Novel Hyaluronic Acid Derivatives to Alleviate Osteoarthritis

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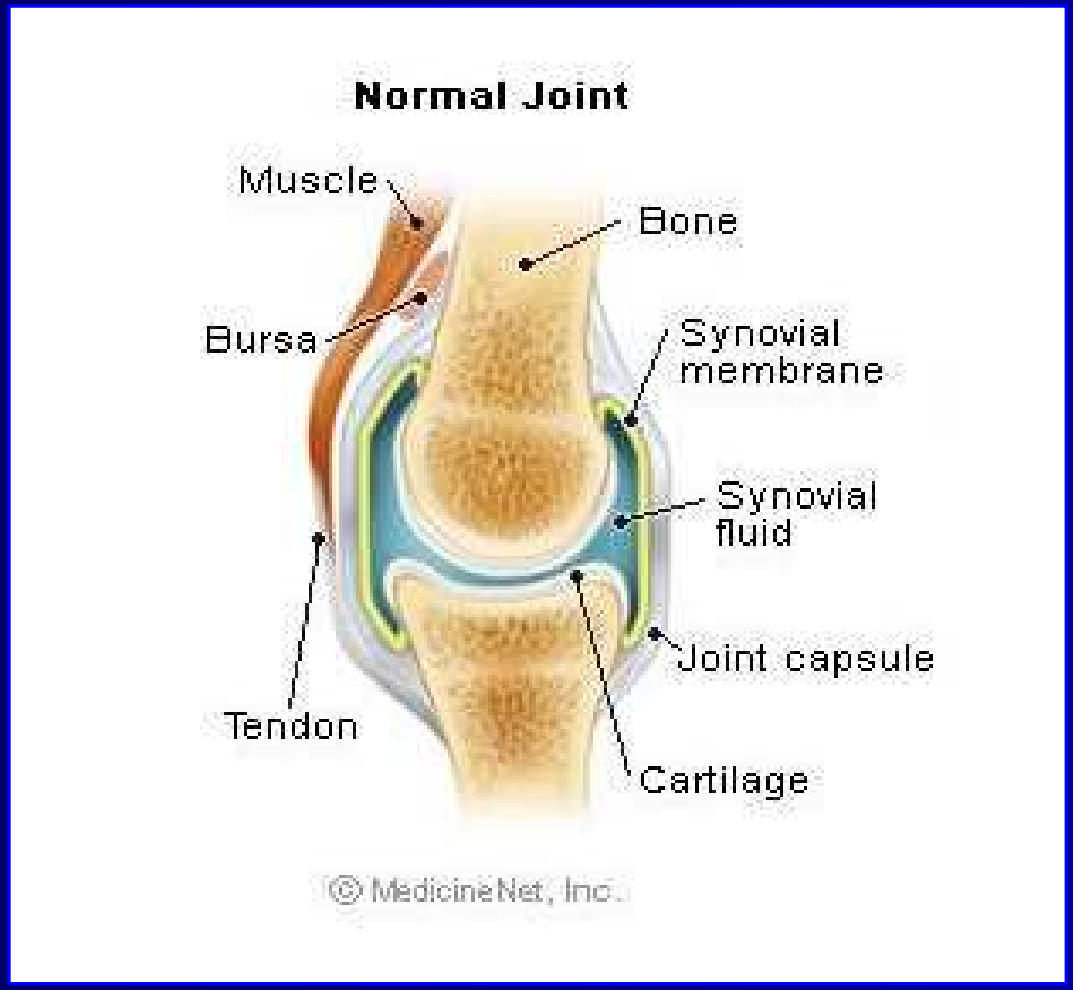
Objective

- To design a polymer to be used in non-surgical injections to relieve symptoms of osteoarthritis
  - Increased retention time
  - Maintain flexibility and strength
  - FDA approved
  - Reasonable cost

Overview

- Osteoarthritis
- Current Treatments
- Novel Hyaluronic Acid Derivative
- Demand
- FDA
- Conclusions

The Knee Joint



www.medicinenet.com/ osteoarthritis/page2.htm

Articular Cartilage

- Transmits load from one surface to another
- 2-3 mm thick
- Components
  - Water (70%)
  - Collagen (10-20%)
  - Proteoglycans (5-10%)
  - Chondrocytes (~5%)

Synovial Fluid

- Fills gap between joints
  - Approx. 50 μm thick
    - During walking can be 0.8-1.5 μm
  - Volume ~1 ml
- Enclosed in synovial membrane
- Major components
  - Water
  - Proteoglycans
  - Hyaluronic Acid (HA)

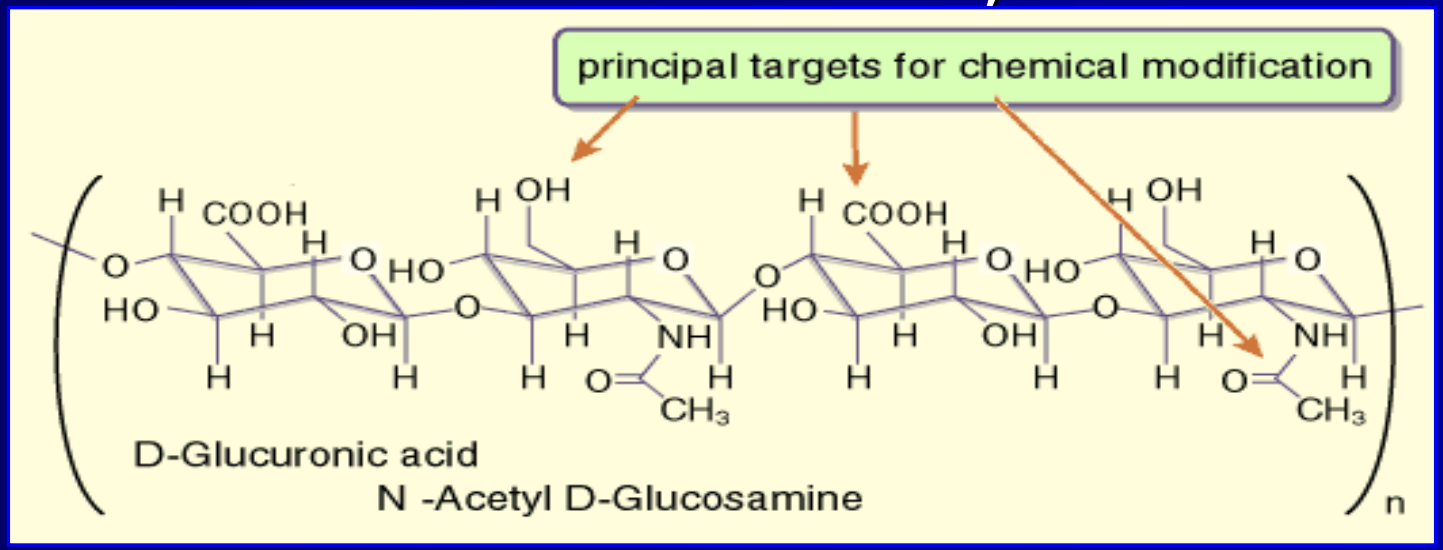
Synovial Fluid

- Properties
  - Viscosity ~ 300-10,000 cP
  - Coefficient of friction ~ 0.02
    - Compare to 0.03 for ice-on-ice
- Main contributor to these properties

  →**Hyaluronic Acid**

Hyaluronic Acid (HA)

- Structure
  - Linear repeating disaccharide
    - D-glucuronic acid and *N*-acetyl-D-glucosamine
  - Number of units = 500 - 25,000



http://www.glycoforum.gr.jp/science/hyaluronan/HA01/HA01E.html

HA in Synovial Fluid

- Average molecular weight for healthy knee
  10^5 – 10^7 Daltons
- Concentration for healthy adult
  3.4 mg/ml
- Non-Newtonian shear thinning
- Viscosity – variable
  - Increases with increasing molecular weight
  - High at low shear rate
  - Low at high shear rate

The Disease

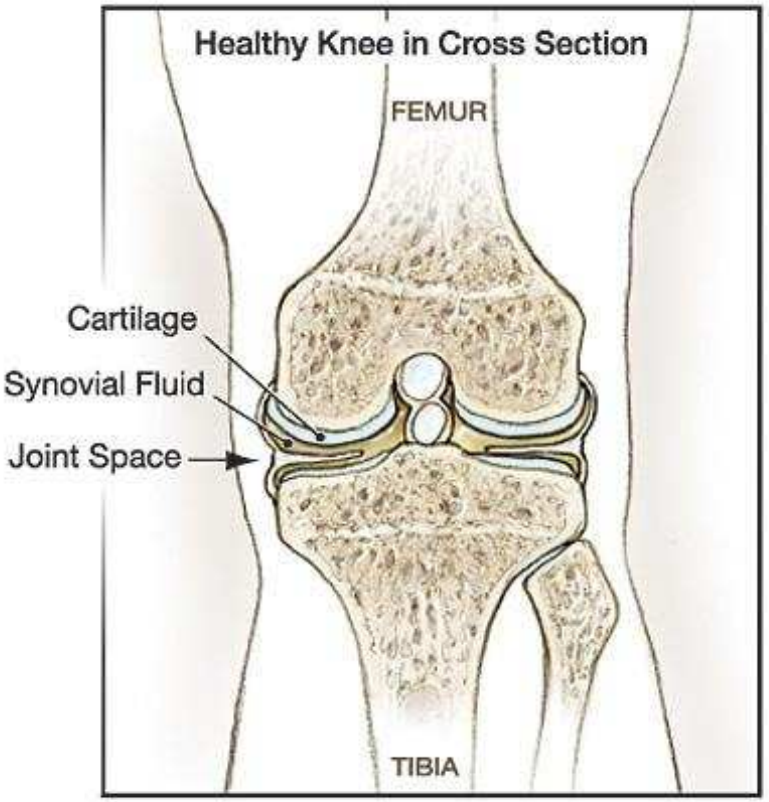
Osteoarthritis – General Facts

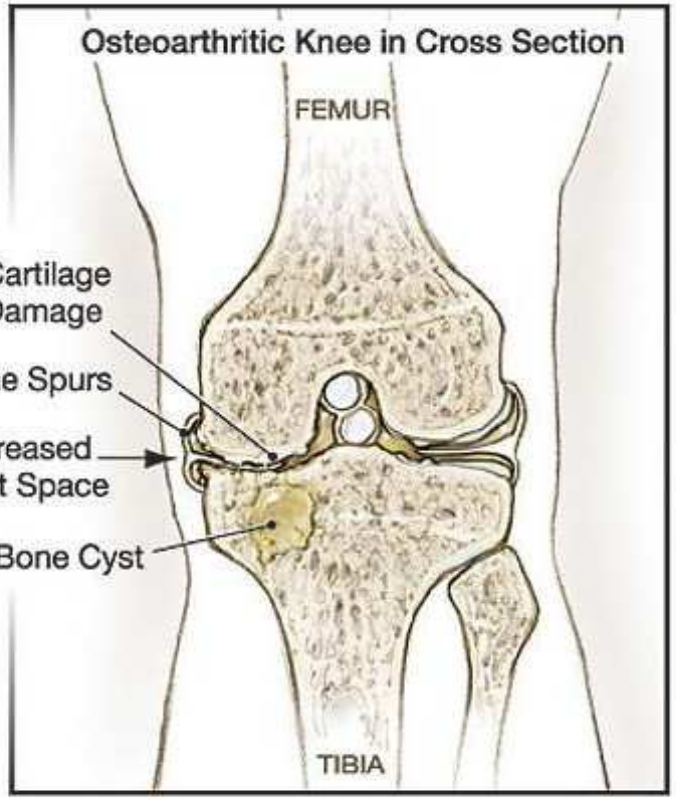
- Over 23 million Americans affected
- Most common in people 65+
- Anyone susceptible
  - Impact injuries
  - Obesity
  - Prolonged elevated activity
- **$1.5 billion/year industry in US**

Osteoarthritis

- Most common in load-bearing joints
  - Knees, hip, lower back, neck
- Possibly due to decreased lubrication and load bearing properties of cartilage and synovial fluid
- Leads to degeneration of cartilage and eventually bone-on-bone contact

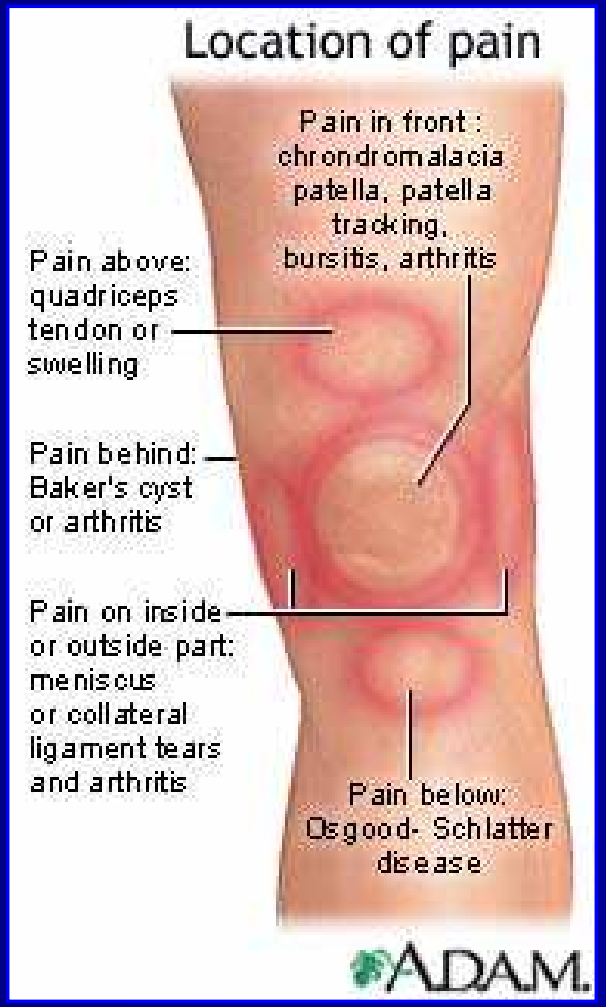
Normal vs. Osteoarthritic Knee





Symptoms

- Mild to severe pain
- Limited range of motion
- Approx. 25% of patients unable perform daily functions



http://adam.about.com/encyclopedia/19506.htm

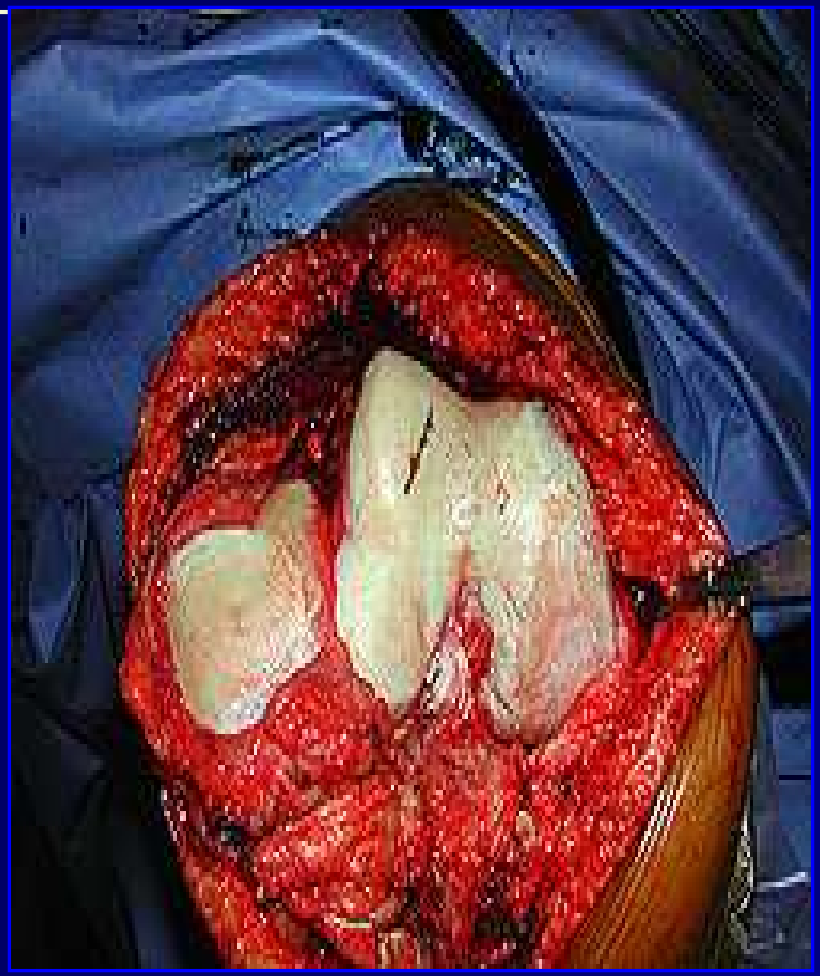
Current Treatments

- Physical therapy
  - exercise techniques
- Oral medication
  - Tylenol, Ibuprofen, Celebrex
  - Effective for moderate pain
- Total Knee Replacement
- Hyaluronic acid (HA) injections

Current Treatments – Total Knee Replacement

- Advantages:
  - Last 15-25 years
- Disadvantages:
  - Time away from work
    - Desk work (3-6 weeks)
    - Labor intensive work (several months)
  - Requires extensive physical therapy
  - Risk of infection
  - Cost ~ $15,000

Total Knee Replacement



www.raphaelmosseri.com/ mi_genou_uk.html

Current Treatments – Hyaluronic Acid Injections

- Intra-articular injections of HA derivatives
  - Series of injections, must be repeated after 6 months
  - Aims to increase the viscoelasticity of the synovial fluid

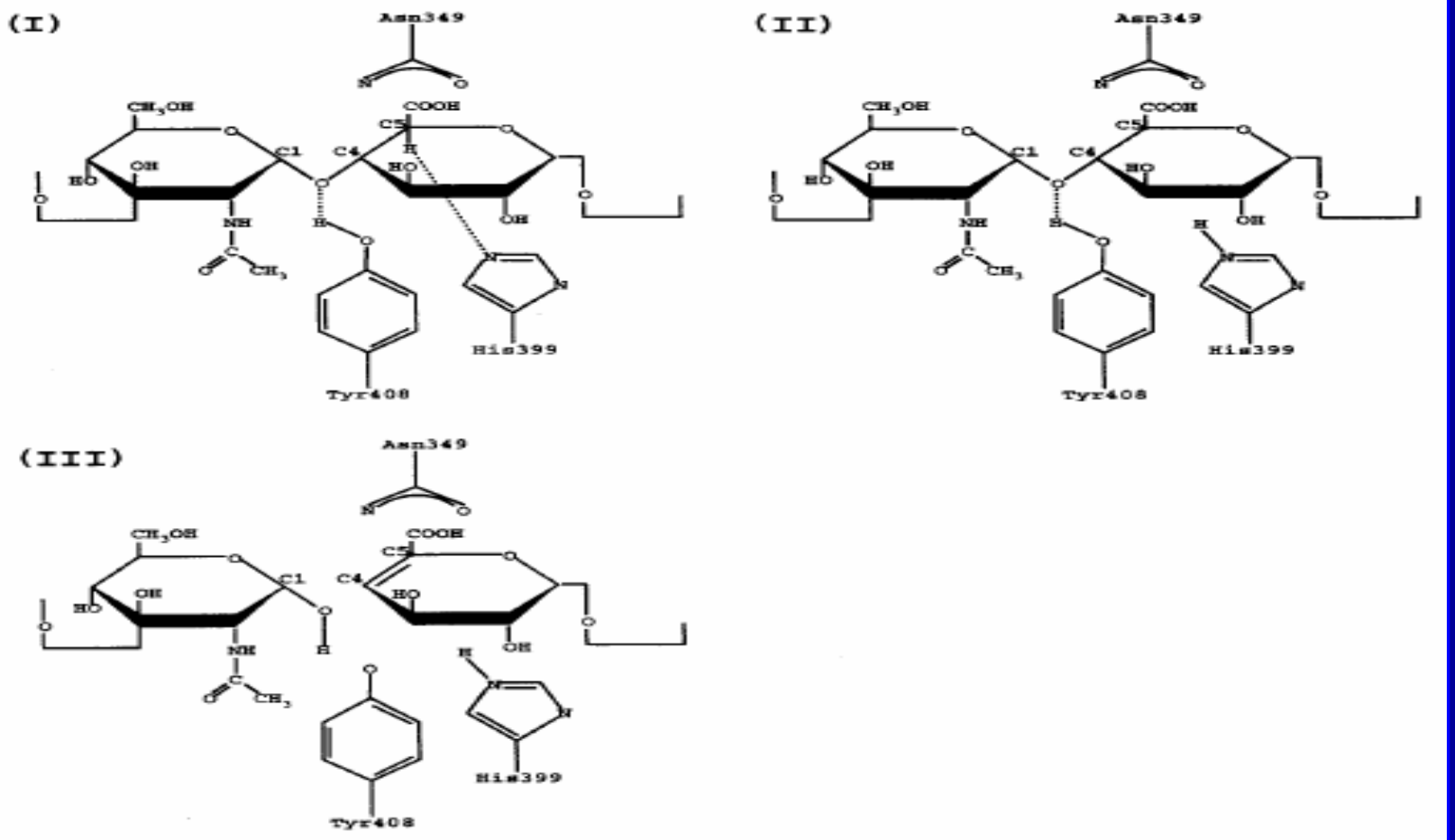
Available HA Injections –

- HYALGAN®
  - Similar in structure and properties to natural hyaluronic acid
  - Mixed results
- Hylan G-F 20 (SYNVISC®)
  - Crosslinked HA
  - Viscosity and elasticity near that of healthy 18-27 year old adult
  - Last up to 6 months
  - $620 for 3-week treatment

Problems

- Current HA injections appear to degrade over time in the body
- It has been proposed by some that the mechanism by which this degradation is occurring is through bond breaking (example of mechanism shown on next slide)

Problem – Degradation



Karthe Ponnuraj and Mark J. Jedrzejas, "Mechanism of hyaluronan binding and degradation: structure of *Streptococcus pneumoniae* hyaluronate lyase in complex with hyaluronic acid disaccharide at 1.7 Å resolution", J. Mol. Biol. (2000) 299, 885-895

Solution – First Consideration

- CF_3 modified HA
  - Increase the dipole to increase viscosity
  - Reduce degradation rate
  - Enzyme used to initiate reaction
  - Low cost of reagents
  - *Problem: Reports of toxicity*

Our Solution

HYAL-VYNE®

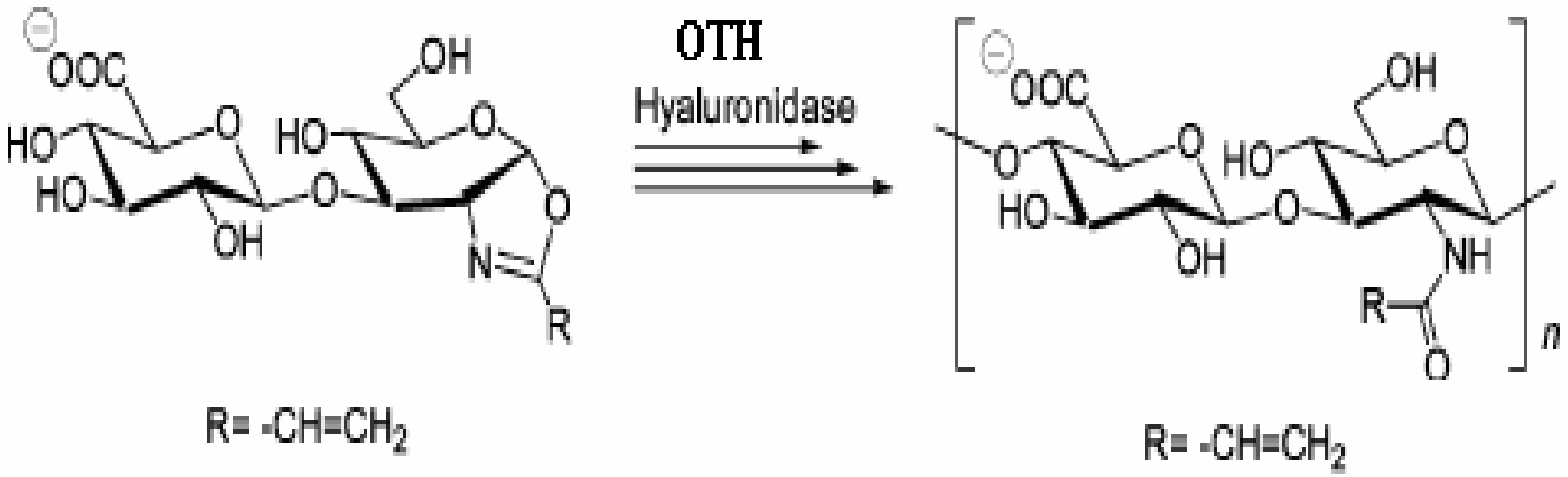
HYAL-VYNE®

- High molecular weight hyaluronic acid derivative

  ~10^6 Da
- HA modified with 2-vinyl
- New crosslinker introduced
- Forms a viscoelastic hydrogel

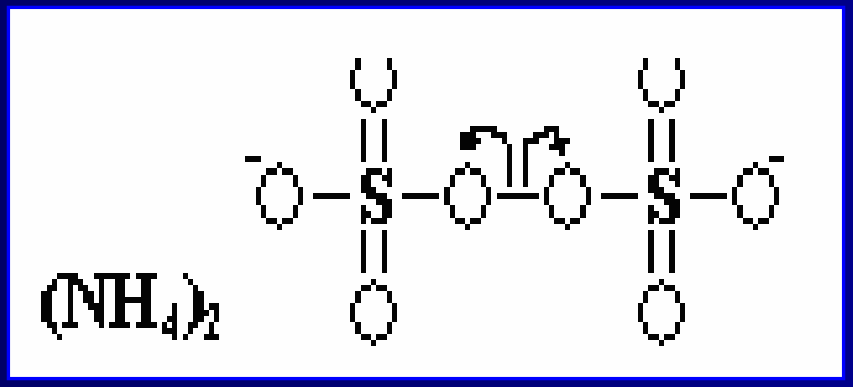
Modified HA

- Hyaluronic acid polymer modified with 2-vinyl (R = -CH=CH_2)
  - Polymerized using Ovine Testicular HAase (OTH)



Crosslinker

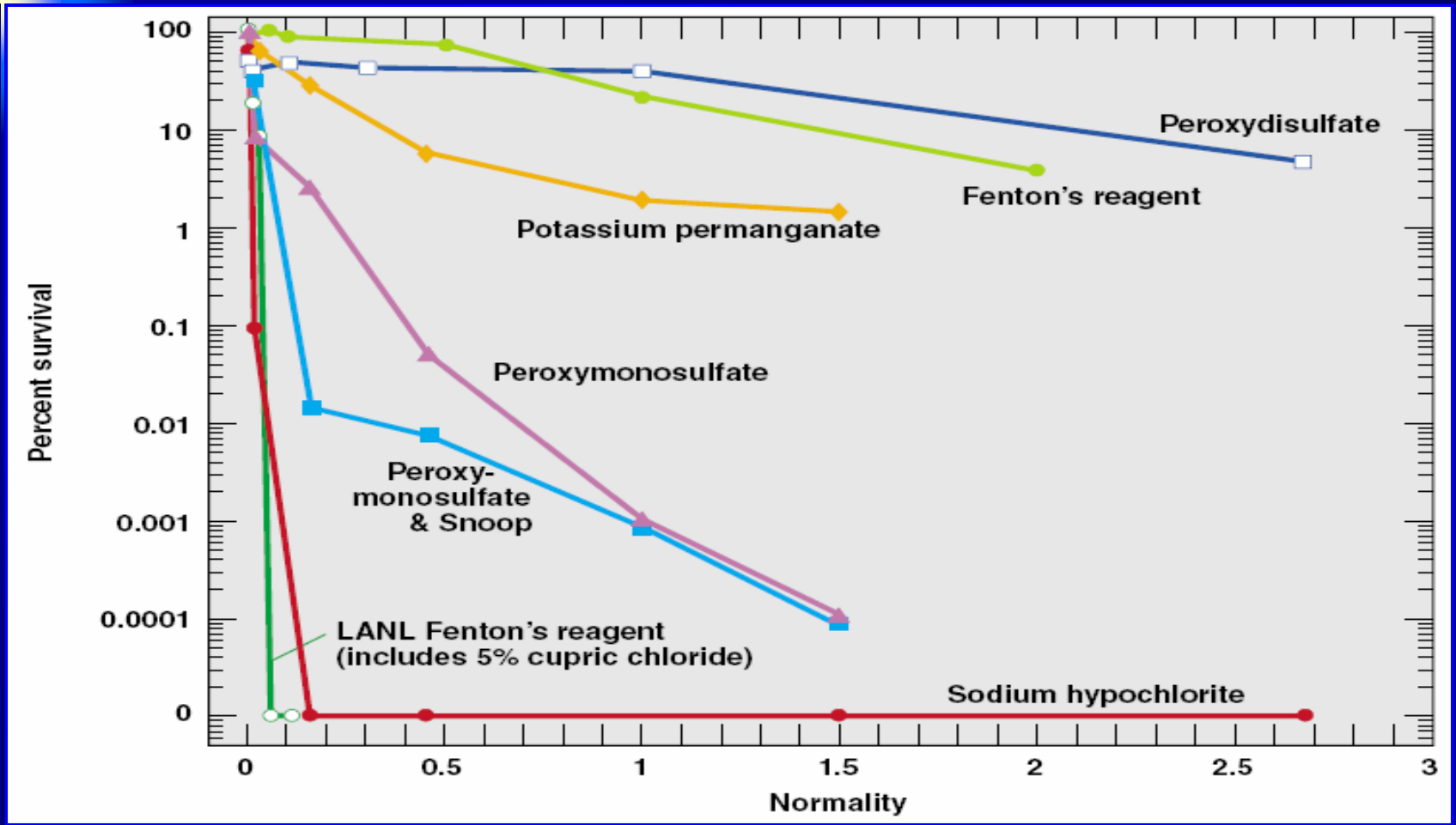
- Ammonium Peroxydisulfate
  - Chemical Formula = $(NH_4)_2S_2O_8$
  - Molecular Weight = 228.18 g/mol
  - Cost = $3.61/500g



Crosslinker

- Advantages
  - Sulfate has been proven to reduce degradation rates
  - Ability to stabilize a structure against denaturation
  - Non-toxic

Crosslinker



http://www.llnl.gov/ipac/technology/profile/environment/GelledDecontaminant/UCRL_AR_143212.pdf

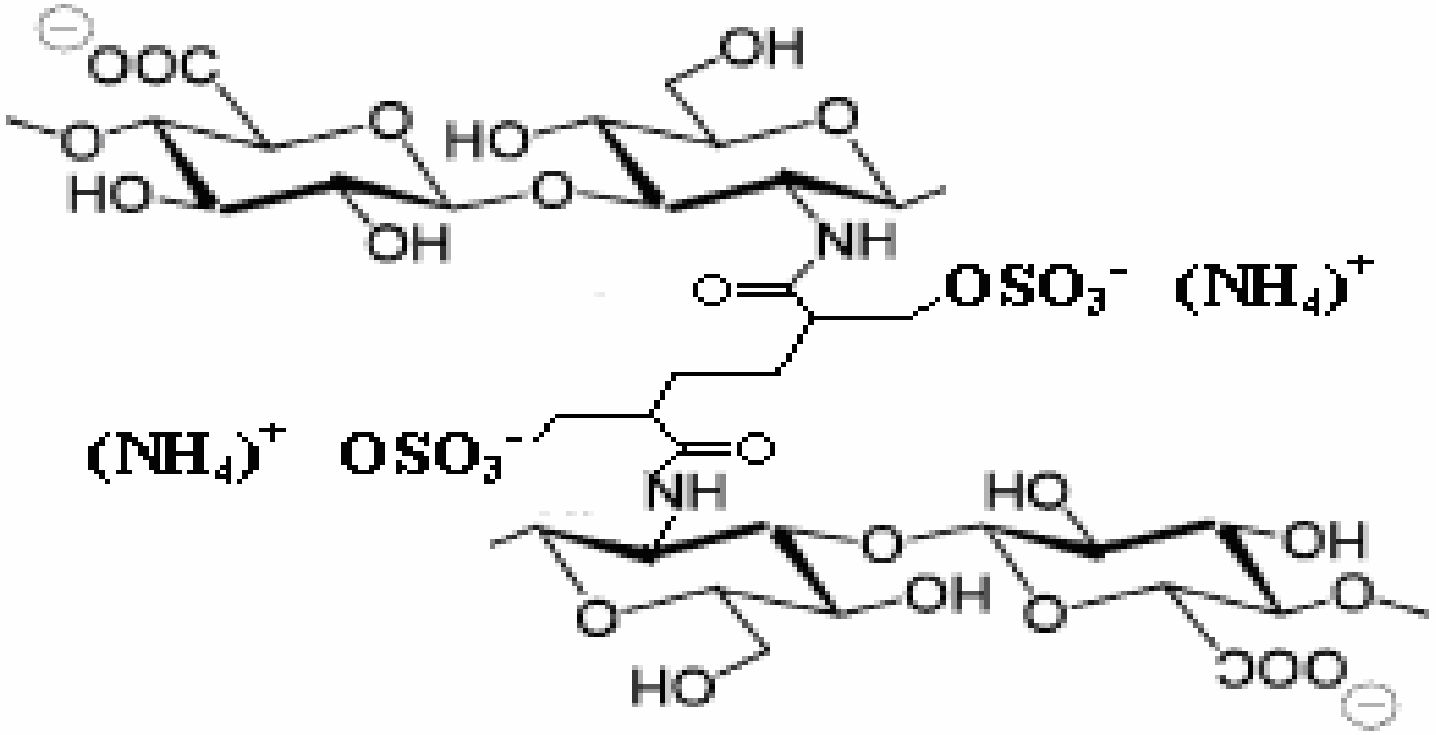
Crosslinking

- Provides greater stability
- Increases size of structure for higher-molecular weight
- Effects the solutions properties such as
  - strength
  - viscosity

Hydrogels

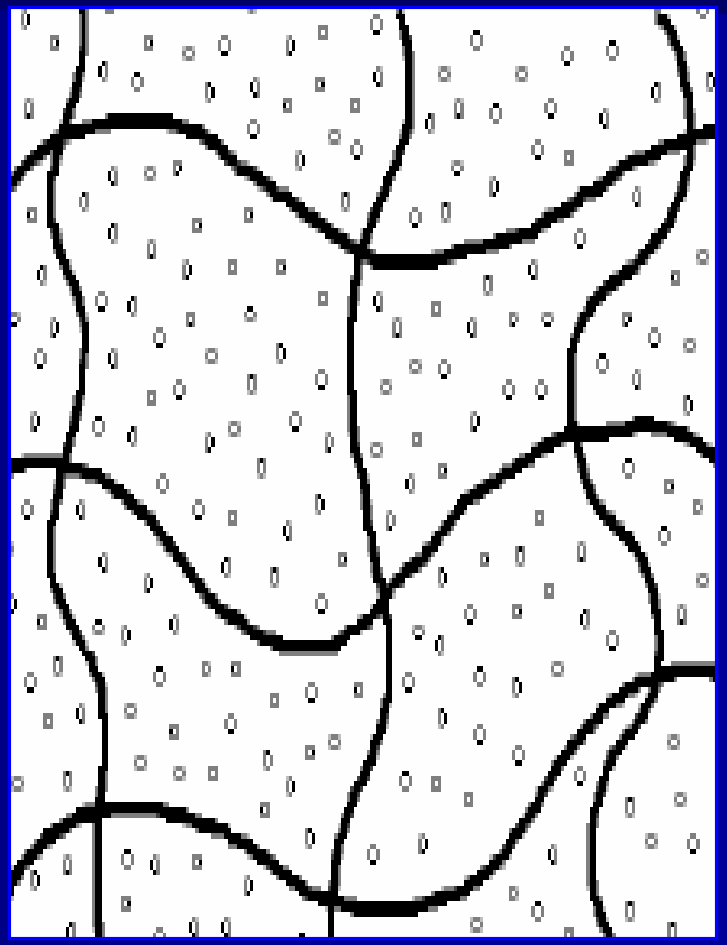
- 3D network of crosslinked hydrophilic polymer chains
- Absorb water
  - Able to retain shape upon loading and unloading
  - Able to absorb more than 20% of their own weight

HYAL-VYNE®



HYAL-VYNE®

- Required concentrations (mol/ml)
  - HA = $2.8*10^{-4}$
  - Crosslinker = $5.5*10^{-5}$
- Reaction controlled by time



HYAL-VYNE ®

- Viscosity ~ 16 Pa·s
- Molecular weight ~ 3 million Daltons
- Crosslinks per polymer chain ~ 24
- Monomers per polymer chain ~ 44
- Total number of monomers ~ 6700
- Total number of crosslinks ~ 1000

Lubrication Theory

- Elastohydrodynamic Lubrication (EHL)
  - Applied in cases of low geometric conformity subject to elastic deformation
  - Applies to most biological systems
  - Synovial fluid falls into Elastic-Isoviscous Regime
  - Use theory to calculate minimum film thickness of new gel to compare to normal synovial fluid

Minimum Film Thickness

- According to the Dowson-Higginson equation, the minimum film thickness for EHL is

$$h_{\min} = 1.6 \frac{\alpha^{0.6} \left(\mu_0 \tilde{u} \right)^{0.7} E'^{0.03} R^{0.43}}{w^{0.13}}$$

Where μ_0 = viscosity at atmospheric pressure
$\tilde{u}$ = effective speed
E' = reduced Young's modulus
R = effective radius
w = load
α = material constant

Minimum Film Thickness

- Parameters:
  - $\tilde{u} = U_0/2 = (0.03+0.03)/2 = 0.03$ m/s
  - R = 0.20 m
  - w = 2.6 MPa
  - $\alpha = 9.9 \times 10^{-9}$
  - E’ = 66.5 kPa

	HYAL-VYNE®	Synovial Fluid
Viscosity, μ_0	16 Pa·s	15.3 Pa·s
Minimum film thickness, h_{min}	1.54 μm	1.50 μm

Demand

Demand Equations

$$p_1 d_1 + p_2 d_2 \leq Y$$

$$p_1 d_1 \beta = \alpha p_2 d_2 \frac{d_1^{\alpha}}{d_2^{\beta}}$$

$$d_1 + d_2 \leq D$$

Parameters

- Product demands, d_1 and d_2 = **???**
- New treatment cost, p_1 = \$2400/injection
- Current treatment cost, p_2 = \$1300/year
- Total market demand, D = 7 million
- Total spent on treatment, Y = \$1.5 billion/ year
- $\alpha \rightarrow$ amount costumers know about new treatment relative to others
- $\beta \rightarrow$ measure of how much better new treatment is compared to competitors

Happiness Function

Attributes	W_i	Design Variables	Min	Max	y_i
Frequency of Treatments	0.75	Crosslink Density	-10.00%	10.00%	10.00%
Pain of Injection	0.125	Molecular Weight	-20.00%	20.00%	20.00%
Cost	0.125	Injection Volume	-5.00%	5.00%	-5.00%
		Viscosity	-10.00%	10.00%	-10.00%

- Old Treatment Happiness = 40%
- **New Treatment Happiness = 62%**

Demand

$$\beta = \frac{\text{competition's happiness}}{\text{new treatment happiness}}$$

$$\beta = \frac{40\%}{62\%} = 0.645$$

- Year (0) = .645
- Changes over time due to improvements in competitor products
- Assume α gradually increases until new product is equally known

Demand

Year	Alpha, α	Beta, β
0	0	0.645
1	0.15	0.715
2	0.4	0.785
3	0.89	0.855
4	0.99	0.925
5	1	0.995
6	1	1.065
7	1	1.135
8	1	1.205
9	1	1.275
10	1	1.345

Demand

- Total demand changes each year
  - Year 1 = 370,000 people
  - Year 2 = 621,000 people
  - Year 3 = 625,000 people (max)
- By Year 6 it is expected that a better treatment will be created due to the increasing competition

FDA Approval Process

FDA Approval - Outline

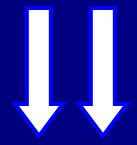
- Classification
- Type of Premarket Approval
- Necessary Experiments
- Possible Scenarios for success/failure
- Time and Money requirements

Product Classification

- Drug or Device?
  - Drug: used for diagnosis or treatment of disease or to affect the structure or function of the body
  - Device: used for diagnosis or treatment of disease or to affect the structure or function of the body, ***but does not depend on metabolic process to achieve primary purpose***

Product Classification

HYAL-VYNE



Medical Device

Device Classification

- Class I: General Controls
  - Least stringent; minimal risk
- Class II: Special Controls
  - More regulations than Class I; no life-threatening health risks
- Class III: Premarket Approval
  - Most strict control; often intended to prevent or treat disease or sustain human life; require extensive review before marketing

Premarket Approval (PMA)

- Traditional PMA
  - All non-clinical and clinical tests completed, then PMA submitted to FDA all at once
  - If denied, possibly have to start completely over
- Modular PMA
  - Non-clinical and clinical tests divided into modules, information from one module reviewed by FDA at a time
  - Allows for easier reassessment in case of denial

Modular PMA

- First a PMA shell must be submitted
  - No predetermined format, customized for particular device
  - Outlines experiments to be conducted in each module
  - Gives approximate time of completion

PMA Shell

HYAL-VYNE		
Module Number	**Contents**	**Time to Complete**
1	Nonclinical Laboratory Studies: Physical and Chemical Property Tests Degradation Tests Toxicity Tests	3 Years
2	Nonclinical Laboratory Studies: Animal Testing Sterilization and Packaging Injection Procedure	3 Years
3	Clinical Studies: Human Patient Testing Physician Instructions Patient Instructions	5 Years

FDA Approval Process Modeling

First-Stage Decisions

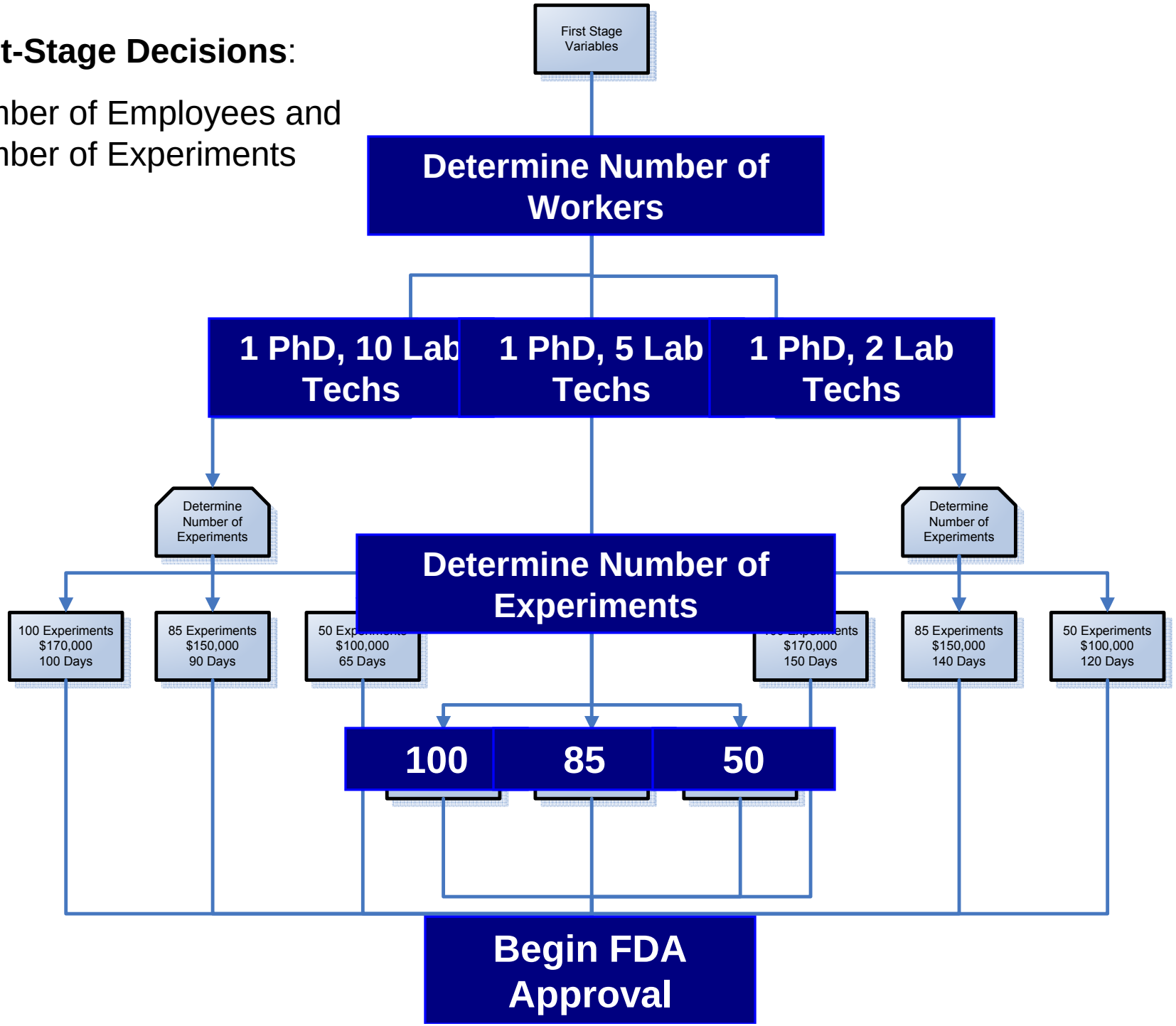
- First-Stage Decision Variables
  - "Here-and-Now" decisions that must be made prior to beginning a project

→ Number of Employees
  - PhDs and lab technicians that will manage and conduct experiments

→ Number of Experiments
  - Number of repeated experiments that will be performed to submit to the FDA to prove consistency of results

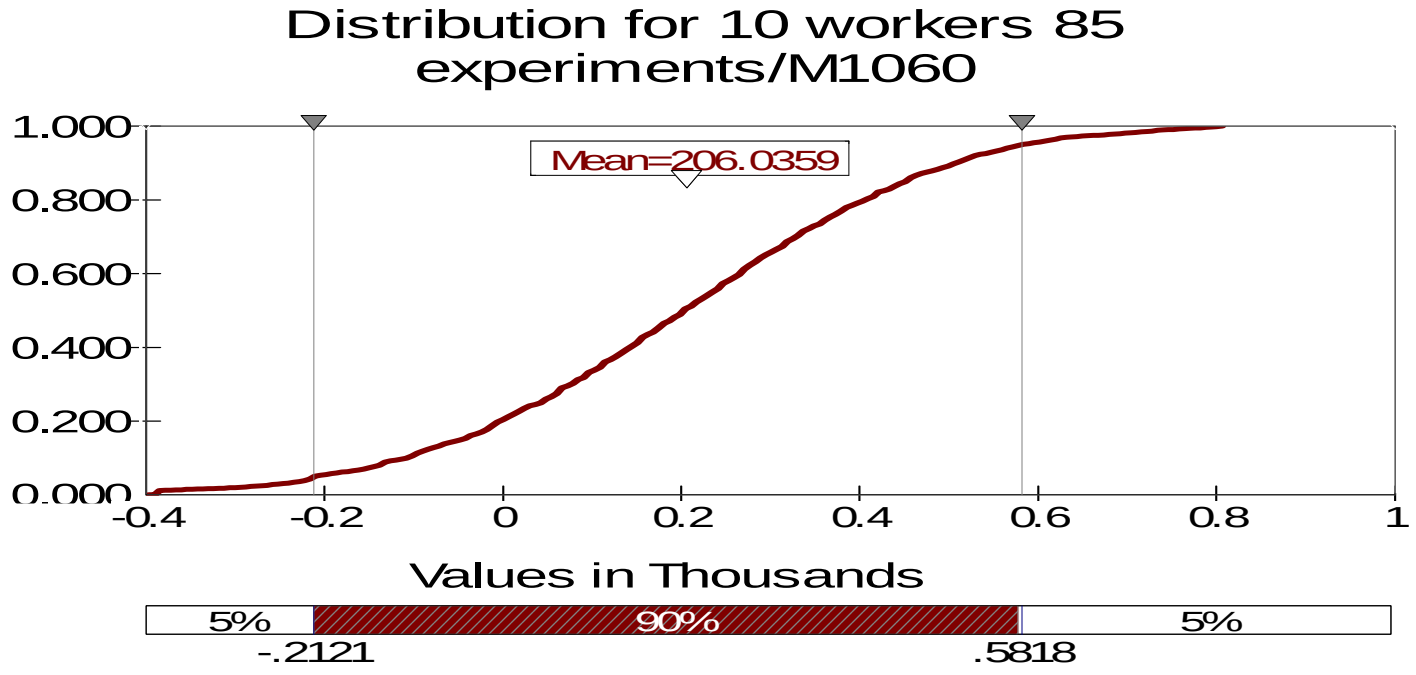
First-Stage Decisions:

Number of Employees and
Number of Experiments



FDA Risk

Name	Min NPV	Max NPV
10 workers 85 experiments	-396.31	805.83
5 workers 85 experiments	-554.35	815.63
2 workers 85 experiments	-415.27	901.93

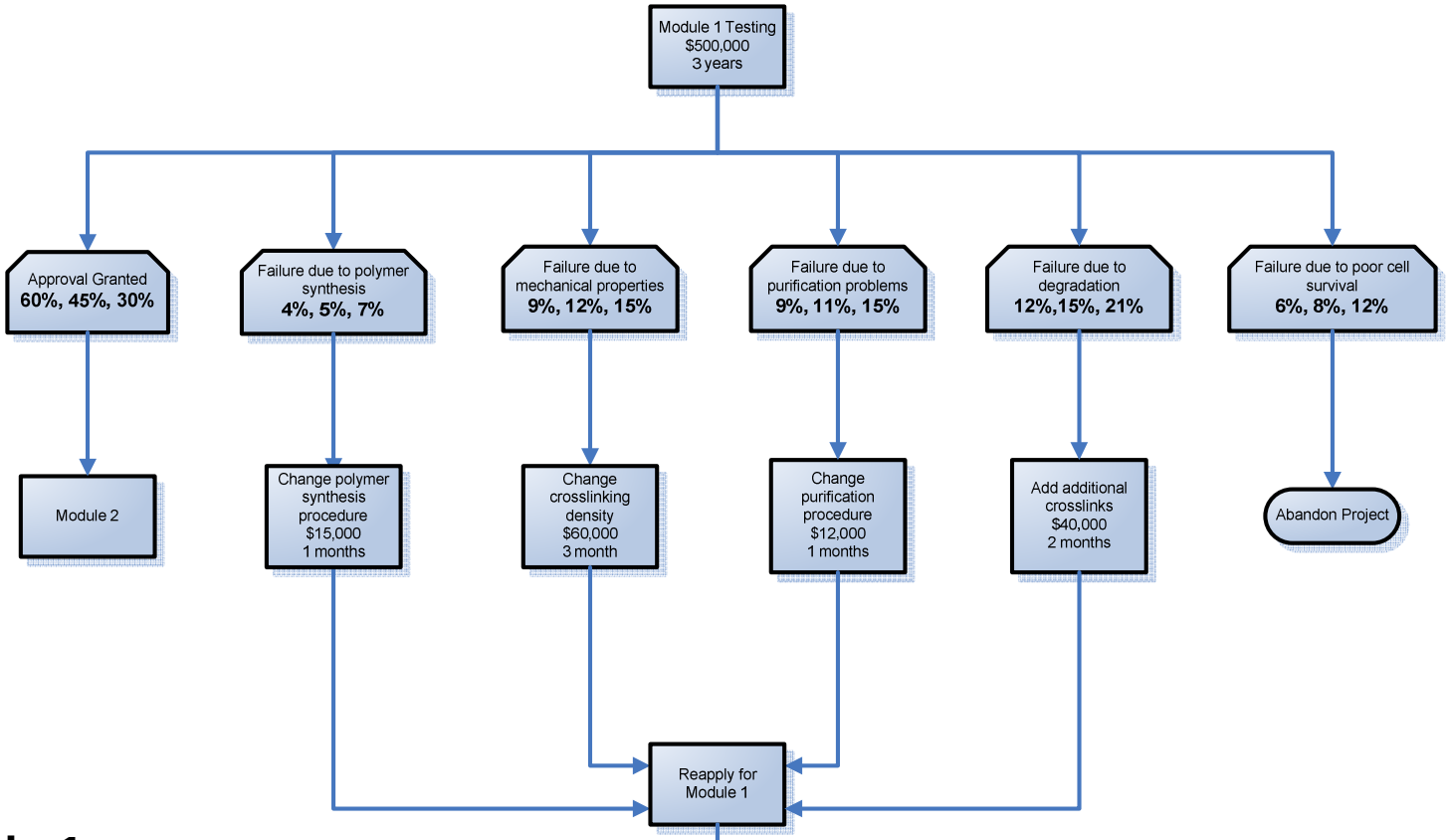


Modules

- We have assigned probabilities (0-100%) for each anticipated result after a particuar module is submission to the FDA
- There are three probabilities listed for each scenario, which correspond to the number of experiments performed
- In the case of disproval, we will re-evaluate our procedures and resubmit the module

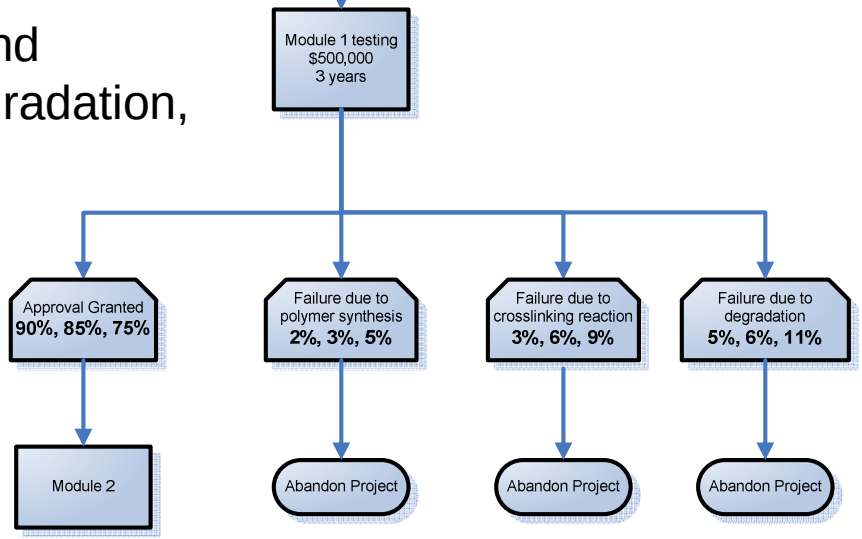
Module 1

- Non-Clinical Testing
  - Compression/tension ratings
  - Viscosity
  - Crosslinking density
  - Product purity
  - Degradation rates
  - Toxicity



Module 1:

Non-clinical chemical and physical properties, degradation, and toxicity tests



Module 2

- Non-Clinical Animal Testing
  - Biocompatability/Immunogenicity
  - Biodegradation
  - Infection
  - Injection procedure (large animals only)
  - Range of mobility (large animals only)

Module 3

- Clinical Trials (Human patient testing)
  - Range of motion
  - Reduction in pain
  - Lasting effects
  - Effectiveness over placebo

Conclusions

Conclusions

- The novel hydrogel HYAL-VYNE® will be hyaluronic acid modified with 2-vinyl and crosslinked with ammonium peroxydisulfate
- $(NH_4)_2S_2O_8$ increases
  - Stability
  - Retention
  - Load support

Conclusions

- Expected demand of 325,000 and will reach 625,000 per year
- Expected project life of 5 years
- Total product cost ~ $210 million
- Cost per injection ~ $688
- Expected FDA approval process cost will be ~ $2 million, and will take ~ 9 years

Further Studies – Scale-up

- With the determined demand, it would required that:
  - The plant capacity for HYAL-VYNE® be approximately 1000 Liters/yr
  - The cost of the treatment to be competitive should be ~ $1500 per year or $2400 per injection

Special Thanks

- Research contacts
  - CBME
    - Miguel J. Bagajewicz
    - Alberto Striolo
  - Chemistry/Biochemistry
    - Daniel T. Glatzhofer – Organic; Polymer Chemistry
    - Vadim A. Soloshonok – Synthetic Organic Chemistry

References

- "Report Measures Impact of Knee Conditions." Knee1.com. 11 November 2003. http://www.knee1.com/news/mainstory.cfm/220/1
- "Osteoarthritis Fact Sheet." Arthritis Foundation. 2005. http://www.arthritis.org/conditions/fact_sheets/OA_Fact_Sheet.asp
- "Dissectors Answers – Joints of the Upper and Lower Limbs." University of Michigan Medical School. 2000. http://images.google.com/imgres?imgurl=http://anatomy.med.umich.edu/images/knee_joint_post.jpg&imgrefurl=
- "Knee Joint – Anatomy and Function." The Center for Orthopaedics and Sports Medicine. Marietta, GA. 3 April 2003. http://www.arthroscopy.com/sp05001.htm
- "Chemical and Other Safety Information." The Physical and Theoretical Chemistry Laboratory, University of Oxford. 7 March 2006. http://ptcl.chem.ox.ac.uk/MSDS/#MSDS
- "Product Search – FisherScientific." 2006. <https://www1.fishersci.com/>
- Peters, Max S., Klaus D. Timmerhaus, Ronald E. West. *Plant Design and Economics for Chemical Engineers*, 5th ed. New York: McGraw Hill, 2003.
- "End of Small Volume High Value Myth in Biotechnology – Process Design for a Mega-plant Producing g-Interferon for Mega Profit."
- "2006 – 2008 General Catalogue." VWR. http://www.vwrsp.com/catalog/index.cgi?parent_id=9000002
- Mironowicz, Magdalena and Sarah Saw. "Electronic Texbook – Joints: Part 1." WorldOrtho, Inc. 1997. http://www.worldortho.com/database/etext/joints.html
- "Articular Cartilage Zones in Normal Articular Cartilage." Histology Atlas. School of Physical Therapy, Slippery Rock University. http://www.sru.edu/depts/pt/histo/artic_cartilage.htm
- Knox, P., J.R. Levick and J.N McDonald. *Quarterly Journal of Experimental Physiology*. 73.1, 33-45.

References

- Tadmor, Rafael; Nianhuan Chen, and Jacob N. Israelachvili. *Journal of Biomedical Materials Research*. 61.4, 514-523.
- Hinton, Ralph et al. "Osteoarthritis: Diagnosis and Therapeutic Considerations." American Family Physician. 1 March 2002.
- "Osteoarthritis Overview." eMedicine Consumer Health. < http://www.emedicinehealth.com/articles/5494-1.asp >
- "Osteoarthritis: medical information about the symptoms, diagnosis and treatment of degenerative joint disease." MedicineNet.com 16 April 2004.
- Hascall, Vincent and Torvard Laurent. "Hyaluronan: Structure and Properties." Glycoforum/Science of Hyaluronan-1.
- Dinnar, U. "Lubrication Theory in Synovial Joints." *CRC Critical Reviews in Bioengineering* **1975.**
- Griffith, Robert W., MD. "Knee Osteoarthritis: Is Arthroscopy Any Help?" Health and Age. 24 January 2003.
- Wen, Dennis Y., MD. "Hyaluronic Acid Injections for knee Osteoarthritis." American Family Physician. The American Academy of Family Physicians. 1 August 2000.
- "Federal Food, Drug, and Cosmetic Act – Chapter II." US Food and Drug Administration.
- "Total Cost to Develop New Prescription Drug, Including Cost of Post-Approval Research, is $897 million." Tufts Center for the Study of Drug Development. 13 May 2003.
- "Device Advice – Premarket Approval." US Food and Drug Administration Center for Devices and Radiological Health. 1 November 2002.
- Jones, D.B. and Middelberg, A. P.J. *Langmuir* **2002**, 18, 10357-10362.
- Kluger, R. and Alagic, A. *Bioorganic Chemistry* **2004**, 32, 451-472.

References

- Langer, Robert S.; Elisseeff, Jennifer, H.; Anseth, Kristi; Sims, Derek; "Semi- interpenetrating or interpenetrating polymer networks for drug delivery and tissue engineering" May 3, 1997.
- Binette, J.S.; Garon, M.; Savard, P.; McKee, M. D.; Buschmann, M. D.; *Journal of Biomechanical Engineering* **2004**, 126, 475-484.
- Huin-Amargier C.; Marchal, P.; Payan, E.; Netter, P.; Dellacherie, E.; *Journal of biomedical materials research* **2006**, 76A, 416-424.
- Uebelhart, Daniel MD.; Williams, James M. PhD.; *Current Opinion in Rheumatology* **1999**, 11(5), 427.
- Soltes, L. and Mendichi, R. *Biomedical Chromatography* **2003**, 17, 376-384.
- Burdick, J. A.; Khademhosseini, A.; Langer, R.; *Langmuir* **2004**, 20(13), 5153-5156.
- Ponnuraj, K. and Jedrzejas, M. J. *Journal of Molecular Biology* **2000**, 299, 885-895.
- Waddell, D. MD; Rein, A. MS; Panarites, C. PhD; Coleman, M. MD; Weiss, C. MD; *The American Journal of Managed Care* **2001**, 7(10), 981-991.
- Ochiai, H.; Ohmae, M.; Mori, T.; Kobayashi, S.; *Biomacromolecules* **2005**, 6, 1068-1084.
- Han, T. *Journal Zhejiang University SCI* **2004**, 5(8), 928-931.
- Chang, W.; Chang, Y.; Chen, Y.; Sung, H.; *Artificial Cells, Blood Substitutes, and Biotechnology* **2004**, 32(2), 243-262.
- Hennink, W.E.; Nostrum, C.F.; *Advanced Drug Delivery Reviews* **2002**, 54, 13-36.
- Lipowitz, A. J.; "Synovial Fluid", Ch. 86, **2006**, 1-19.
- Anggiansah, C. L.; Scott, D.; Poli, A.; Coleman, P. J.; Badrick, E.; Mason, R. M.; Levick, J. R.; *The Journal of Physiology* **2003**, 550.2, 631-640.
- Milas et al. *Biopolymers* **2000**, 59.4, 191-204.
- Kirker, Kelly R. and Glenn D. Prestwich. *Journal of Polymer Science: Part B: Polymer Physics* **2004.** 42.23, 4344-4356.

Questions?