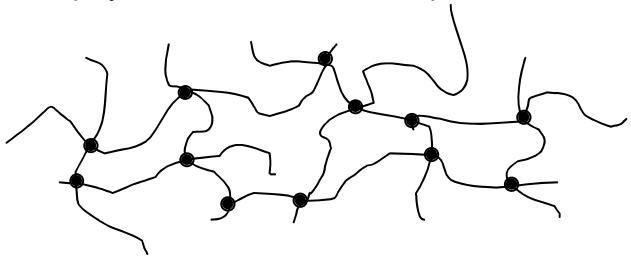
Lecture 7: Hydrogel Biomaterials: Structure and Physical Chemistry

Last Day:	programmed/regulated/multifactor controlled release for drug delivery and tissue engineering
Today:	Applications of hydrogels in bioengineering Covalent hydrogels structure and chemistry of biomedical gels Thermodynamics of hydrogel swelling
Reading:	N.A. Peppas et al., 'Physicochemical foundations and structural design of hydrogels in medicine and biology,' *Annu. Rev. Biomed. Eng*., **2**, 9-29 (2000).
Supplementary Reading:	P.J. Flory, 'Principles of Polymer Chemistry,' Cornell University Press, Ithaca, pp. 464-469, pp. 576-581 (Statistical thermodynamics of networks and network swelling)

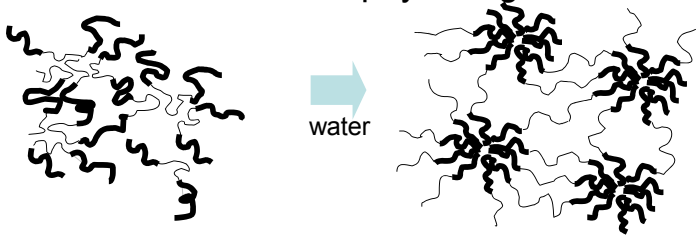
Applications of hydrogels in bioengineering

- Hydrogels: insoluble network of polymer chains that swell in aqueous solutions



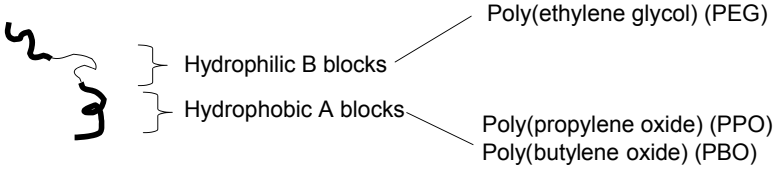
- Gels can be classified by the type of crosslinker:[1]
  - Covalent - covalent junctions
  - Physical - non-covalent junctions

Physical gels: example- Hydrophobic interactions in physical gels



Physical gels are formed by *noncovalent* cross-links

Example blocks:



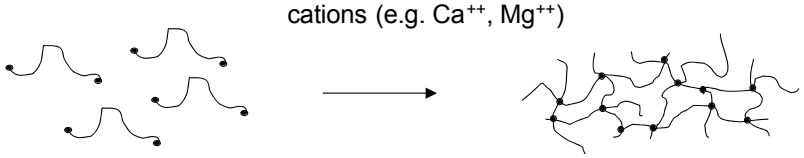
- Key properties of gels for bioengineering applications:
    1. *in situ* formability
    2. degradability
    3. responsive swelling
    4. tissue-like structure/properties

- *In situ* formability
    - *Gelation of liquid solutions by:*
        - Irradiation with light
        - Temperature change (e.g. 4°C to 37°C)
        - Cross-linking enzymes
        - Presence of divalent salts

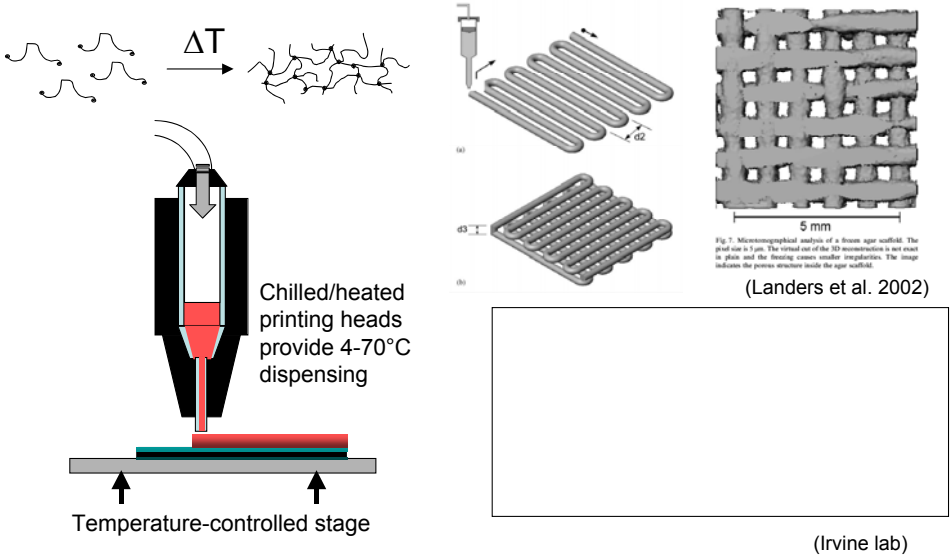
ON BOARD:

In situ formation

¥ħν
¥Heat
¥Crosslinking by enzymes
¥Introduction of divalent cations (e.g. Ca^{++}, Mg^{++})



Key properties of hydrogels for bioengineering applications: example: ÔprintableÕgels



(Landers et al. 2002)

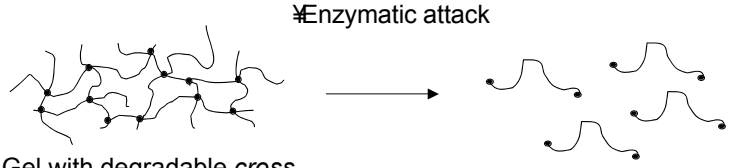
(Irvine lab)

- Degradability

ON BOARD:

Degradability

¥Hydrolysis
¥Enzymatic attack



Gel with degradable *cross-links* or *network chains*

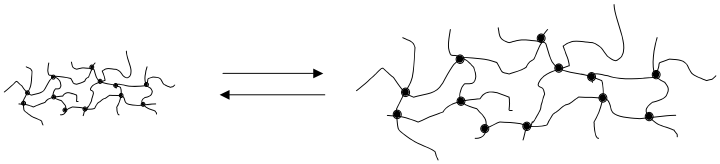
Eliminible/metabolizable Water-soluble fragments

- Responsive swelling
  - Temperature-, pH-, and molecule-responsive swelling
  - Basis of sensors and ‘smart’ materials
  - (to be covered later)

ON BOARD:

Responsive swelling

¥ΔpH
¥ΔT
¥Δc (change in concentration of a molecule



- Tissue-like structure/properties
  - Form swollen networks similar to collagen, elastin, proteoglycans

- General areas of application in bioengineering:
  - Controlled release

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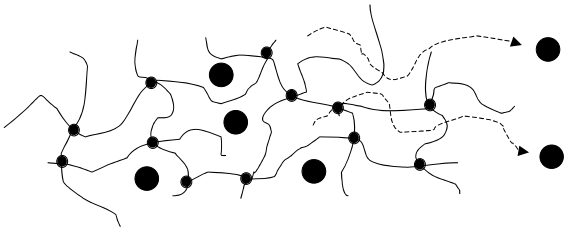
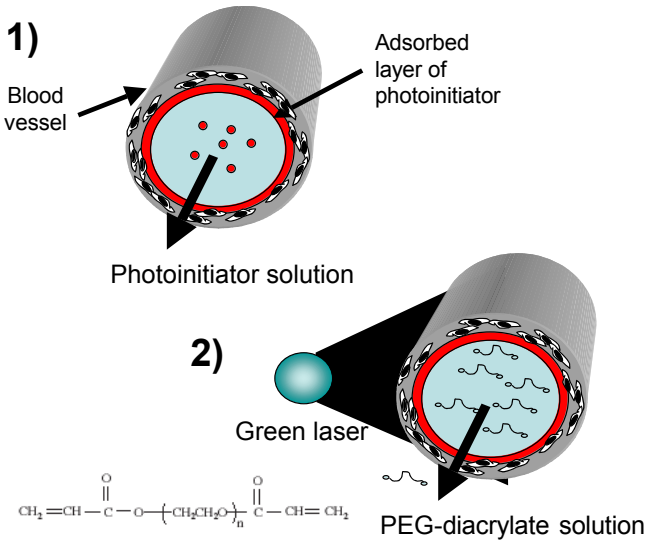
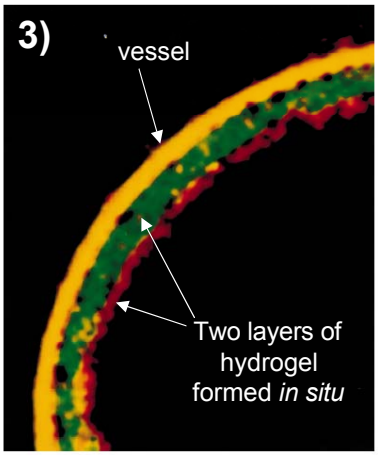


Controlled release

  - Tissue barriers (Hubbell[2,3])
    - Prevent thrombosis (vessel blocked by coagulating platelets) and restenosis (re-narrowing of blood vessel after operation) in vessels after vascular injury/angioplasty/etc.
    - Prevent tissue-tissue adhesion after an operation

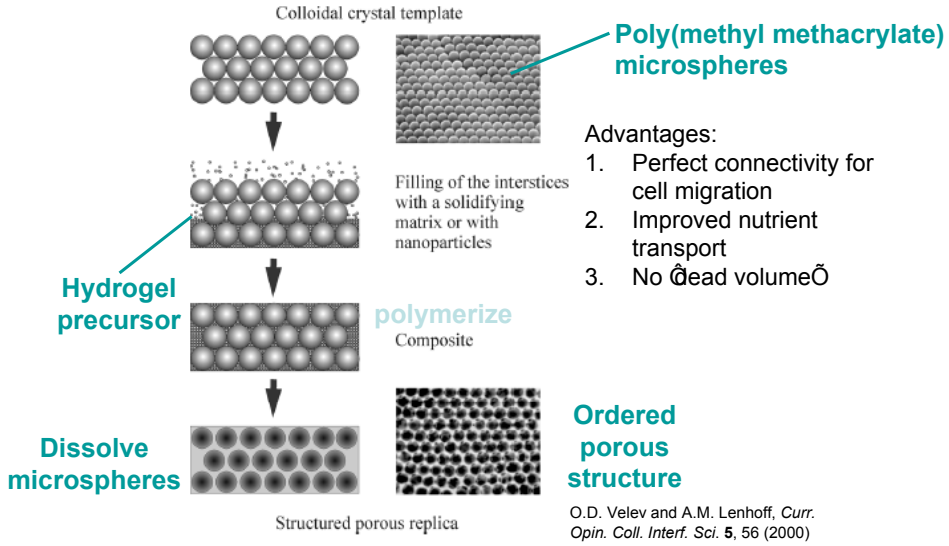
Tissue barriers and conformal coatings





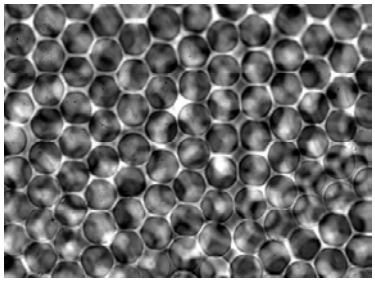
(An and Hubbell 2000)

- TE scaffolds/cell encapsulation/immunoisolation[4,5]

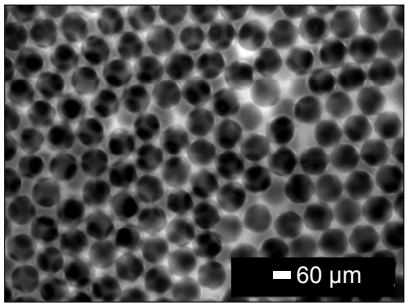


Hydrogel ôinverse opalsÕ

Optical micrograph/20 µm pores



Fluorescence micrograph/60 µm pores

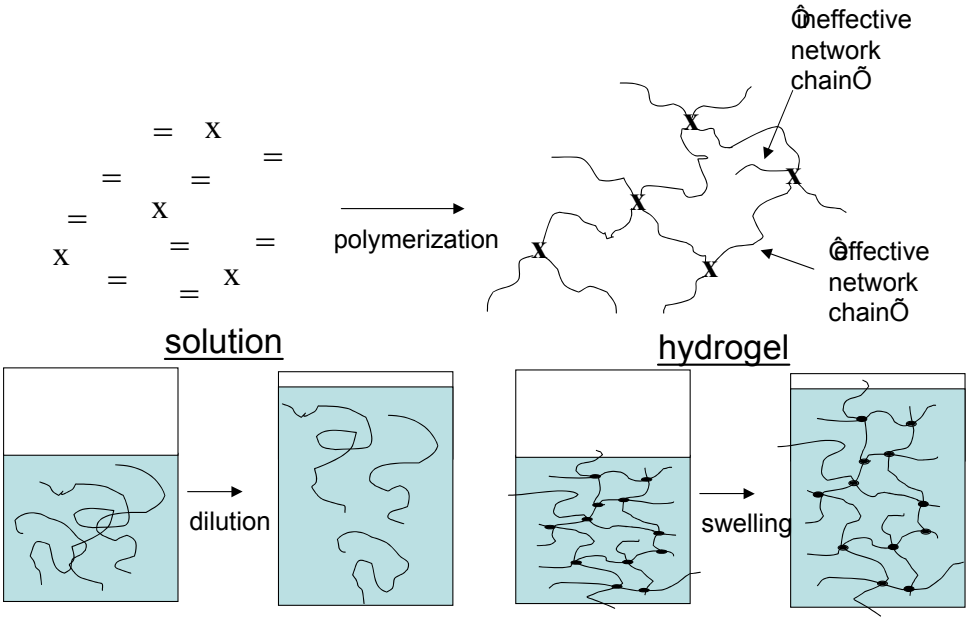


- Biosensors (to be covered later)
- Contact lenses

Structure of covalent hydrogel biomaterials

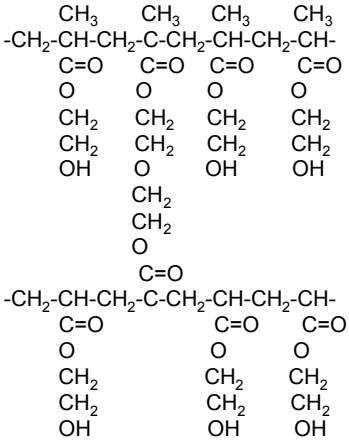
Chemical and physical structure

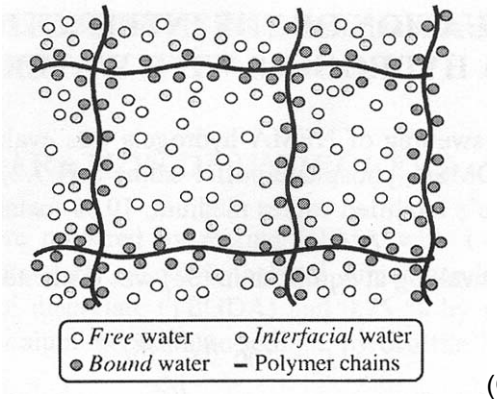
Structure and swelling of hydrogel materials



- Networks formed by stitching together monomers in aqueous solutions via cross-linkers that are multifunctional units
  - Draw an example of a crosslinker: bisacrylamide
  - networks from hydrophilic vinyl monomers
    - hydroxyethyl methacrylate
    - poly(ethylene glycol) methacrylate
    - acrylic acid
    - acrylamide, N-isopropylacrylamide
  - Common crosslinkers:
    - PEGDMA, EGDMA
    - bis-acrylamide
- Hydrogels undergo swelling in analogy to dilution of free polymer chains in solution
  - Difference lies in limit to 'dilution' when chains are cross-linked together (ENTROPIC)
- Poly(2-hydroxyethyl methacrylate) hydrogels
  - One of the first biomedical hydrogels; applied to contact lenses in late 1950s[6]

PEGDMA-co-PHEMA



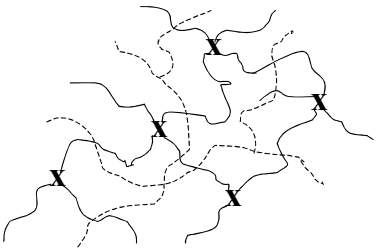


(Chielline et al.[7])

Interpenetrating networks

- Useful for obtaining gels with properties in between two different materials
  - E.g. mix a swelling polymer with a temperature- or pH-responsive polymer to obtain networks that have a defined amount of swelling in response to changes in temperature or pH

Interpenetrating networks

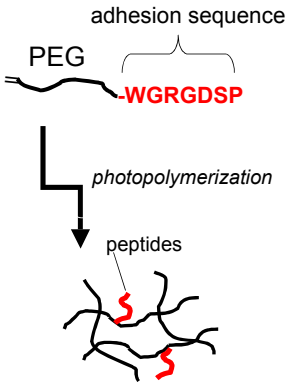


  - Sem-interpenetrating networks: second component is entangled with first network but not cross-linked

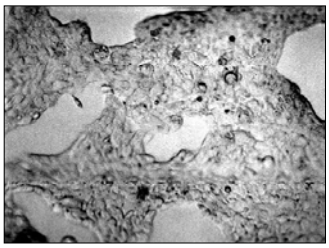
Biological recognition of hydrogels

- Inclusion of peptide-functionalized co-monomers allows hydrogels to have tailored biological recognition properties similar to solid degradable polymers
  - Promoting cell adhesion:

Incorporating biological recognition:

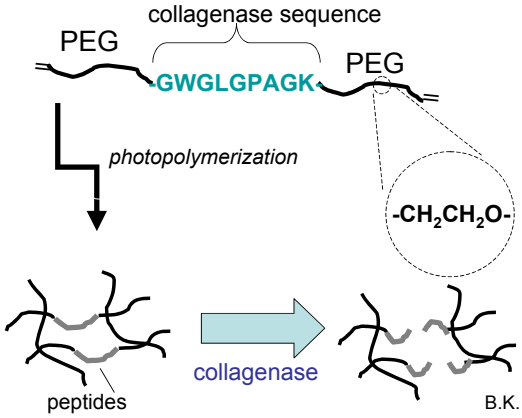


NR6 fibroblast adhesion on PEG-RGD hydrogel



(no cell adhesion on ligand-free hydrogels)

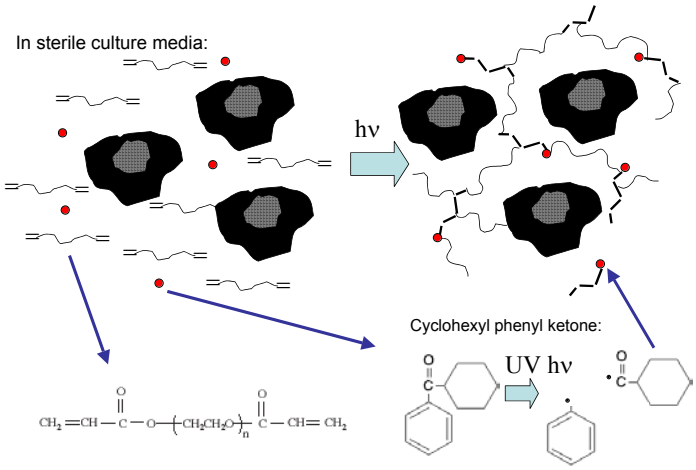
- Promoting remodeling/cell migration through synthetic networks:



B.K. Mann, A.S. Gobin, A.T. Tsai, R.H. Schmedlen, J.L. West, *Biomaterials* **22**, 3045 (2001)

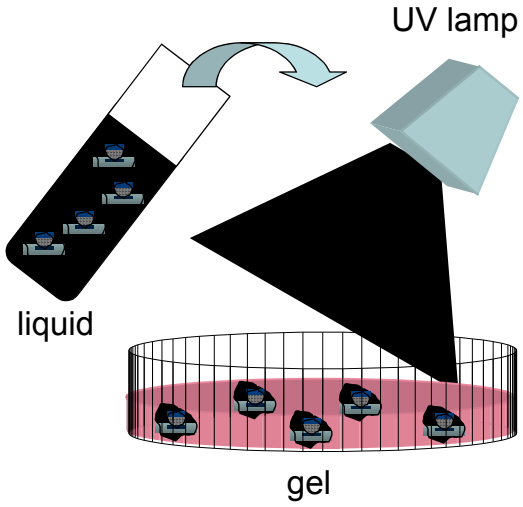
Example synthesis strategy: photoencapsulation of live cells[5]

- Photoencapsulation: expose solution of cells, prepolymer/cross-linker/monomer, and photoinitiator to light to initiate free radical polymerization



- Provides very rapid polymerization (2-20 seconds typical), at neutral pH and room temp. – 37°C
- 'soft' UV photoinitiators are common and non-toxic (illuminate at 365 nm)

- Cells can be entrapped with high viability[4,8]:



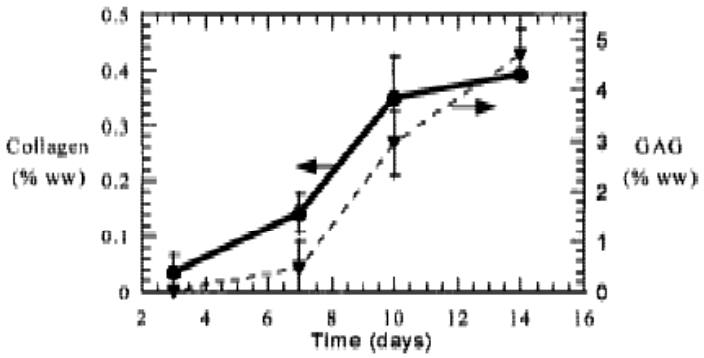


Figure 3. Biochemical analysis. Evolution of GAG and total collagen contents (% wet weight) over 14 days of photoencapsulated bovine chondrocytes.

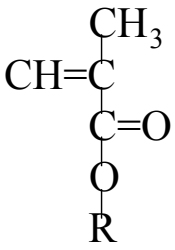
Example Biomedical Hydrogel Materials[6]

Formed from hydrophilic biocompatible polymers, often polymers that can be safely eliminated by the body if the gel breaks down.

TABLE 1 Important hydrogel polymers in medicine

Hydrogel polymer	Medical applications
Poly(vinyl alcohol) [PVA]	**Blood-Compatible Hydrogels**
Polyacrylamide [PAAm]	
Poly(N-vinyl pyrrolidone) [PNVP]	
Poly(hydroxyethyl methacrylate) [PHEMA]	
Poly(ethylene oxide) [PEO]	
Poly(ethylene glycol) [PEG]	
Poly(ethylene glycol) monomethyl ether [PEGME]	
Cellulose	
Poly(hydroxyethyl methacrylate) [PHEMA] copolymerized with:	**Contact Lenses**
NVP	
Methacrylic acid [MAA]	
Butyl methacrylate [BMA]	
Methyl methacrylate [MMA]	
3-methoxy-2-hydroxypropylmethacrylate [MHPM]	
PHEMA/poly(ethylene terephthalate) [PTFE]	**Artificial Tendons**
	Other Medical Applications
Cellulose acetate	Artificial kidney
PVA and cellulose acetate	Membranes for plasmapheresis
PNVP, PHEMA, cellulose acetate	Artificial liver
PVA and PHEMA	Artificial skin
Terpolymers of HEMA, MMA and NVP	Mammaplasty
PHEMA, P(HEMA-co-MMA)	Maxillofacial reconstruction
PVA	Vocal cord reconstruction
P(HEMA-b-siloxane)	Sexual organ reconstruction
PVA, poly(acrylic acid) [PAA], poly (glyceriyl methacrylate)	Ophthalmic applications
PVA, HEMA, MMA	Articular Cartilage
	Controlled Drug Delivery*
Poly(glycolic acid) [PGA], Poly(lactic acid) [PLA], PLA-PGA, PLA-PEG, Chitosan, Dextran, Dextran-PEG, polycyanoacrylates, fumaric acid-PEG, sebacic acid/1,3-bis(p-carboxyphenoxy) propane [P (CPP-SA)]	*Biodegradable hydrogels*
	Non-Biodegradable Hydrogels
PHEMA, PVA, PNVP, poly(ethylene-co-vinyl acetate) [PEVAc]	Neutral
Poly(acrylamide) [PAAm], Poly (acrylic acid) [PAA], PMAA, poly (diethylaminoethyl methacrylate) [PDEAEMA], poly (dimethylaminoethyl methacrylate) [PDMAEMA]	pH-Sensitive

(*continued*)



General methacrylates

TABLE 1 (*Continued*)

Hydrogel polymer	Medical applications
Poly(methacrylic acid-grafted-poly(ethylene glycol)) [P(MAA-g-EG)], poly(acrylic acid-grafted-poly(ethylene glycol) [P(PAA-g-EG)]	Complexing hydrogels
Poly(N-isopropyl acrylamide) [PNIPAAm]	Temperature-sensitive
PNIPAAm/PAA, PNIPAAm/PMAA	pH/Temperature-sensitive

*These drug delivery applications have been used for the controlled release of several therapeutic agents such as contraceptives, antiarrhythmics, peptides, proteins, anticancer agents, anticoagulants, antibodies, among others. This table does not include all the copolymers of such hydrogels.

Chemical structure of biodegradable hydrogels

***Mechanism I*: (non-degradable water-soluble polymers with degradable cross-links)**

- Degradable cross-links
  - e.g. dextran hydrogels[9]
    - bacterial exo-polysaccharide
    - branched polymer composed of α-1,6-linked D-glucopyranose residues with a low % of α-1,2 and 1,3 side chainsDextran with polylactide crosslinks: hydrolyzable crosslinks[9]

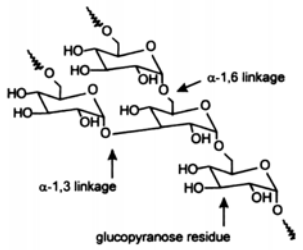


Figure 1. The chemical structure of dextran.

- dextran can be functionalized with methacrylate and then crosslinked in the presence of a small amount of vinyl monomer:

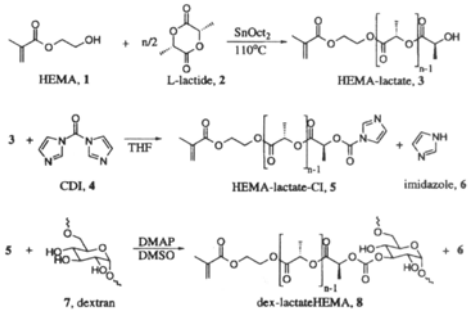


Figure 8. Reaction scheme for the synthesis of dex-lactateHEMA.

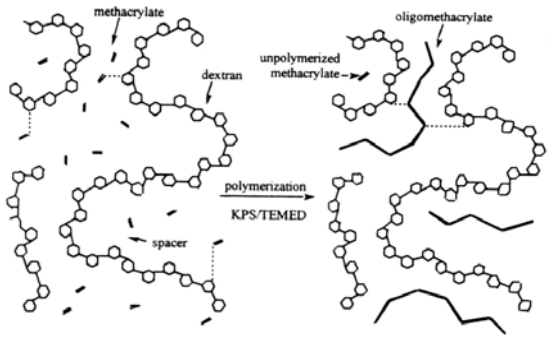


Figure 3. Schematic representation of the formation of dextran hydrogels.

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degradable gels show first swelling then dissolution as cross-links are hydrolyzed:

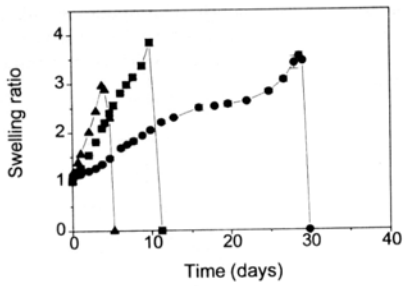


Figure 12. Swelling behavior of dex-HEMA (●), dex-lactate-HEMA (■) and dex-lactate$_2$-HEMA (▲) hydrogels in aqueous solution (pH 7.2, 37 °C). The initial water content of the hydrogels was 80%, the degree of methacryloyl substitution was approximately 6.

Mechanism III:

- Co-encapsulation of degradation catalyst
  - e.g. dextran hydrogels[9] encapsulating dextranase enzyme
    - polymerization is carried out in the presence of protein to be delivered and a bacterial dextranase: dextranase breaks down the dextran chains over time, releasing protein
    - degradation/protein release rate depends on amount of enzyme encapsulated

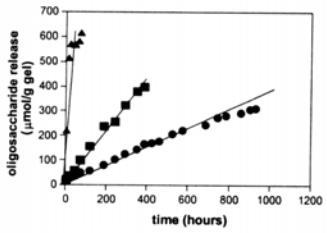


Figure 6. The cumulative release of reducing oligosaccharides from dex-MA hydrogels (DS 4, initial water content 70% w/w) containing dextranase (0.03 U/g gel (●), 0.1 U/g gel (■), 1 U/g gel (▲).

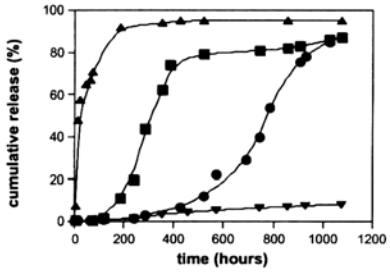


Figure 7. The cumulative release of IgG from dextran hydrogels (DS 4, initial water content 70% w/w) containing dextranase ((▼), 0.03 U/g gel (●), 0.1 U/g gel (■), 1 U/g gel (▲)).

Thermodynamics of hydrogel swelling

Derivation of the free energy of polymer chains cross-linked in the presence of solvent

- Theory originally developed by Flory and Rehner for solid rubber networks exposed to solvent[10,11]
- Adapted to describe hydrogels in biomedical applications by Bray and Merrill[12]

Description of the model

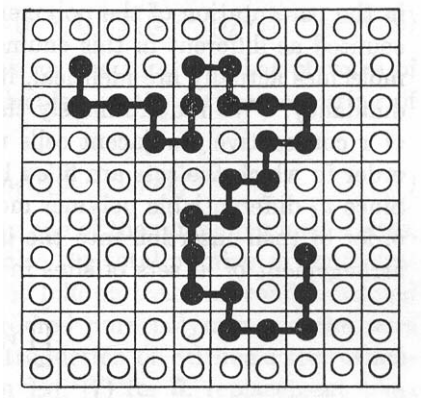
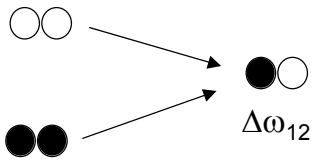


FIG. 110.—Segments of a chain polymer molecule located in the liquid lattice.

Polymer and solvent (water) are modeled as *segments* of equal volume- polymer chains are composed of connected segments

Energy of contacts:



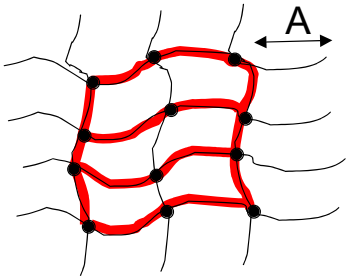
(Flory[13])

Model parameters

μ_1^{bath}	chemical potential of water in external bath (= μ_1^0)
μ_1	chemical potential of water in the hydrogel
μ_1^0	chemical potential of pure water in standard state
Δw_{12}	pair contact interaction energy for polymer with water
z	model lattice coordination number
x	number of segments per polymer molecule

M	Molecular weight of polymer chains before cross-linking
M_c	Molecular weight of cross-linked subchains
n_1	number of water molecules in swollen gel
χ	polymer-solvent interaction parameter
k_B	Boltzman constant
T	absolute temperature (Kelvin)
$v_{m,1}$	molar volume of solvent (water)
$v_{m,2}$	molar volume of polymer
$v_{sp,1}$	specific volume of solvent (water)
$v_{sp,2}$	specific volume of polymer
V_2	total volume of polymer
V_s	total volume of swollen hydrogel
V_r	total volume of relaxed hydrogel
ν	number of subchains in network
ν_e	number of 'effective' subchains in network
ϕ_1	volume fraction of water in swollen gel
$\phi_{2,s}$	volume fraction of polymer in swollen gel
$\phi_{2,r}$	volume fraction of polymer in relaxed gel

- Subchains, M_c, and 'effective' chains



Assume cross-links are randomly placed; on average, all are equidistant
ν = number of subchains in cross-linked network
ν_e = number of "effective" subchains: tethered at both ends
M = MW of original chains
M_c = MW of subchains = MW between cross-links

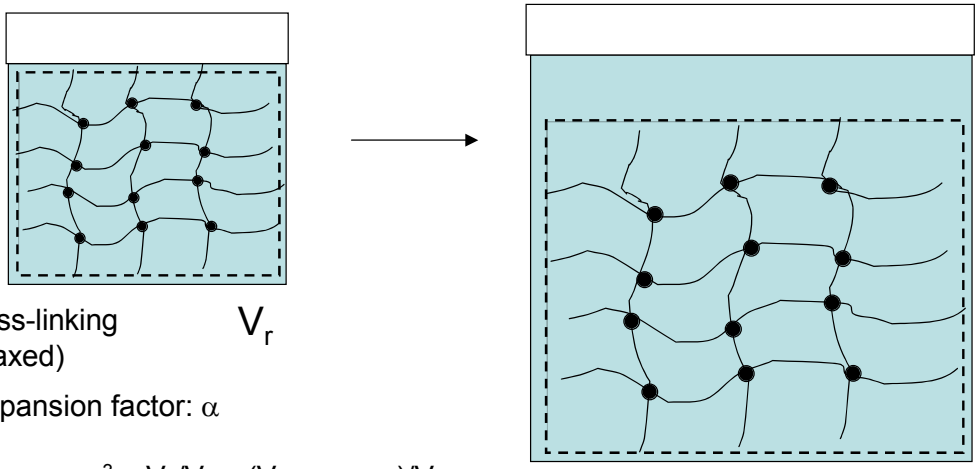
Example: assume polymer chains have a molecular weight M = 4A and each "subchain" has molecular weight A:

$\nu = 24$ $\nu_e = 12$

Two useful relationships:

$\nu = V_2/v_{sp,2}M_c$
$\nu_e = \nu(1 - 2(M_c/M))$

- Physical picture of the equilibrium described:
  - Polymer chains are cross-linked in water
  - Relaxed network is moved to a large bath of water and swells to a new equilibrium



Cross-linking (relaxed) V_r

Expansion factor: α

$\alpha_x\alpha_y\alpha_z = \alpha^3 = V_s/Vr = (V_2 + n_1v_{m,1})/V_r$

swelling V_s

$\phi_{2,s} = V_2/(V_2 + n_1v_{m,1})$ volume fraction of polymer in swollen gel

$\phi_{2,r} = V_2/V_r$ volume fraction of polymer in relaxed gel

Derivation of the equilibrium properties

- We want to calculate the change in free energy as the network is cross-linked and first exposed to a surrounding solvent bath that can trigger solvent to enters/leave the hydrogel
- The free energy of the system can be written as a contribution from mixing and an elastic retracting energy:

$$\Delta G_{total} = \Delta G_{mix} + \Delta G_{el}$$

At equilibrium, the chemical potentials of solvent inside and outside the gel are equal:

Eqn 1 $\mu_1^{bath} = \mu_1$

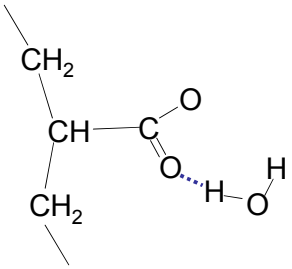
Eqn 2 $\mu_1^0 = \mu_1$ **chemical potential of bath is water's standard state**

Eqn 3
$$0 = \Delta(\mu_1)_{total} = \left(\frac{\partial(\Delta G_{total})}{dn_1}\right)_{T,P} = \Delta(\mu_1)_{mix} + \Delta(\mu_1)_{el}$$

- $\Delta(\mu_1)_{mix}$ and $\Delta(\mu_1)_{el}$ will depend on the degree of swelling and thus allow us to calculate the swelling if we know the physicochemical parameters of the system…
- Determining the contribution from mixing:
  - Based on Flory's original lattice liquid model

Eqn 4 $\Delta G_{mix} = \Delta H_{mix} - T\Delta S_{mix}$

- Free energy can be decreased by entropy gain on mixing (more configurations, $\Delta S_{mix} > 0$) and favorable solvent-polymer interactions ($\Delta H_{mix} < 0$)



  - …drives SWELLING of hydrophilic networks in water
- Enthalpy of mixing: count contacts and provide $\Delta\omega_{12}$ energy per contact:
  - $\Delta\omega_{12}$ accounts for energy of moving a molecule of solvent from pure water into pure polymer
  - # contacts between 1 and 2 = (total number of polymer segments in system)(# contacts with solvent)
    = $(n_2 x)$[(number neighbors per segment)(probability that neighbor is solvent)]
    = $(n_2 v_2)(z)(\phi_1) = z n_1 \phi_2$

Eqn 5 $\Delta H_{mix} = z\Delta\omega_{12} x_1 n_1 \phi_2$

- Define the polymer-solvent interaction parameter:

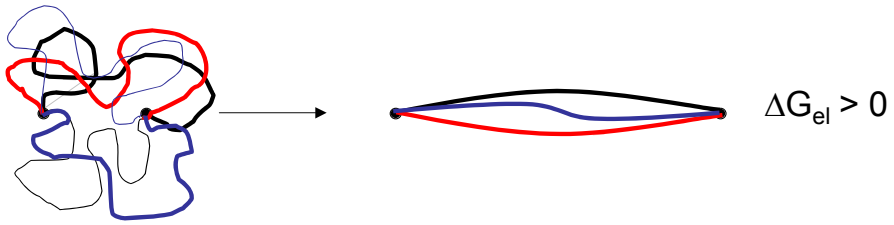
Eqn 6 $\chi = z\Delta\omega_{12} x_1 / k_B T$ **(unitless)**

Eqn 7 **therefore** $\Delta H_{mix} = k_B T n_1 \phi_2$

- Now derive ΔS_{mix}: we won't derive it here:
  - Based on fundamental equation:

Eqn 8 $S = k_B \ln \Omega$

- Where Ω is the number of configurations possible in the system.
- Lower configurational entropy if chains of network are stretched



- Resists chain stretching, competes *against* ΔG_{mix} and ΔG_{ion}, driving network *collapse*
- Flory derived an expression for the # ways free polymer chains could be arranged on the lattice:

Eqn 9 $\Delta S_{mix} = k_B \ln(\Omega^{solution}/\Omega^{separate}) = -k_B [n_1 \ln \phi_1 + n_2 \ln \phi_2]$

- For a gel, the number of 'free' polymer chains n2 = 0, so:

Eqn 10 $\Delta G_{mix} = k_B T[n_1 \ln \phi_1 + \chi n_1 \phi_2]$

- The chemical potential change can be obtained by differentiating Eqn 10:

Eqn 11

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