Unlocking the Potential of Nature's Dendrimer: From Serendipitous Discovery to Fundamental Science to Commercialization

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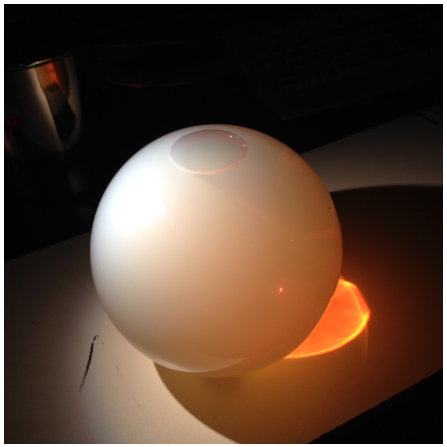
Dutcher Lab

Outline

- natural phytoglycogen nanoparticles
  - unique structure
    - highly branched particles – dendrimer
    - nano-confined water
  - soft & deformable
    - soft colloid & glassy physics
  - PhytoSpherix natural nanotechnology
    - Mirexus
- summary

Serendipitous Discovery

- fundamental, multidisciplinary collaboration involving physics & biology
- complicated, multi-step chemical process
- waste product from one step
  - interesting optical properties
    - opalescence
- kept waste instead of throwing it out
- eventually measured on atomic force microscope



Serendipitous Discovery

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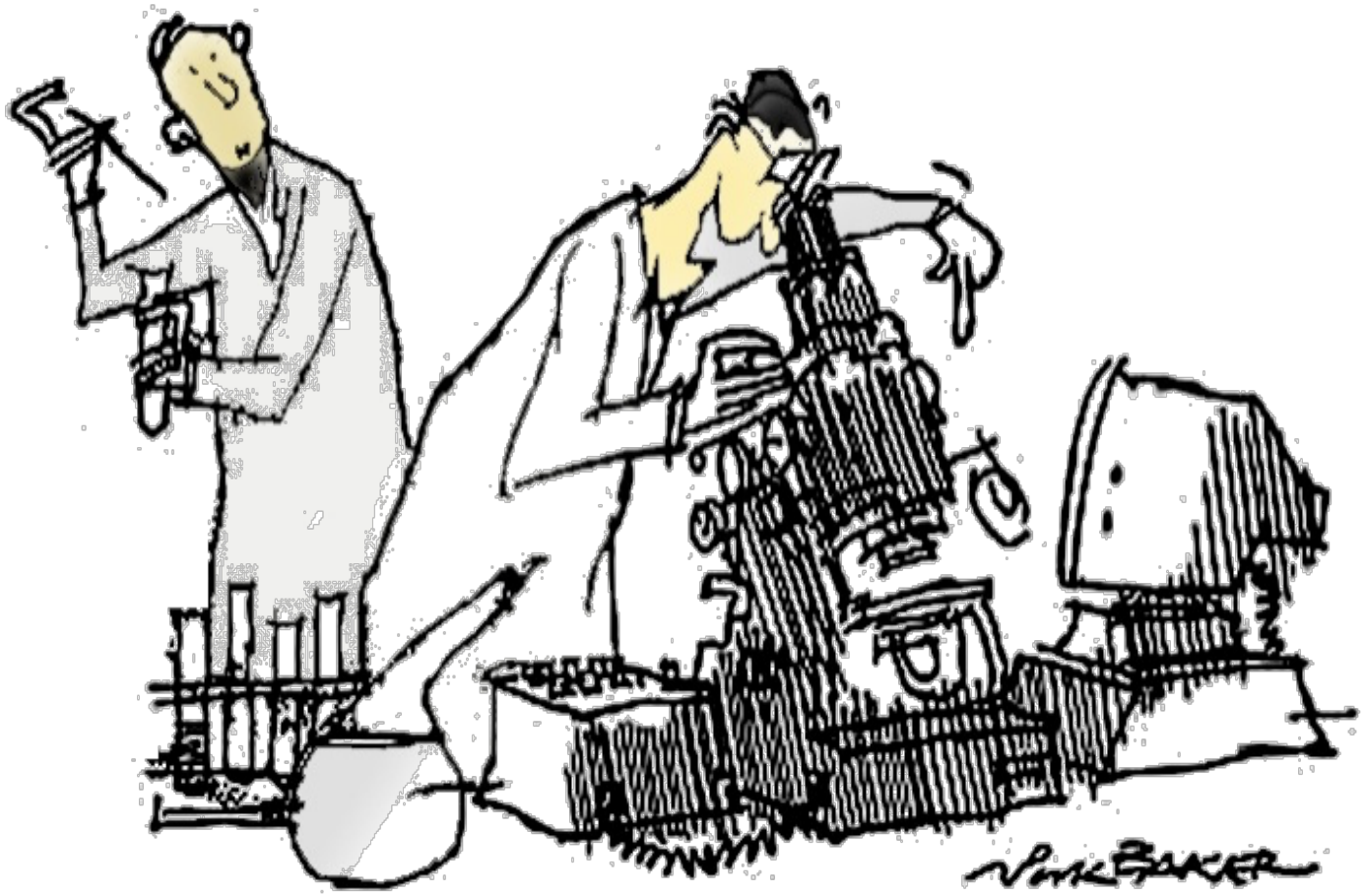

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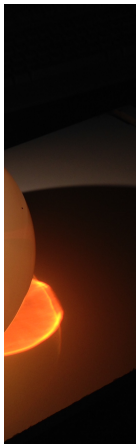
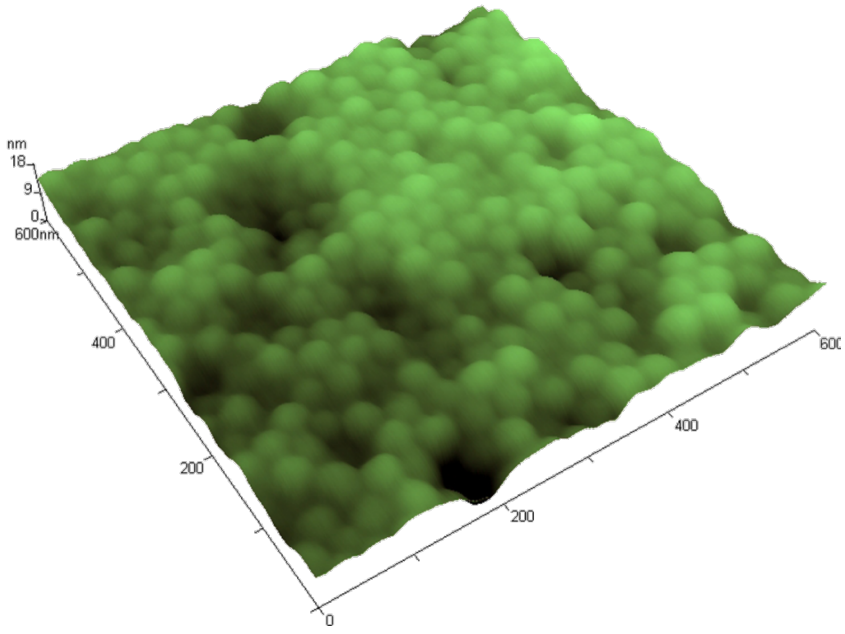


Phytoglycogen Nanoparticles

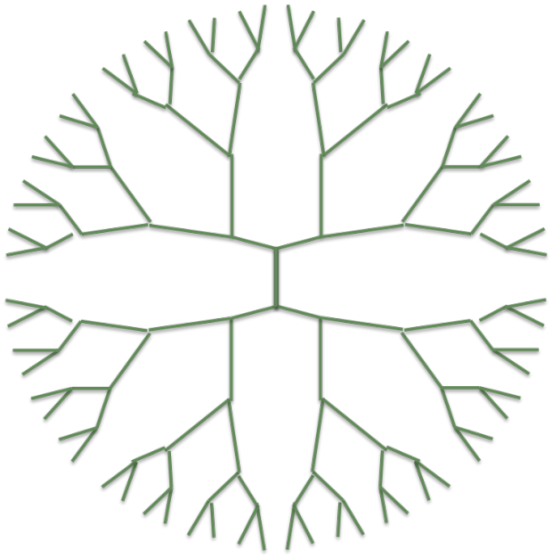


PhytoSpherix™

- uniform nano-size (35 nm dia)
  - highly-branched, soft, compact nanoparticles
- uniform surface chemistry
  - tunable surface properties
- biodegradable & edible
  - extracted & purified in their natural state from sweet corn
  - no harsh chemicals or enzymes

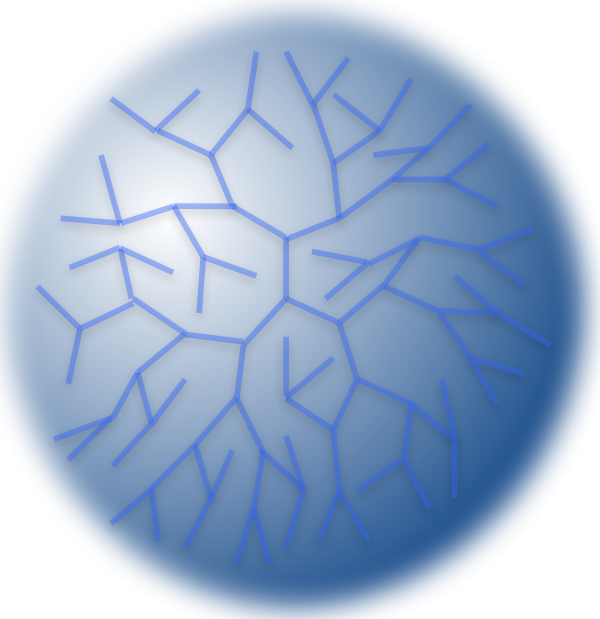
MIREXUS
NANO BY NATURE

Phytoglycogen Nanoparticles



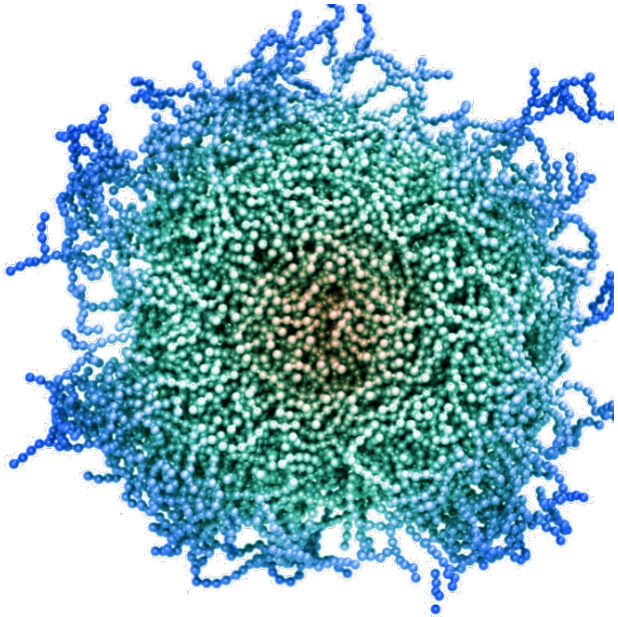
- Nature's dendrimer
  - glucose polymer with dendrimeric structure
- highly-branched spherical single molecule

Phytoglycogen Nanoparticles



- Nature's dendrimer
  - glucose polymer with dendrimeric structure
- highly-branched spherical single molecule
- real structure has
  - uniform density core
  - highly-branched throughout

Phytoglycogen Nanoparticles



[de Haan (2017)]

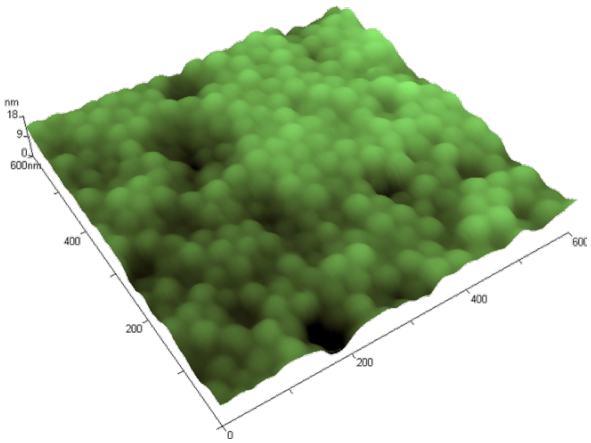
- Nature's dendrimer
  - glucose polymer with dendrimeric structure
- highly-branched spherical single molecule
- real structure has
  - uniform density core
  - highly-branched throughout

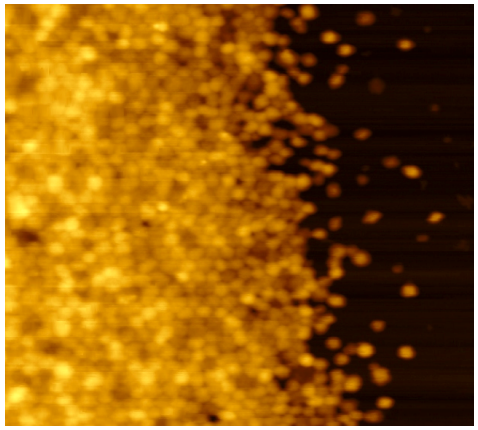
Unique Properties of Phytoglycogen

- special structure results in special properties
  - monodispersity
  - extraordinary water retention
  - low viscosity in water
  - high stability in water

special interaction with water

  - ability to stabilize & disperse bioactive compounds
  - film forming capacity





Techniques To Study Water

- neutron scattering
  - scattering of neutrons from particles dispersed in water
    - uniform particle size & density, molar mass
    - high water content & slow water dynamics

[Nickels *et al*., Biomacromolecules (2016)]

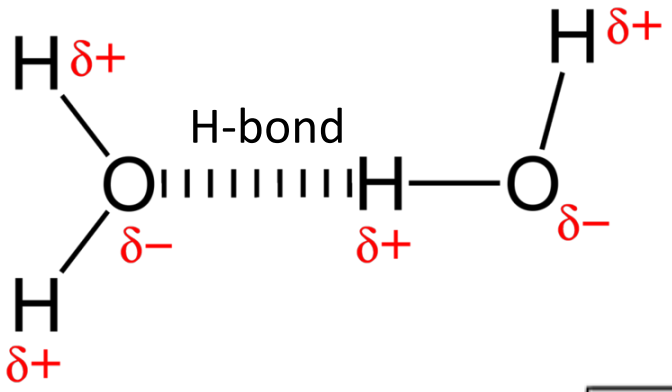
- infrared spectroscopy
  - absorption of infrared by O-H bonds
    - highly structured water inside particles

[Grossutti & Dutcher, Biomacromolecules (2016)]

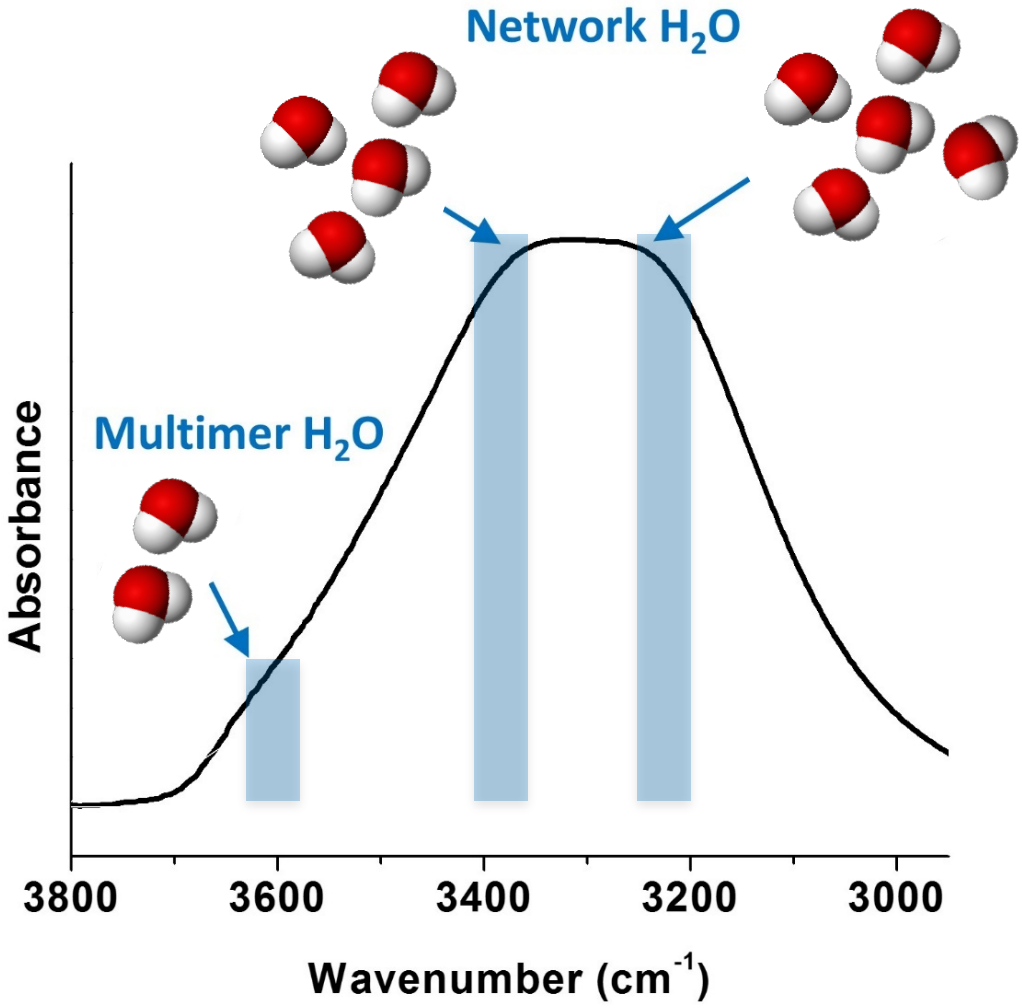
- ellipsometry
  - swelling of ultrathin films of particles
    - short-range hydration forces [Grossutti *et al*., Langmuir (2017)]
- rheology
  - viscosity of aqueous dispersions of particles
    - deformation & colloidal glass [Shamana *et al*. (2017)]

IR Spectroscopy & Water Structure

- excellent technique to study water structure
  - O-H stretching mode of bulk water

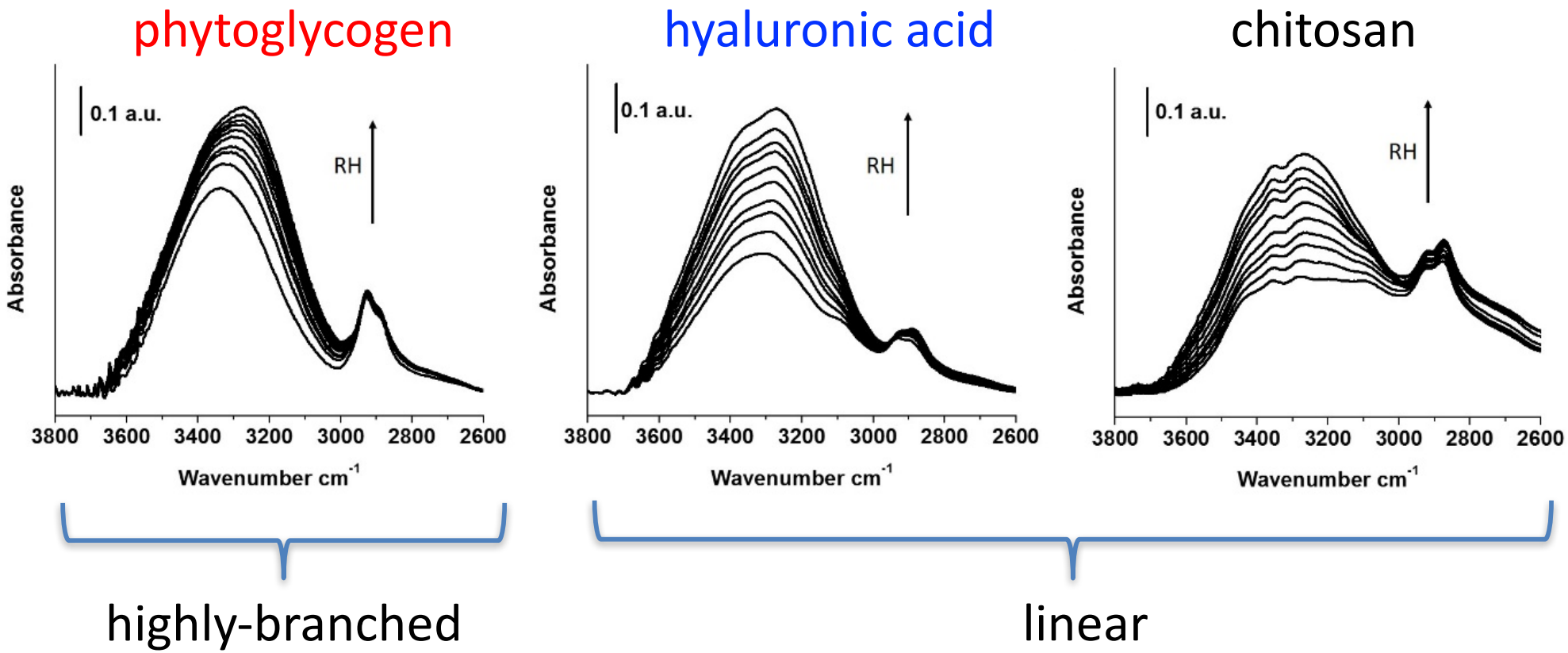


$$\upsilon_{radiation} = \upsilon_{vibration} = \frac{1}{2\pi}\sqrt{\frac{k_{vibration}}{m_{vibration}}}$$



Absorbance Spectra

- compare highly-branched phytoglycogen & two linear polysaccharides

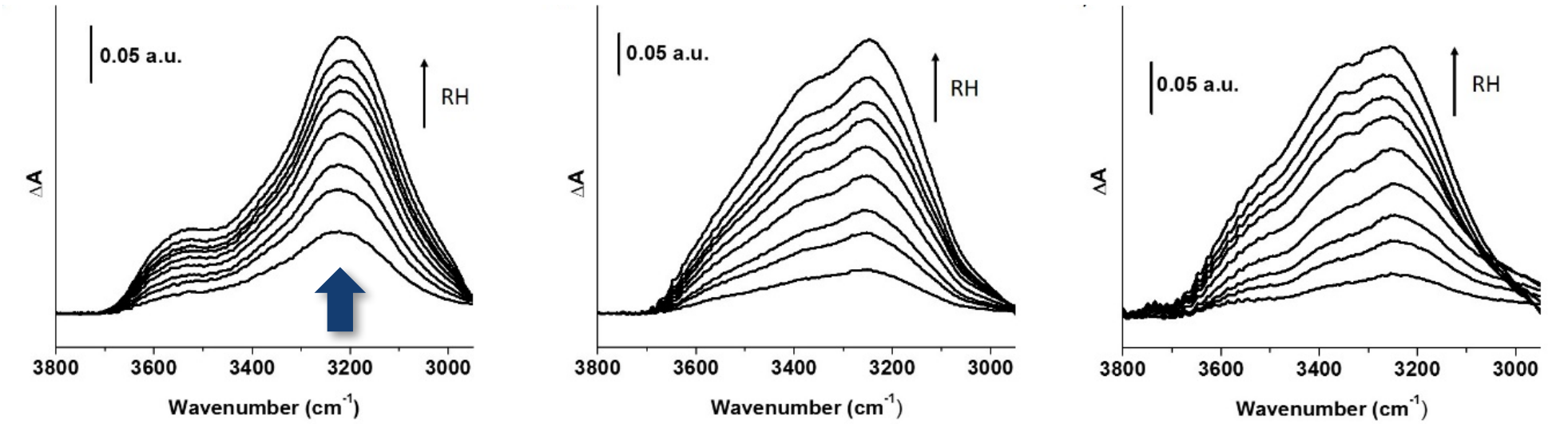


[Grossutti & Dutcher, Biomacromolecules (2016)]

Difference Spectra

- difference between spectra at different RH & reference spectrum at lowest RH

phytoglycogen | hyaluronic acid | chitosan



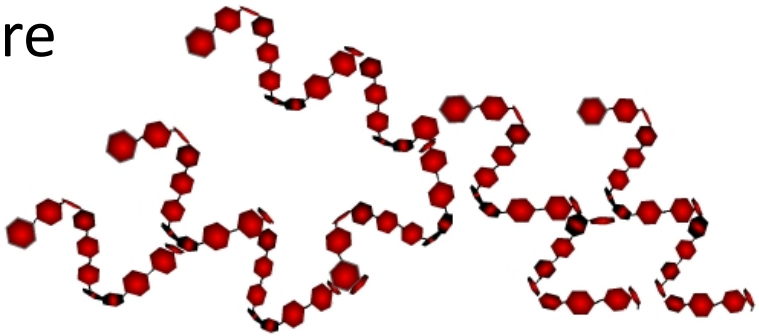
highly-branched | linear

- phytoglycogen has more highly connected, bound water

[Grossutti & Dutcher, Biomacromolecules (2016)]

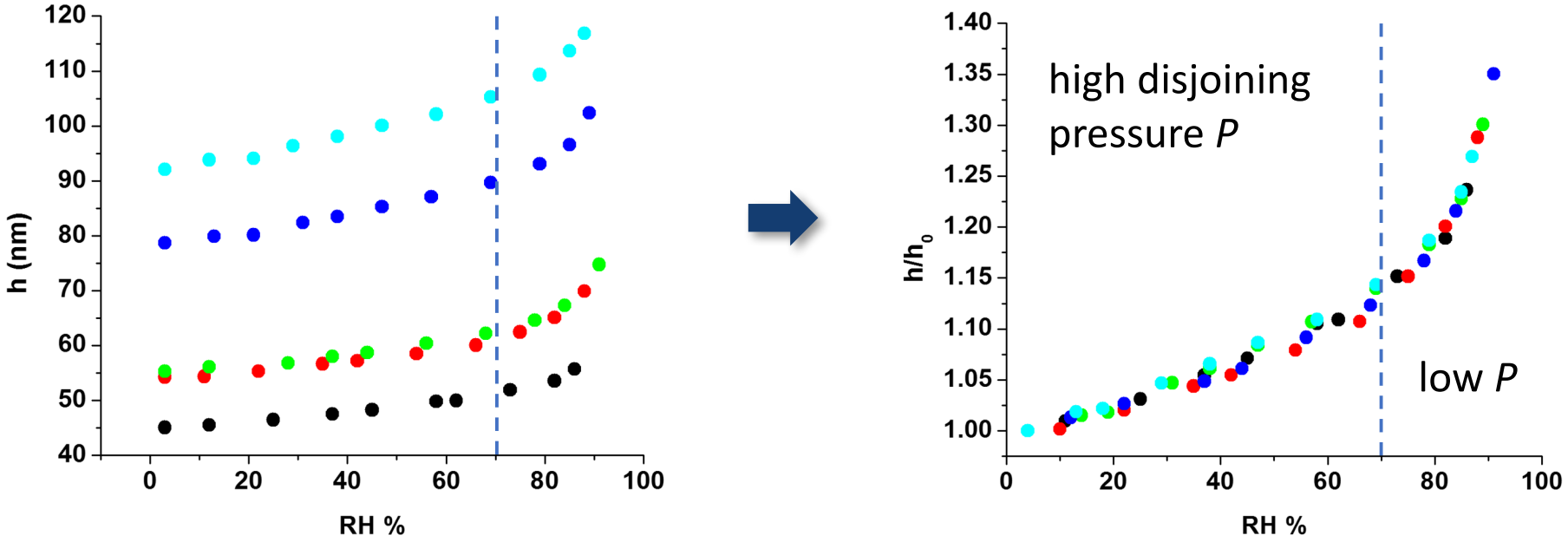
Nano-confined Water

- what is effect of nano-confinement on water structure?
- previous studies have shown
  - sensitivity to confinement
  - insensitivity to chemical environment
- generally introduces hydrogen bond defects & reduces water connectivity (polymers, micelles, liposomes)
- in contrast, connectivity increased for water confined in cylindrical glass nanopores
  - perhaps water in phytoglycogen more highly ordered because of water nanochannels along helical chains

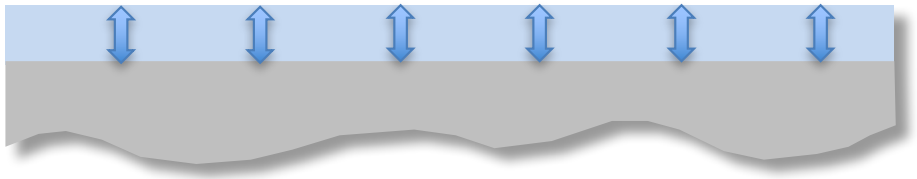


Ultrathin Film Swelling

- ellipsometry measurements of ultrathin phytoglycogen films



- interpret results in terms of disjoining pressure *P* acting across film

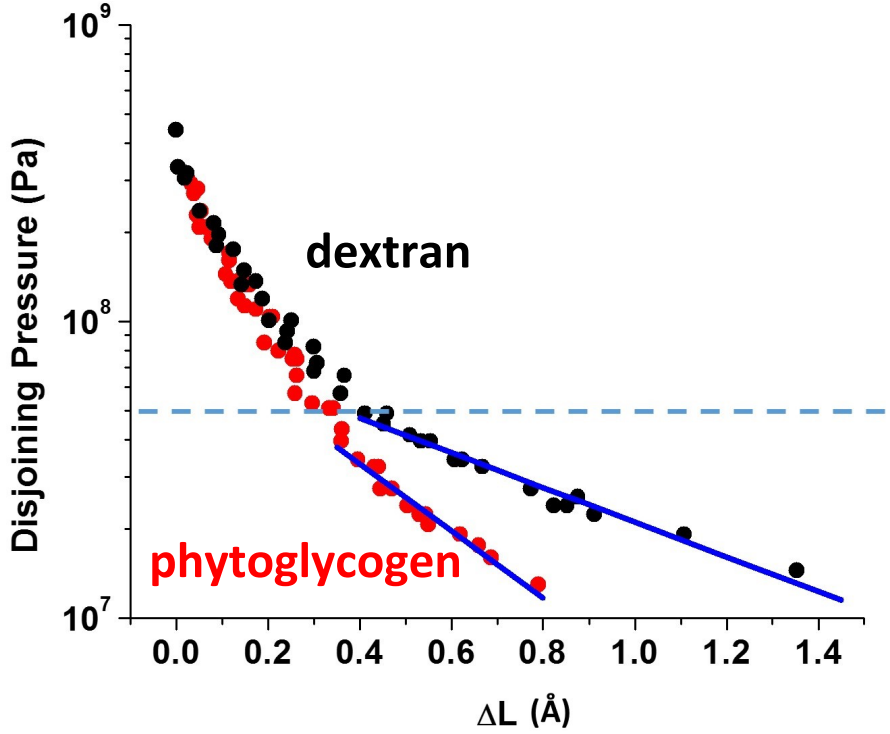


[Grossutti *et al*., Langmuir (2017)]

Hydration Forces

- can relate swelling results at high RH to hydration forces

$$P(\Delta L) = P_0 e^{-\Delta L/\lambda_0} \qquad \Delta L = a_0 \left[\left({}^{h}/_{h_0} \right)^{1/3} - 1 \right]$$



compare glucose-based polysaccharides

polysaccharide	P_0 (10^7 Pa)	λ_0 (Å)
dextran	8.1 ± 0.5	1.71 ± 0.10
phytoglycogen	9.3 ± 0.8	0.89 ± 0.06
cellulose*	14	0.32

- decay length λ_0 depends on chain architecture

[Grossutti *et al*., Langmuir (2017)]

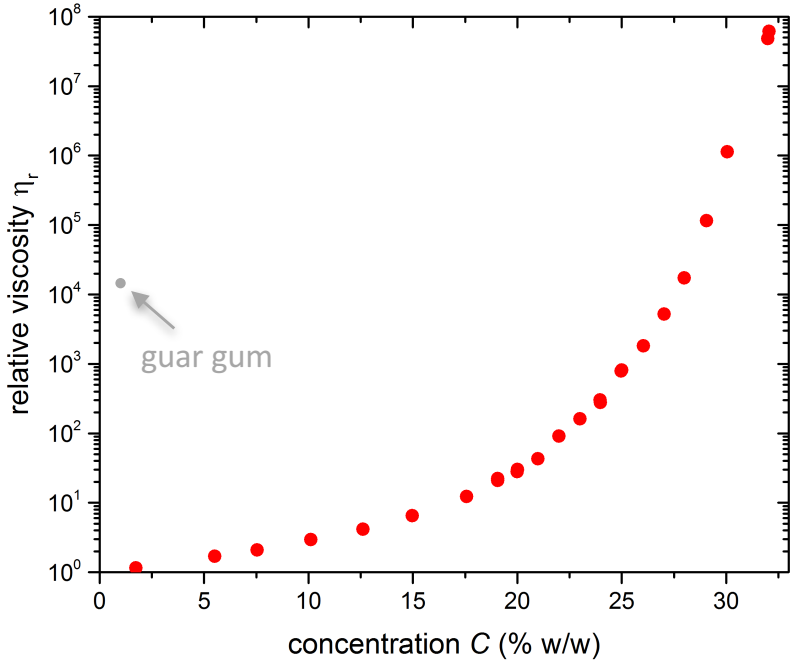
*[Rehfeldt *et al*., Langmuir (2003)]

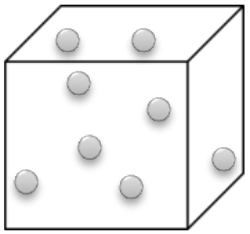
Phytoglycogen Particles In Water

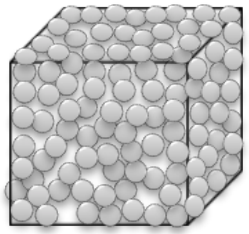
- can be loaded up to 20% by mass before significant increase in viscosity

➡ “hard spheres”
(small interaction except when they touch)

“solid-like”







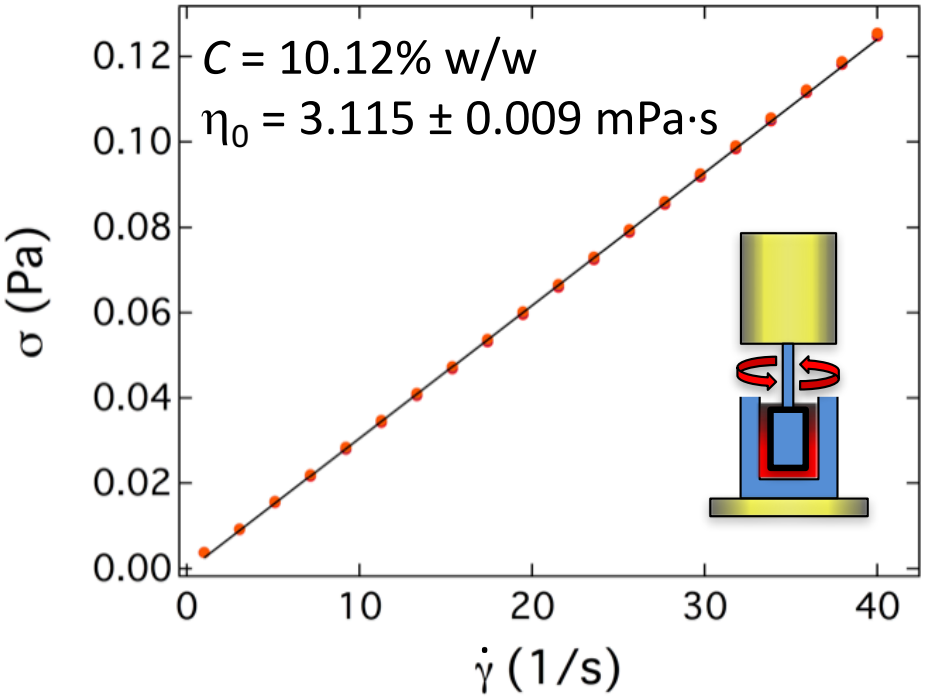
[Shamana *et al*. (2017)]

Viscosity Measurements

- mix spray dried particles at room temp into Milli-Q H_2O

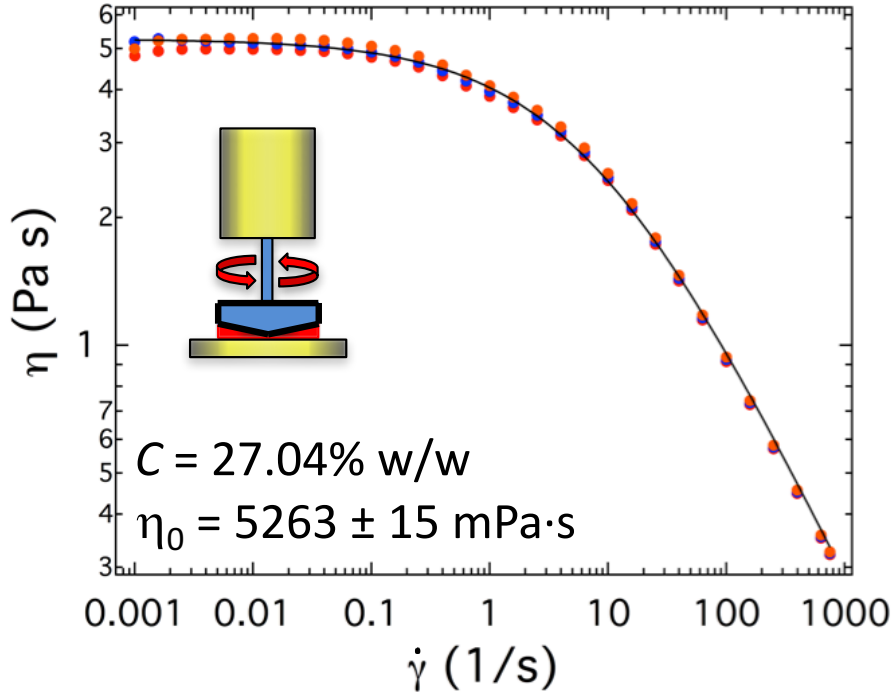
$C \leq 20\%$ w/w: concentric cylinders

$$\sigma = \eta\dot{\gamma} = \eta_0\dot{\gamma}$$



$C \geq 20\%$ w/w: cone & plate

$$\eta(\dot{\gamma}) = \frac{\eta_0}{1 + (k\dot{\gamma})^m}$$



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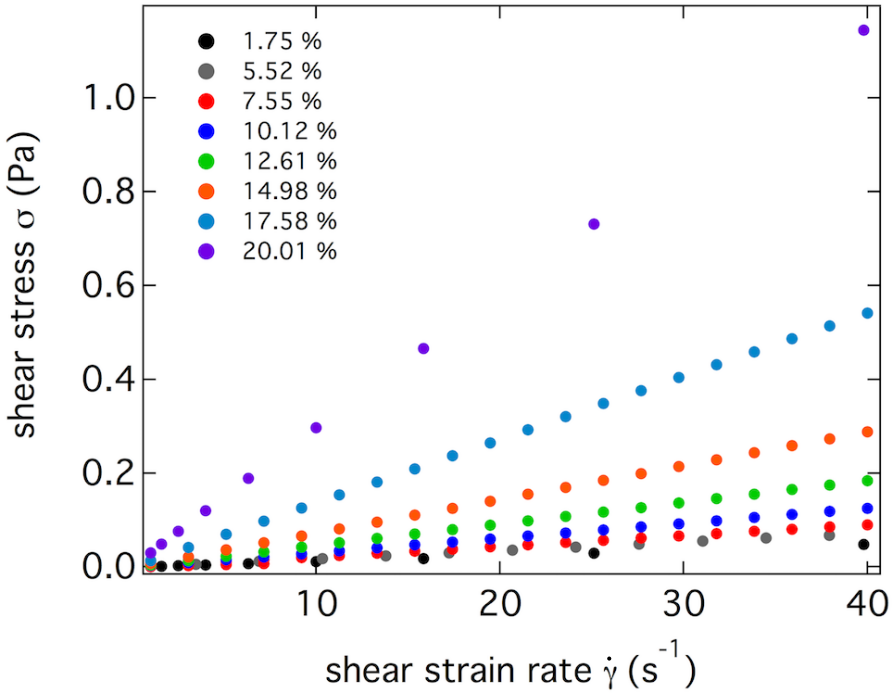
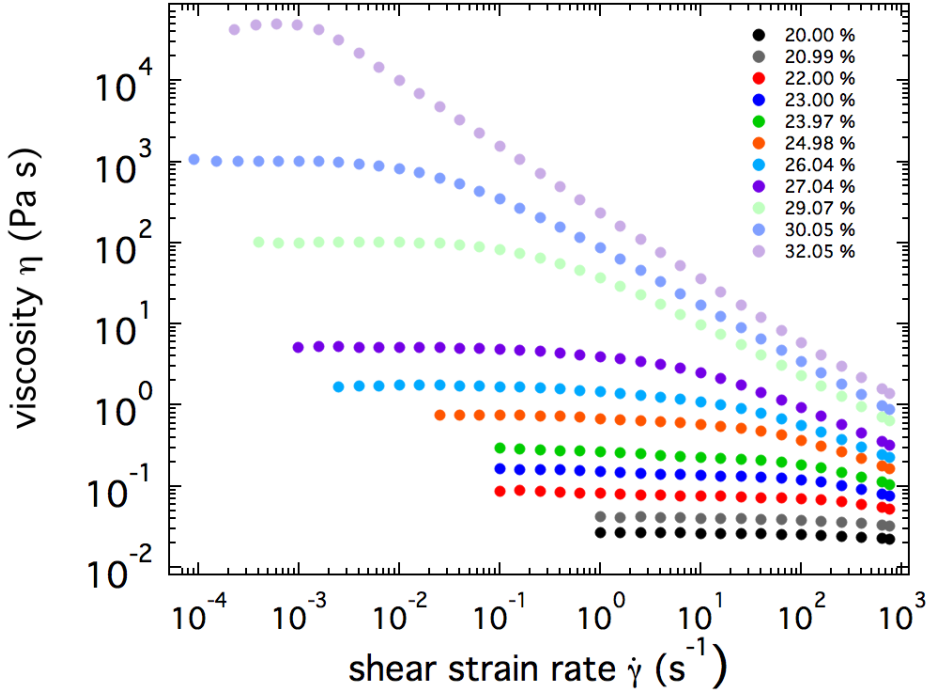
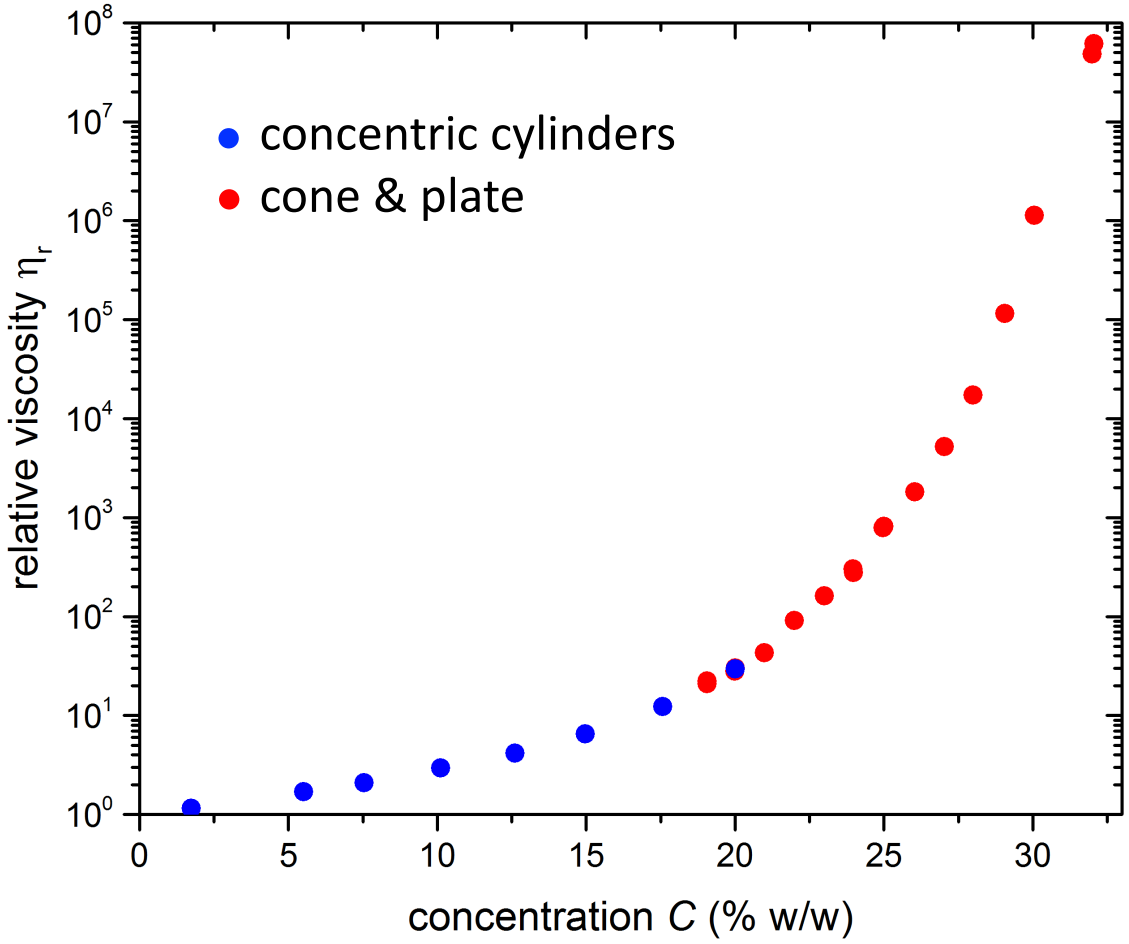




[Shamana *et al*. (2017)]

Zero Shear Viscosity



[Shamana *et al*. (2017)]

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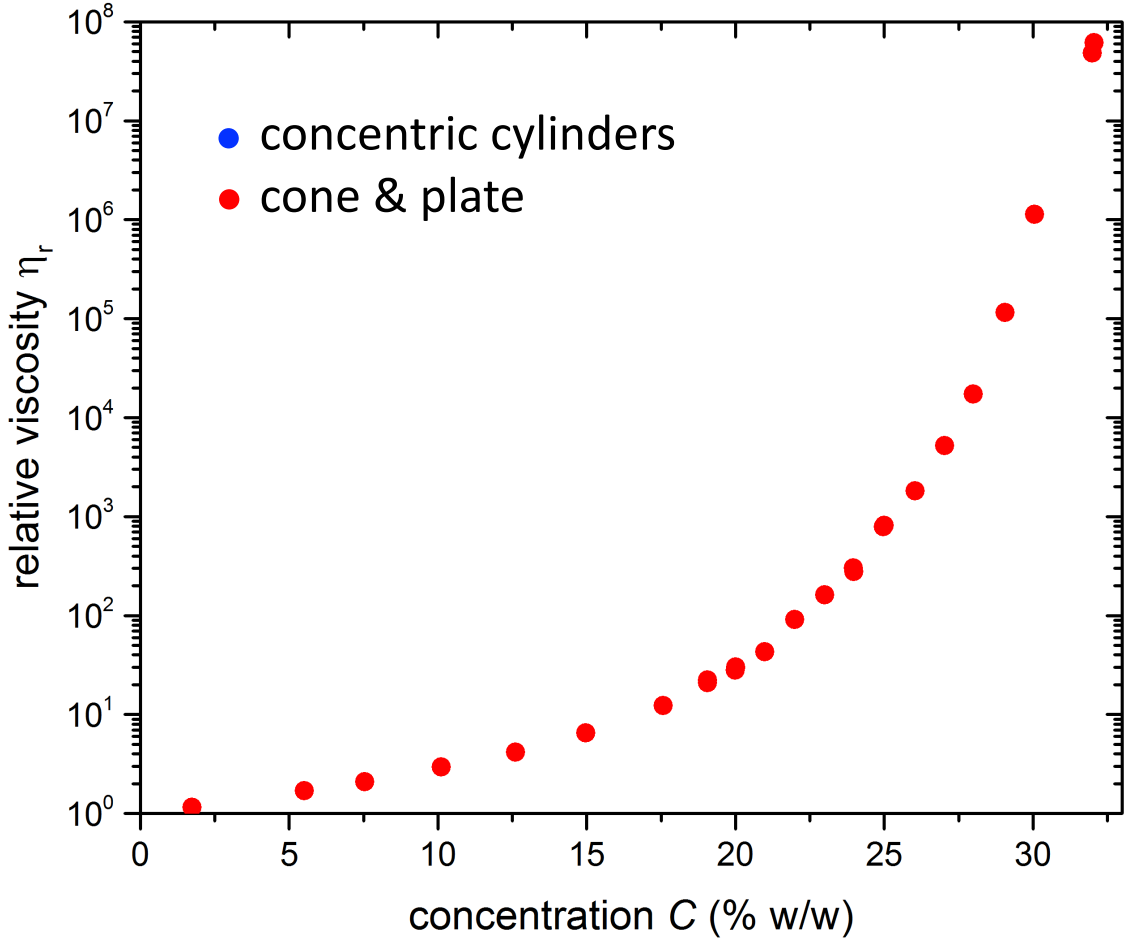


[Shamana *et al*. (2017)]

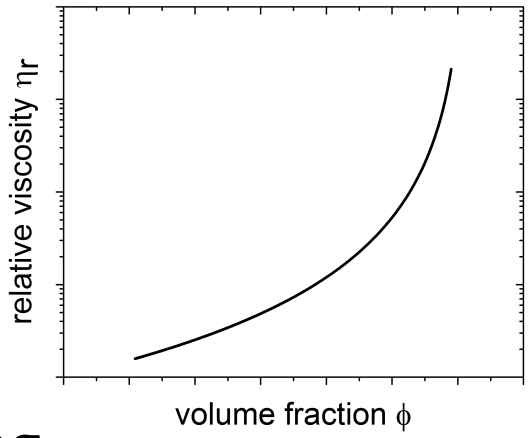
Hard Sphere Dispersion Theories

- properties of dispersion defined by volume fraction ϕ
- at low concentrations, viscosity described by Einstein relation

$$\eta_r = 1 + 2.5\,\phi$$

- at high concentrations, semi-empirical approach
  [Krieger & Dougherty, Trans. Soc. Rheol. (1959)]

$$\eta_r = \left(1 - \frac{\phi}{\phi_m}\right)^{-[\eta]\phi_m}$$



  - describes divergence of viscosity due to packing of hard spheres at high volume fractions ϕ
- viscosity diverges at colloidal glass transition at ϕ_g = 0.58
  [Mason & Weitz, PRL (1995)]

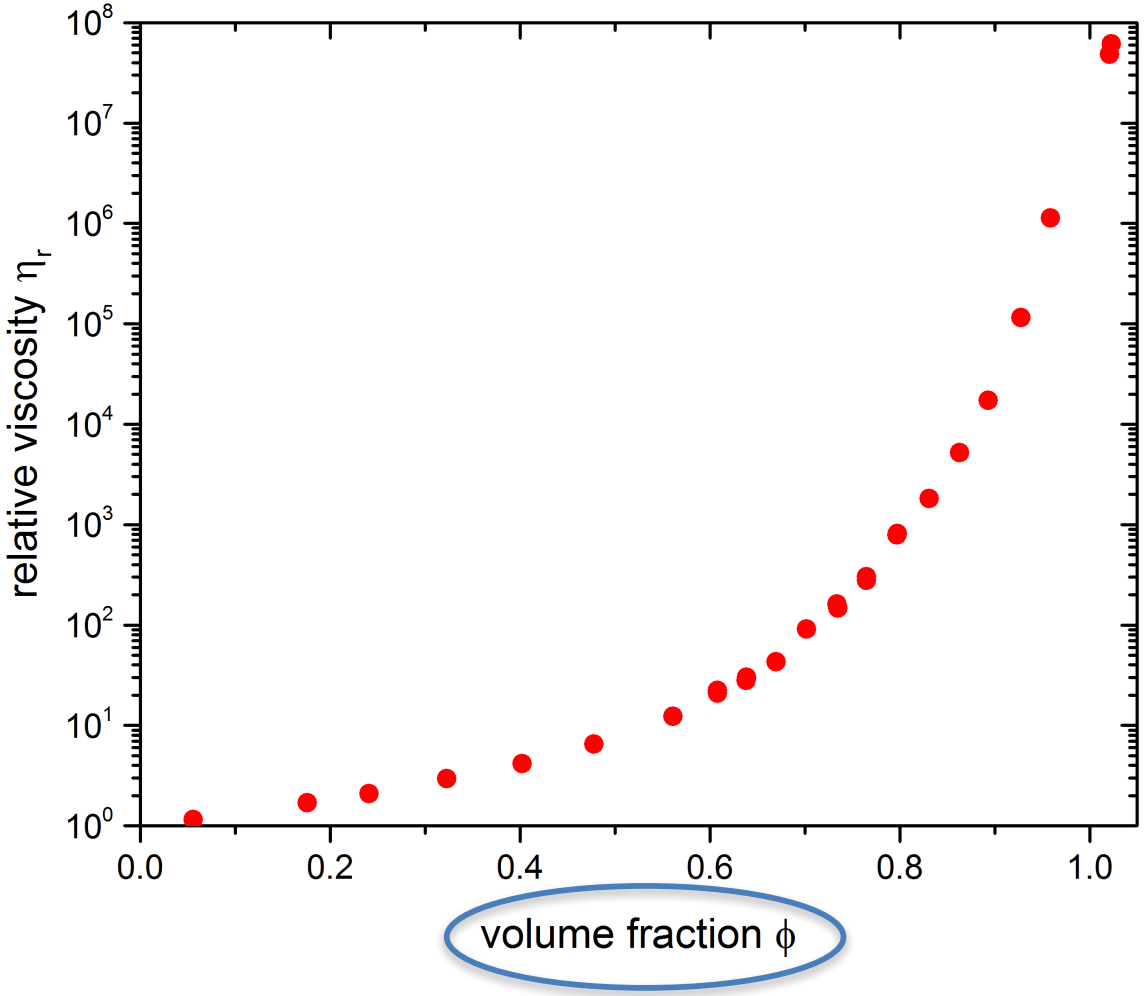
Volume Fraction Values

- need to convert concentrations C into volume fractions ϕ
  - spray dried particles sorb considerable amount of water
- depends on quantities that we can measure
  - mass of spray dried particles
  - volume of added water
  - residual water in spray dried particles
  - density of hydrated particles

} measured independently

[Shamana *et al*. (2017)]

Viscosity Versus Volume Fraction



[Shamana *et al*. (2017)]

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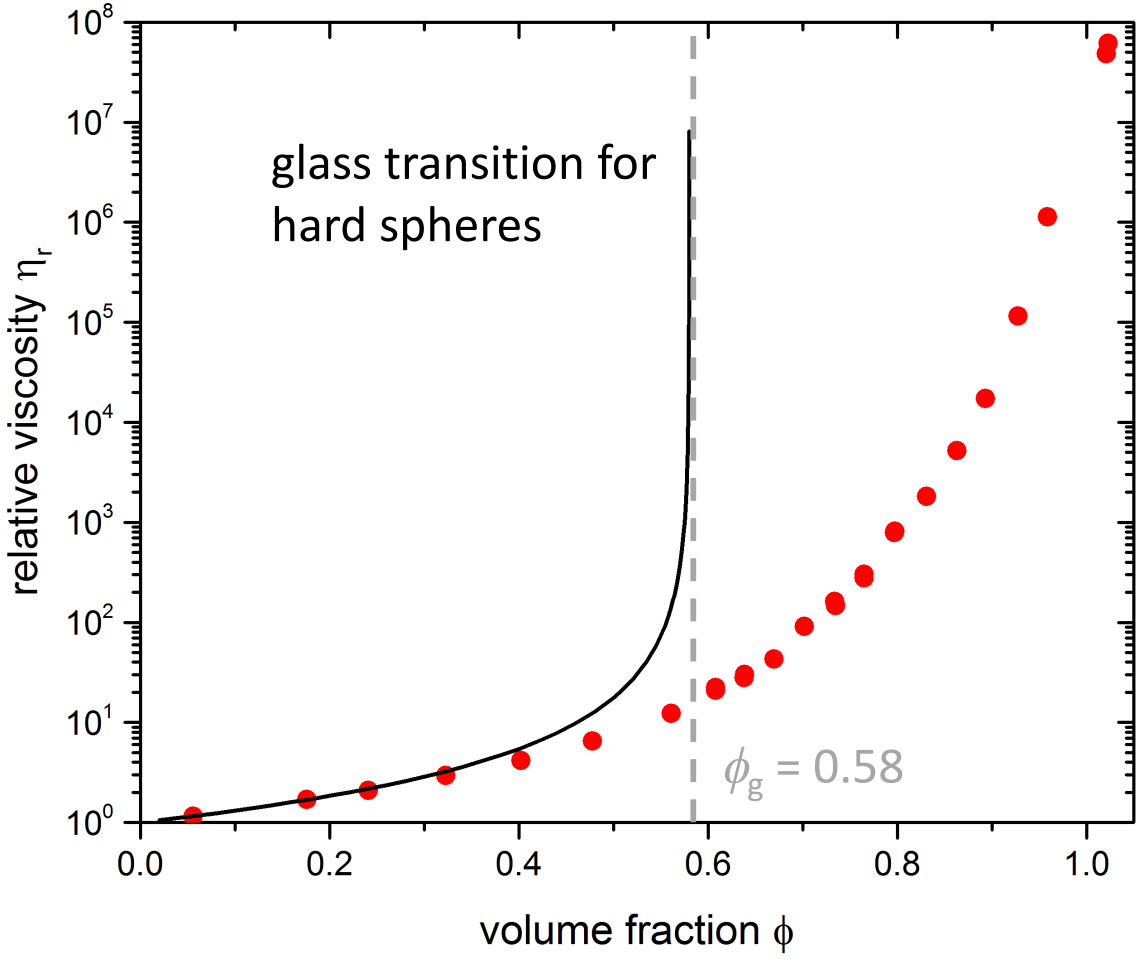
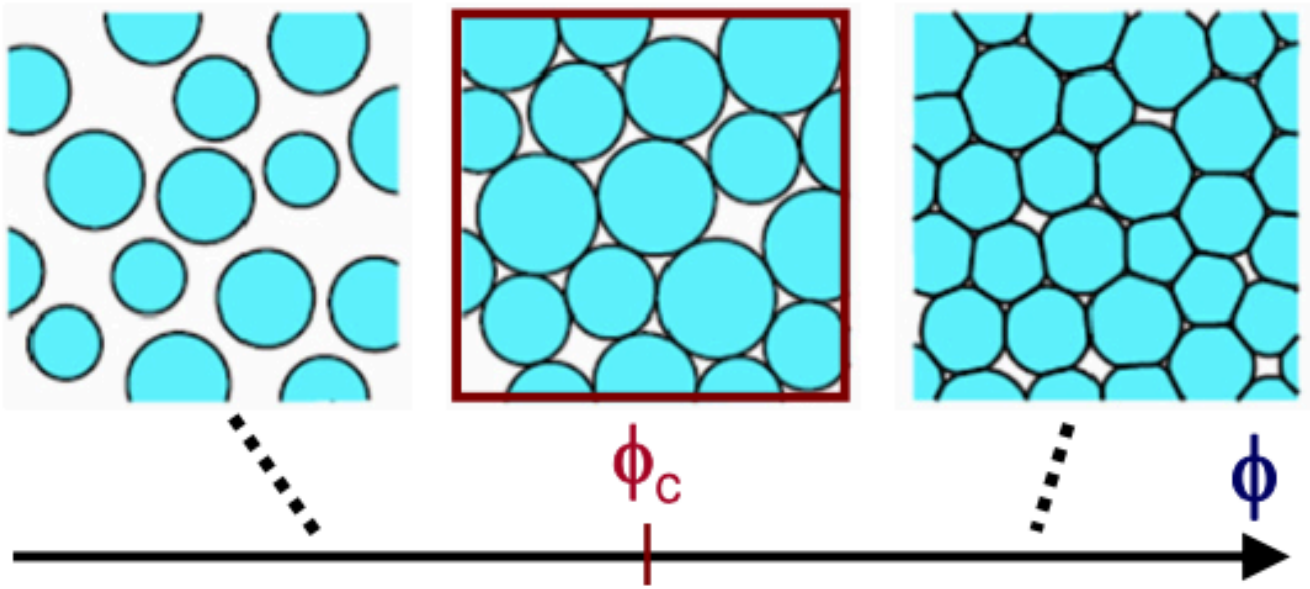


- considerable deformation of soft particles at high ϕ
  - consistent with neutron scattering & AFM [Nickels *et al*., Biomacro (2016)]

[Shamana *et al*. (2017)]

Deformation at Large ϕ

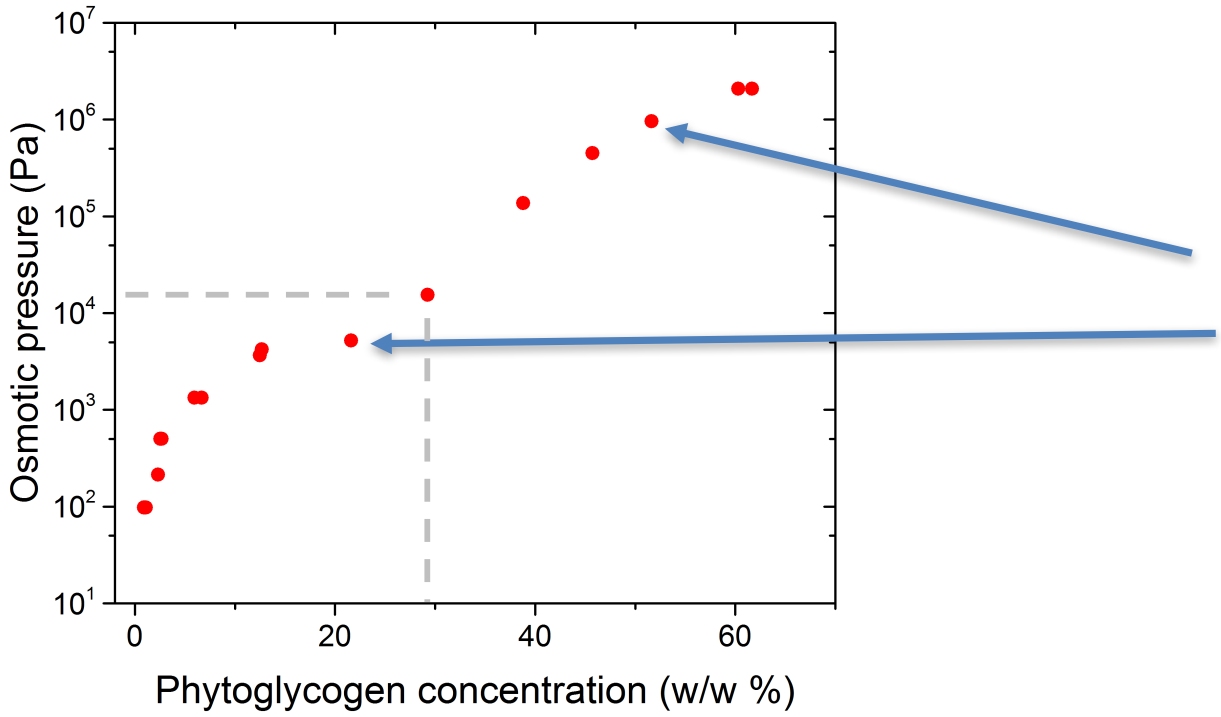
- soft particles will readily deform at large ϕ
  - facets will develop between particles to fill in voids up to $\phi = 1$ ($C \sim 30$ % w/w)



[M. Van Hecke, J. Phys.: Condens. Matter (2010)]

Osmotic Pressure Study

- apply osmotic pressure Π by doing dialysis with PEG & dextran solutions & measure equilibrium concentration
  - achieve concentrations > 60 % w/w

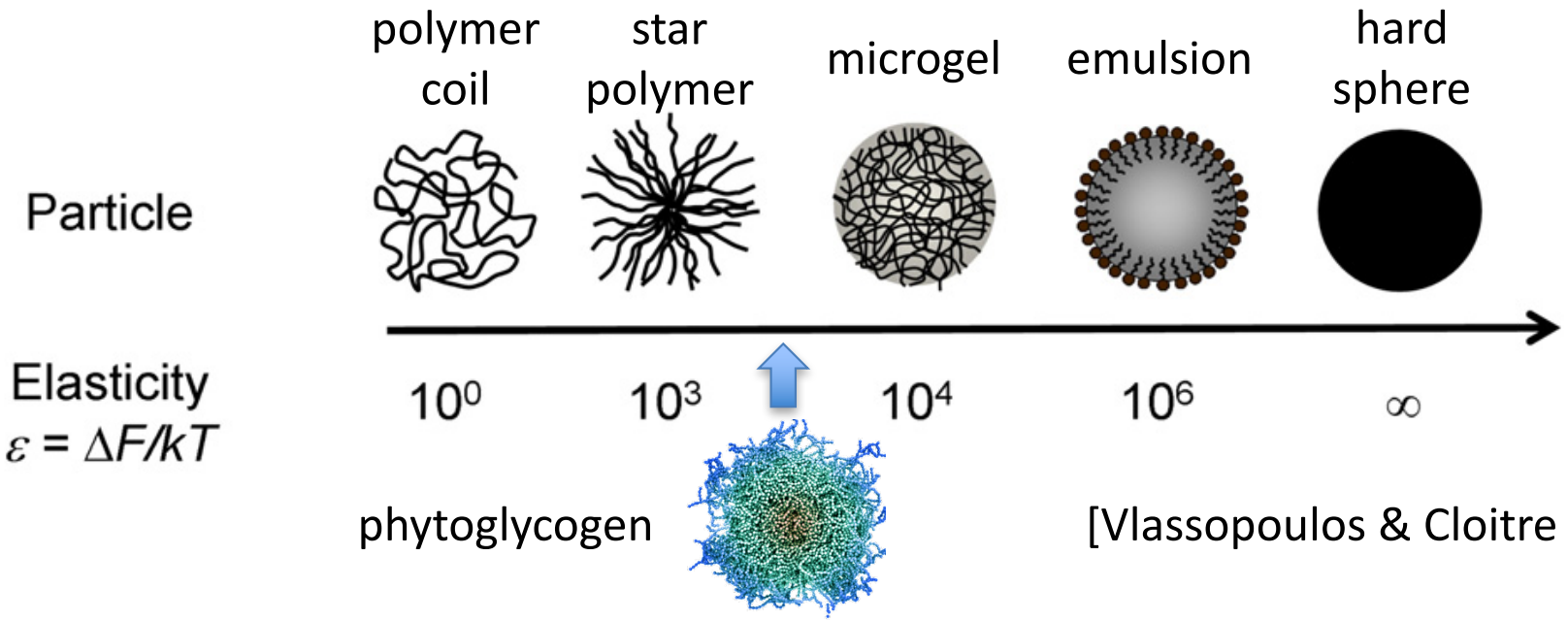


Π = 5 x 10^5 Pa

- for concentration C = 30 % w/w, $\phi \sim 1$
  - value of Π provides estimate of bulk modulus of particles: K ~ 20 kPa

Soft Colloid Systems

- soft colloid systems offer new insight into particle interactions & deformations



[Vlassopoulos & Cloitre (2014)]

- phytoglycogen dispersions are a new model soft colloid system
  - native particles
  - chemically & enzymatically modified particles

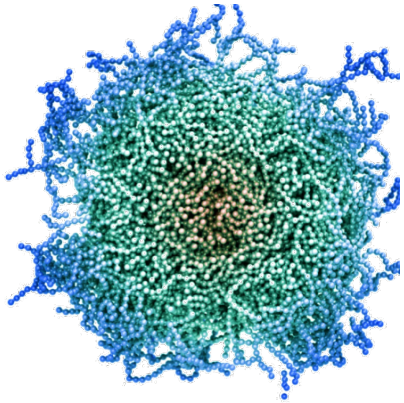
PhytoSpherix™ Technology

- research in my U of Guelph laboratory
  - particles from non-GMO corn
  - successful crop trials
  - pilot scale production of tens of kg

- MIREXUS
  SAFE & NATURAL NANOMATERIALS
  - IP protected by patent portfolio
  - angel investment, $7 M investment in 2015, $12 M in 2017
  - R&D facility in Guelph has 18 FT employees (9 PhD)
  - new 12,000 ft^2 facility under construction, 16 T/y production
  - approvals for use in cosmetics & food
  - products developed, orders shipped, products available

Applications of PhytoSpherix Particles

- many advantages to using PhytoSpherix in personal care, nutritional & biomedical applications
  - water soluble
  - monodisperse
  - non-ionic
  - low viscosity
  - can be sterilized



  - renewable resource
  - biocompatible
  - non-toxic
  - non-allergenic
  - biodegradable
  - easy to modify with high density of functional groups

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MIREXUS

Fortis Ingredients

glysantis

Personal Care
enhanced natural cosmetics & anti-aging products

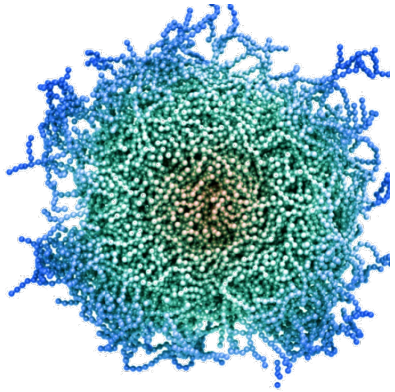
Nutrition
functional additives & enhanced nutrition

Health Care
anti-infectives, drug delivery & immunomodulation

Phytoglycogen Nanoparticles

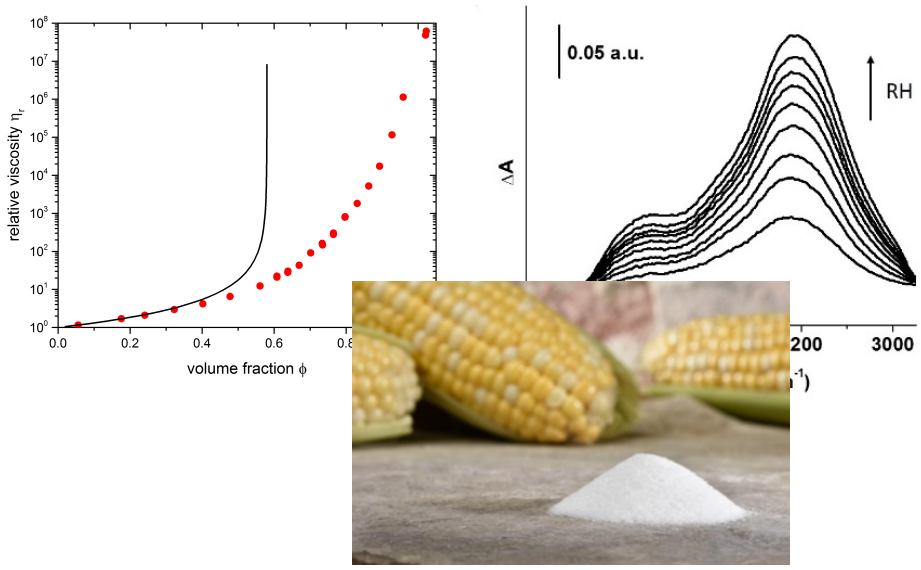
- nano from nature
  - safe, non-toxic – even edible!
- special structure ➡ special properties
  - unique strong interaction with water
  - model soft colloid system

    ➡ promising new material for fundamental research & new technologies



Dutcher Lab

www.physics.uoguelph.ca/dutcherlab



The Team

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- JRD
- Michael Grossutti
- John Atkinson
- Hurmiz Shamana
- Erzsi Papp-Szabo
- Ben Baylis
- Eric Bergmann
- Perry Mahon
- Aidan Maxwell

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