18**, 415-9 (2000).
17. Ito, Y. Tissue engineering by immobilized growth factors. *Materials Science and Engineering C* **6**, 267-274 (1998).
18. Ito, Y. Regulation of cell functions by micropattern-immobilized biosignal molecules. *Nanotechnology* **9**, 200-204 (1998).
19. Kuhl, P. R. & Griffith-Cima, L. G. Tethered epidermal growth factor as a paradigm for growth factor-induced stimulation from the solid phase. *Nat Med* **2**, 1022-7 (1996).
20. Chen, G. & Ito, Y. Gradient micropattern immobilization of EGF to investigate the effect of artificial juxtacrine stimulation. *Biomaterials* **22**, 2453-7 (2001).
21. Ito, Y. Surface micropatterning to regulate cell functions. *Biomaterials* **20**, 2333-42 (1999).