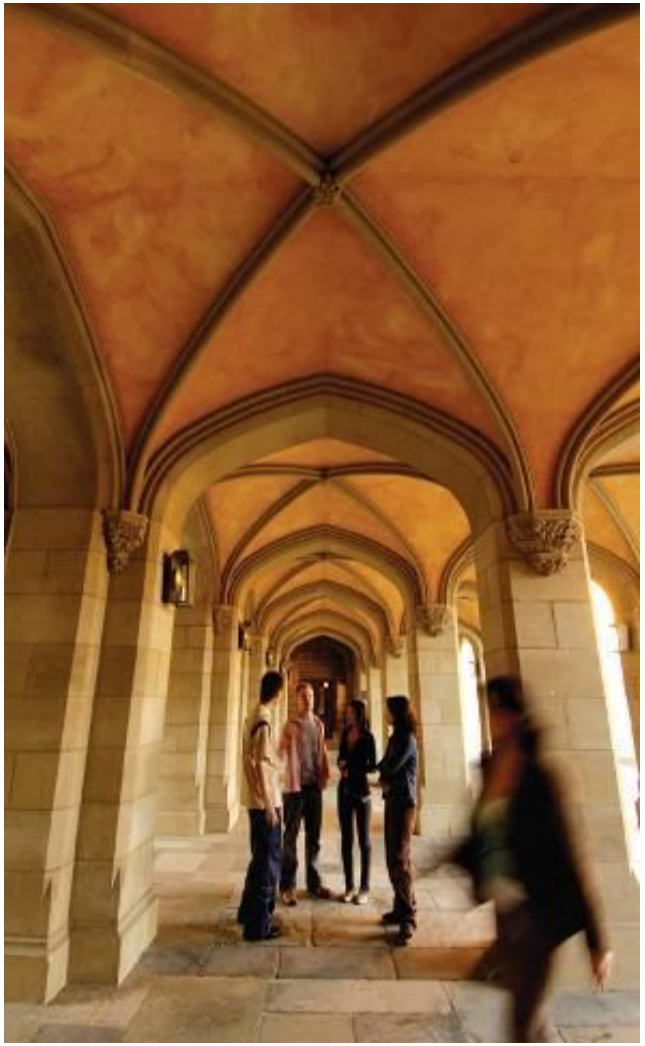
Ultrasonic Technology for Food/Bioprocessing

Muthupandian Ashokkumar

SUSU, October 12, 2016

History of Leadership

- Established in 1853
  - Second oldest university in Australia
  - 160th Anniversary in 2013
- History of Leadership
  - Graduated Australia's first female student in 1883
  - Offered Australia's first PhD program in 1945
  - Home to first computer built in Australia in 1955
  - Launched major curriculum reform in 2008
  - First Australian member of Massive Open Online Courses (MOOCs) provider Coursera 2013



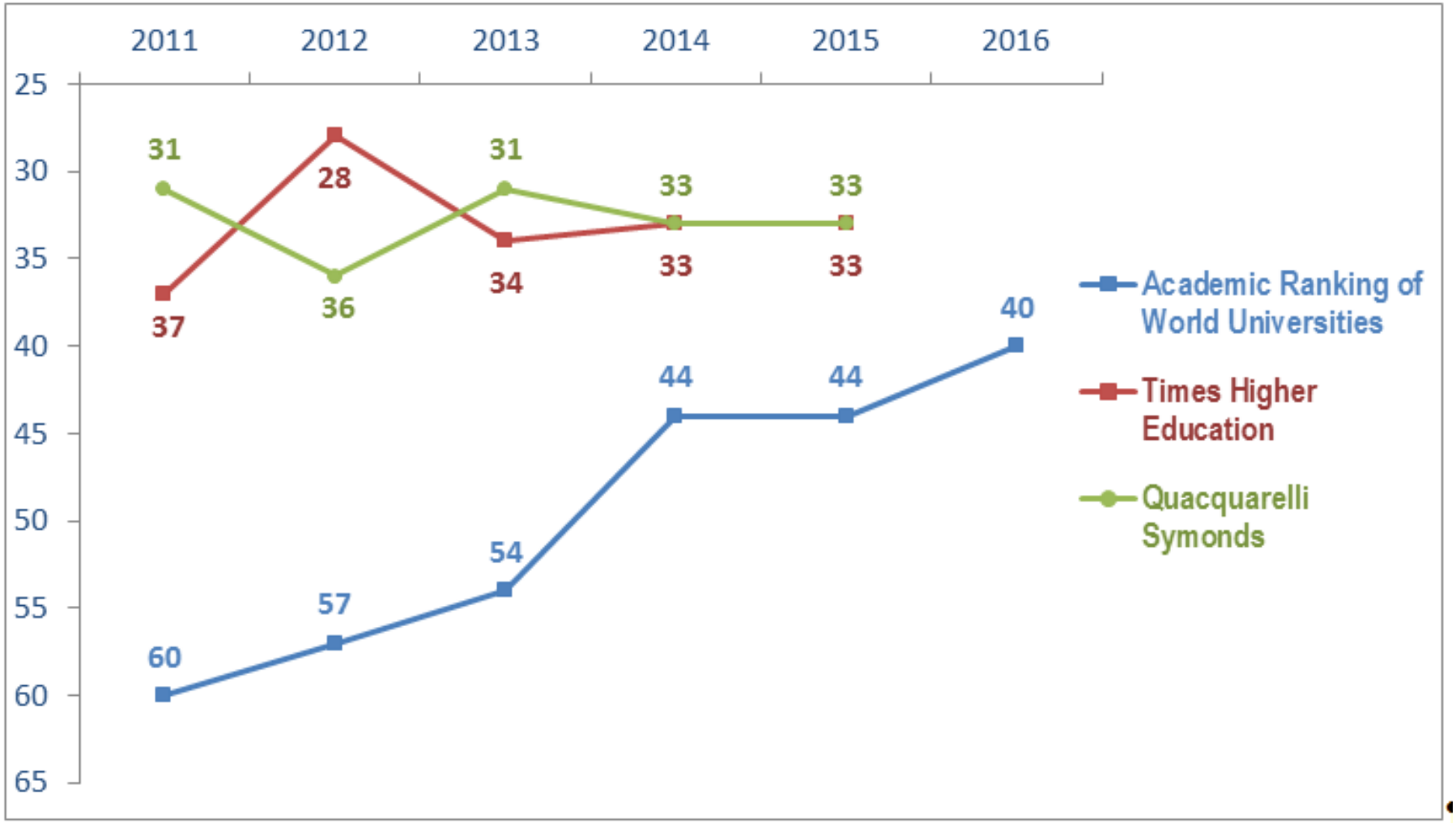
Old Quadrangle

THE UNIVERSITY OF MELBOURNE

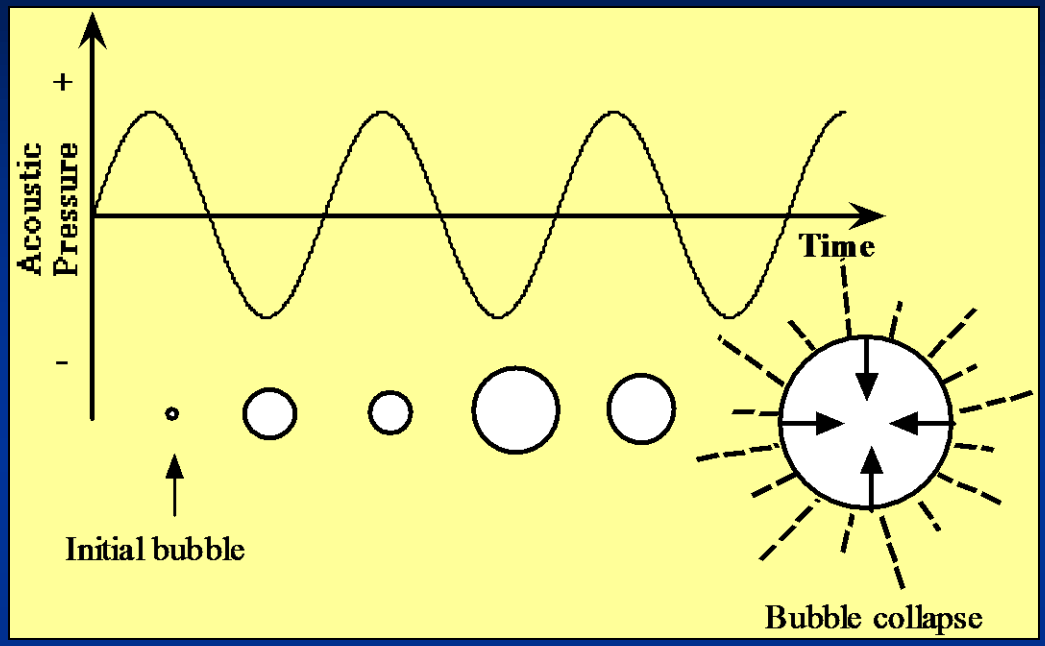
Snapshot

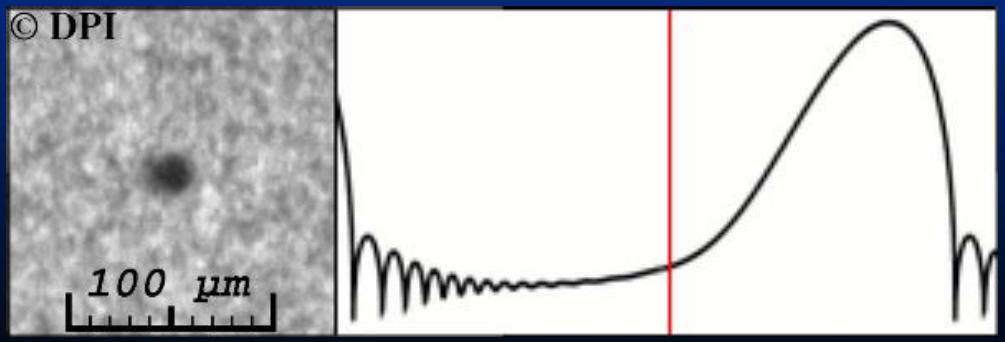
- 45,411 total student load
  - 23,384 undergraduate
  - 18,417 postgraduate coursework
  - 3,610 research higher degree
- 15,208 international student load
  - **33.5%** international students
  - from 130 countries
- Level of Study
  - 8% research higher degree
  - 41% postgraduate coursework
  - 51% undergraduate
- 8,075 staff
  - 4,068 academic
  - 3,995 professional

The only Australian University ranked in the top 50 THE, ARWU and QS global rankings

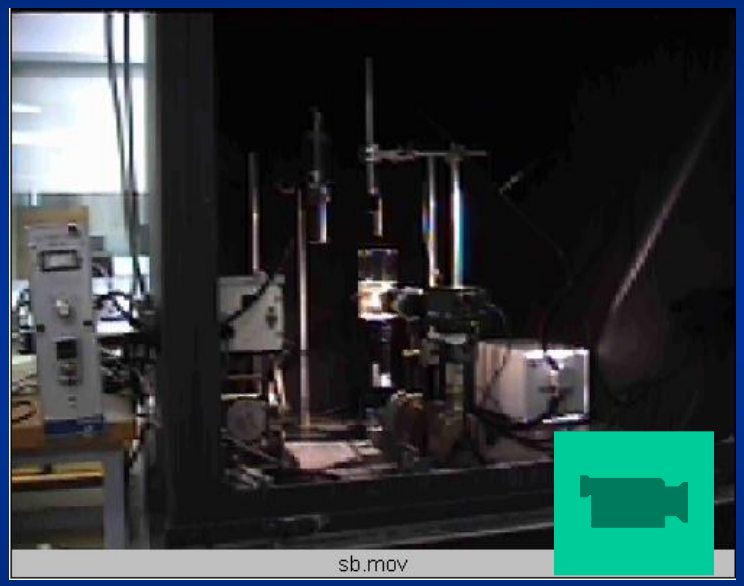


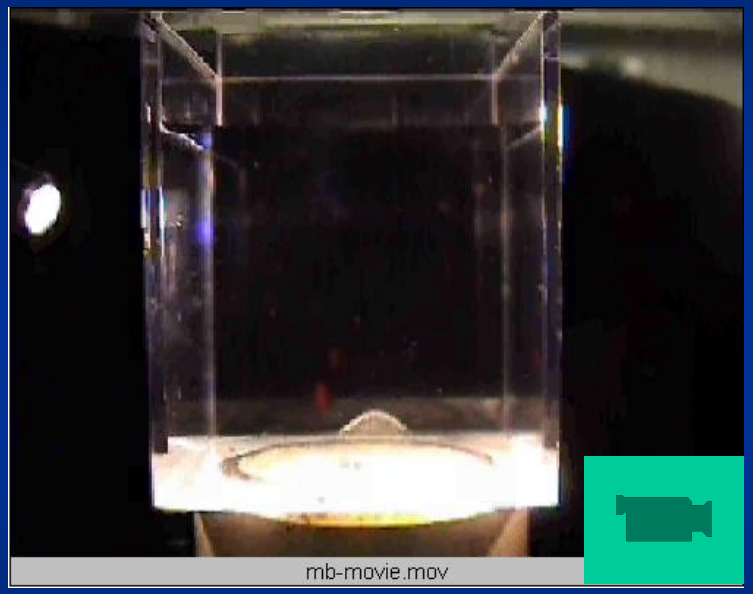
Acoustic Cavitation





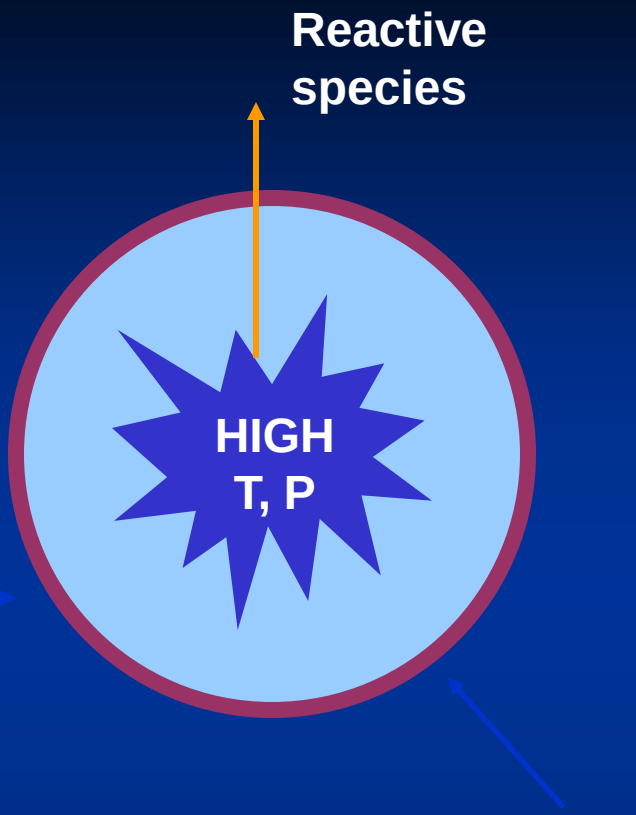
Single & multi bubble cavitation



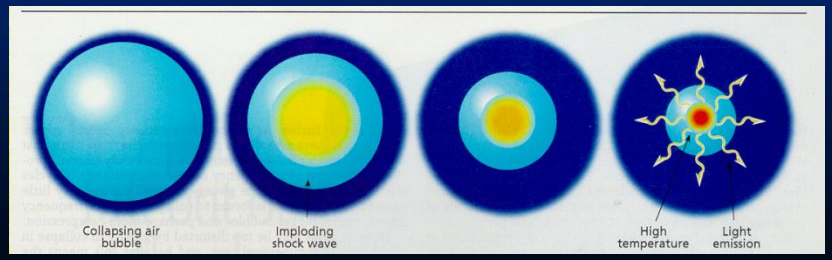


The power of cavitation

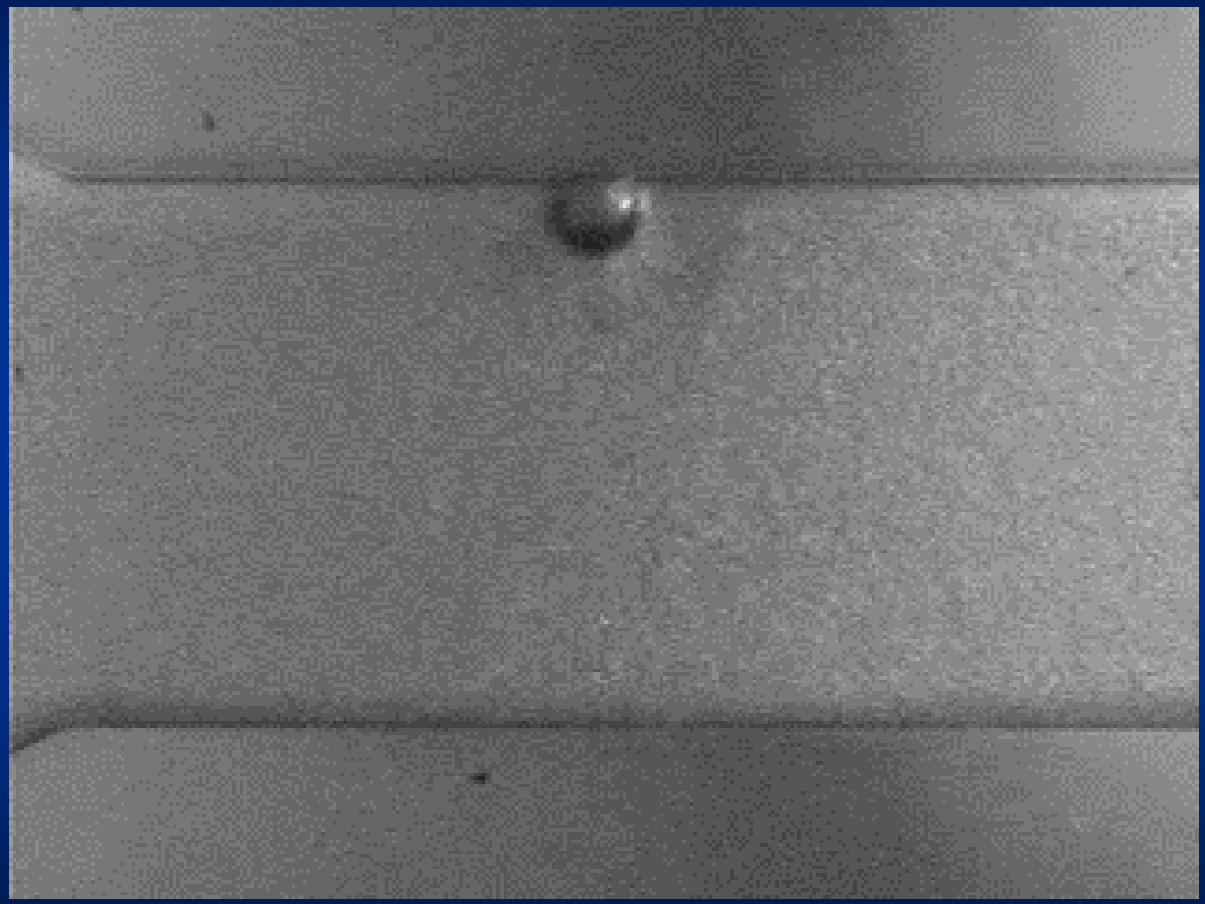
- Bubble Implosion - “near adiabatic”
  - Leads to localised areas of high T and P
  - T_{max} of the order of 5000 - 15000 K
  - P_{max} of the order of 100 - 1000 atm



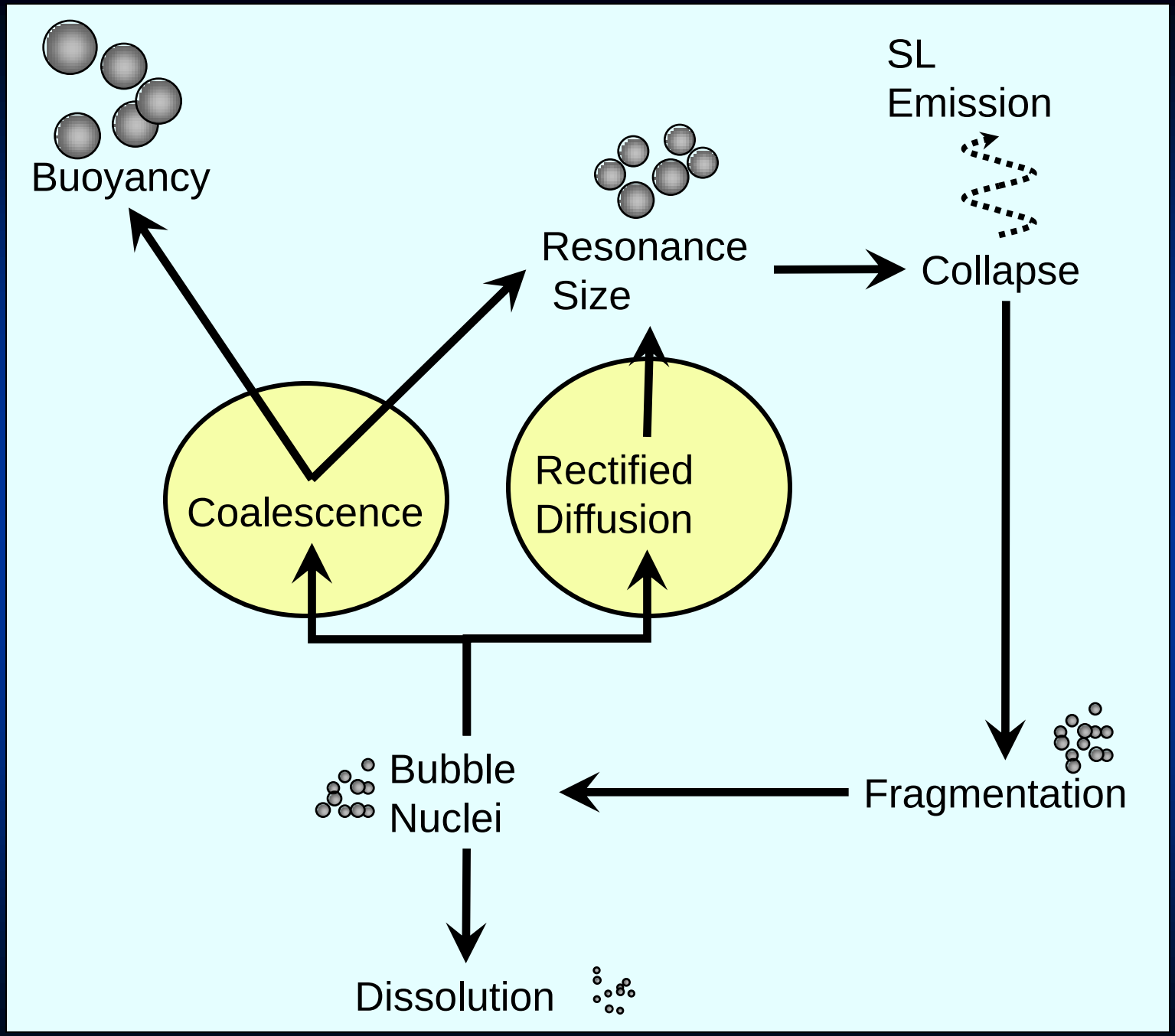
- Consequences of these extreme conditions
  - Radical generation - Sonochemistry
  - Light emission - Sonoluminescence
  - Shock waves, microjet, shear forces, etc.



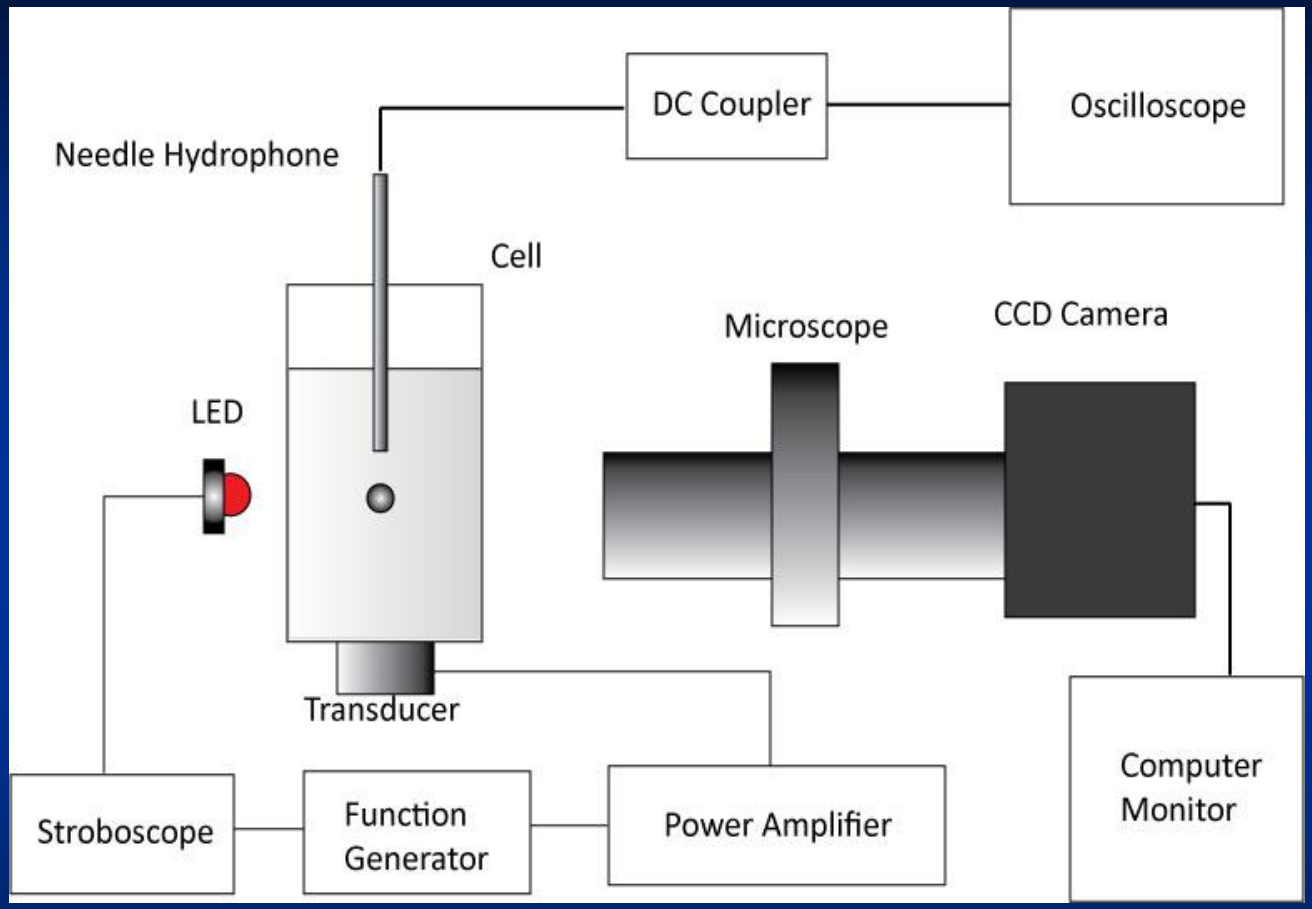
Physical forces

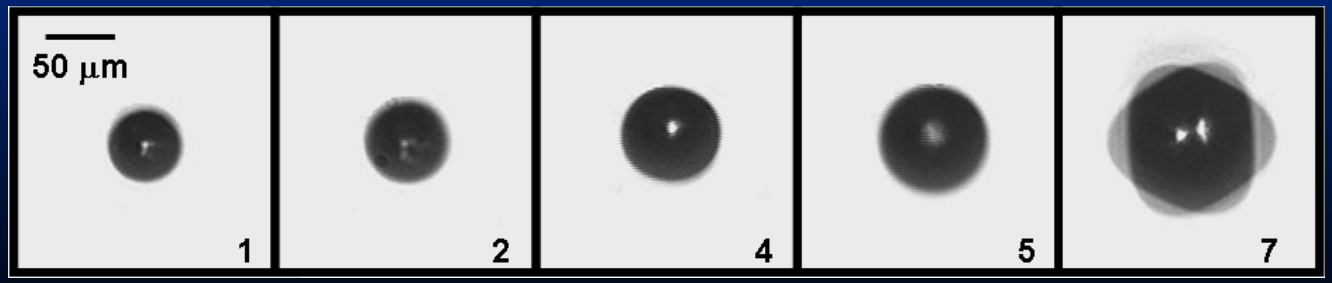


Courtesy of Dr Yasuo Iida

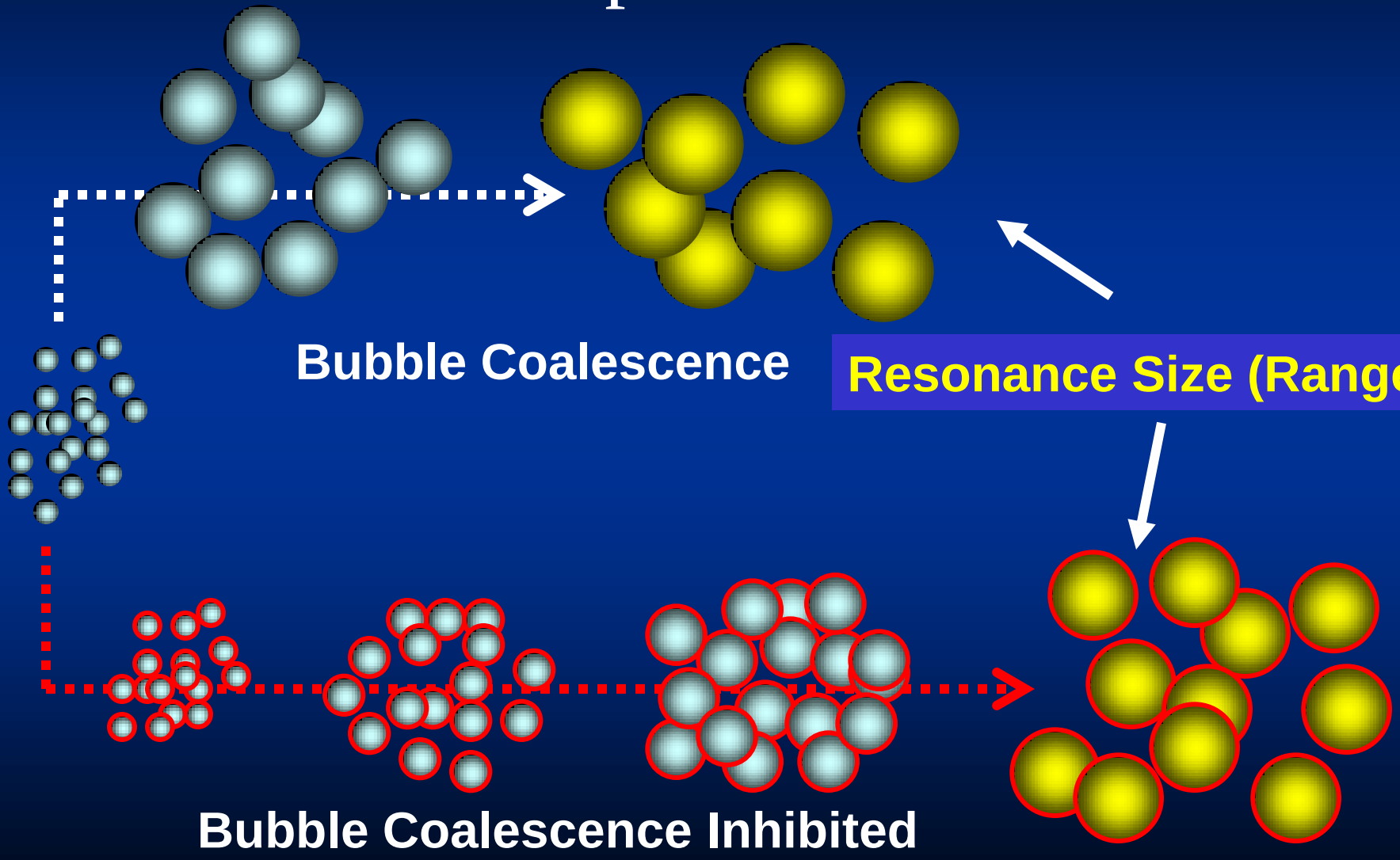

Buoyancy
Resonance
Size
SL
Emission
Collapse
Coalescence
Rectified
Diffusion
Bubble
Nuclei
Fragmentation
Dissolution

Growth by Rectified Diffusion

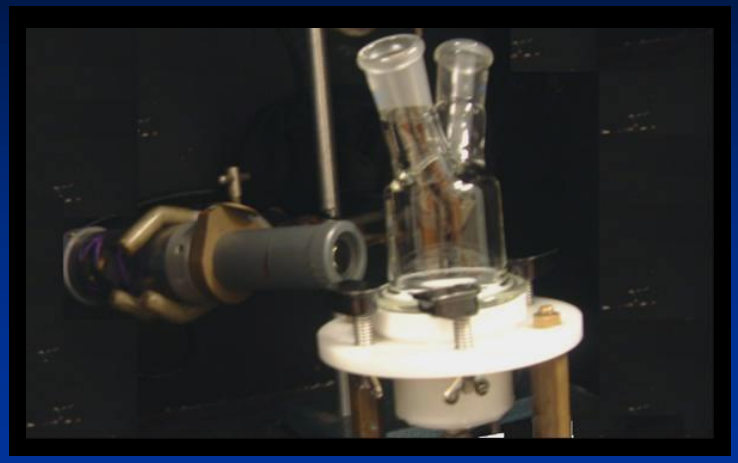




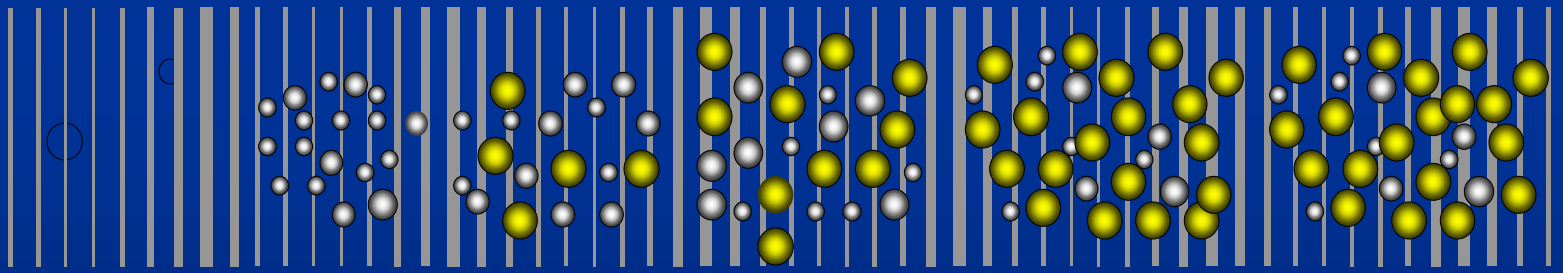
Effect of Coalescence on Active Bubble Population

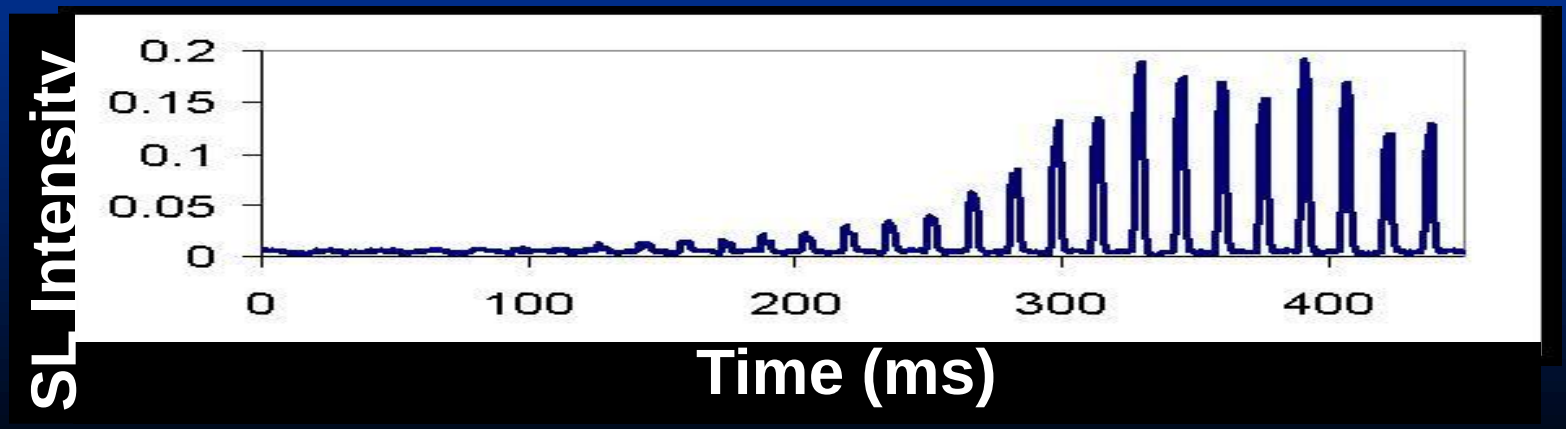


Initial Growth of Active Bubbles

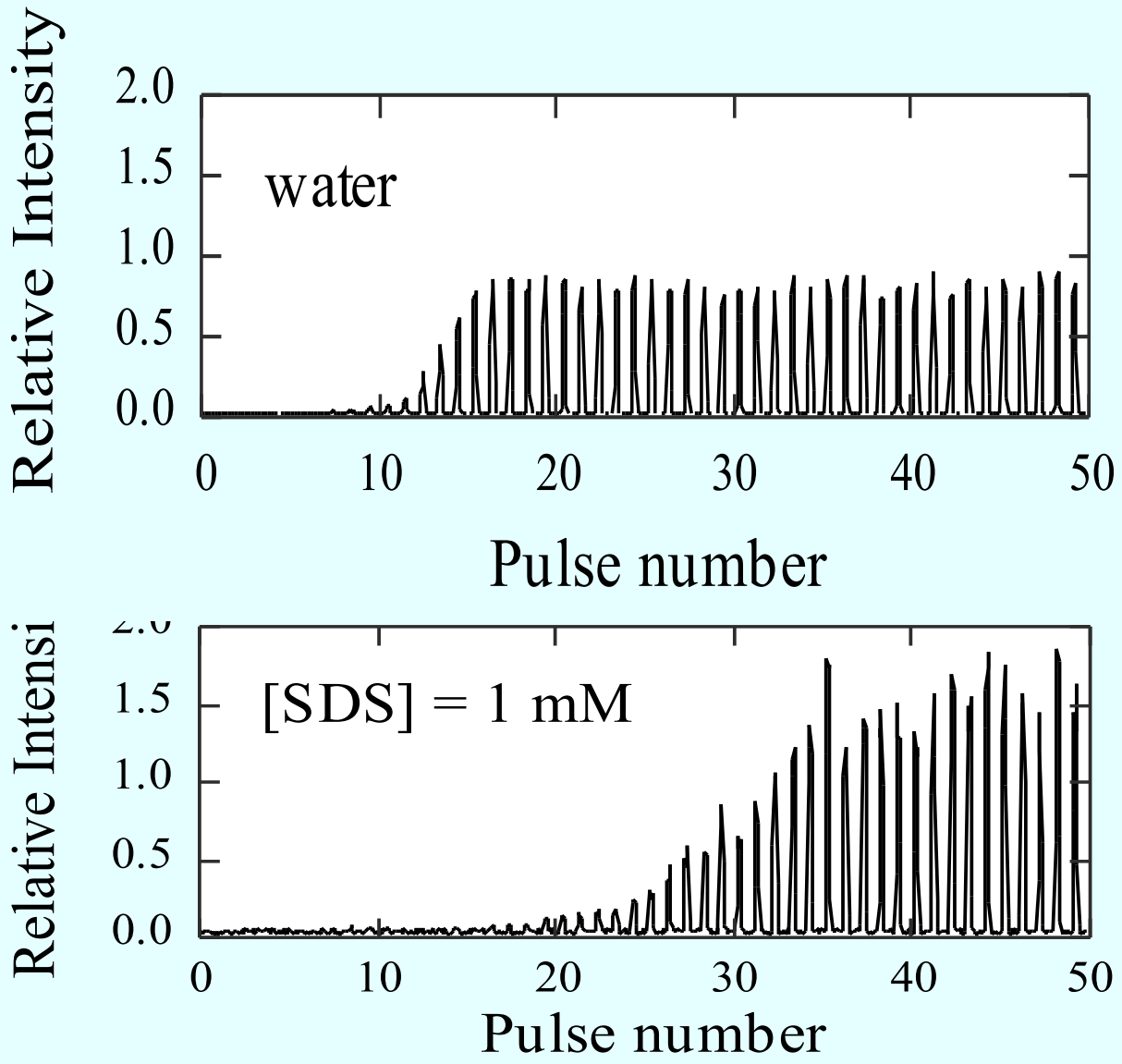


Active bubbles

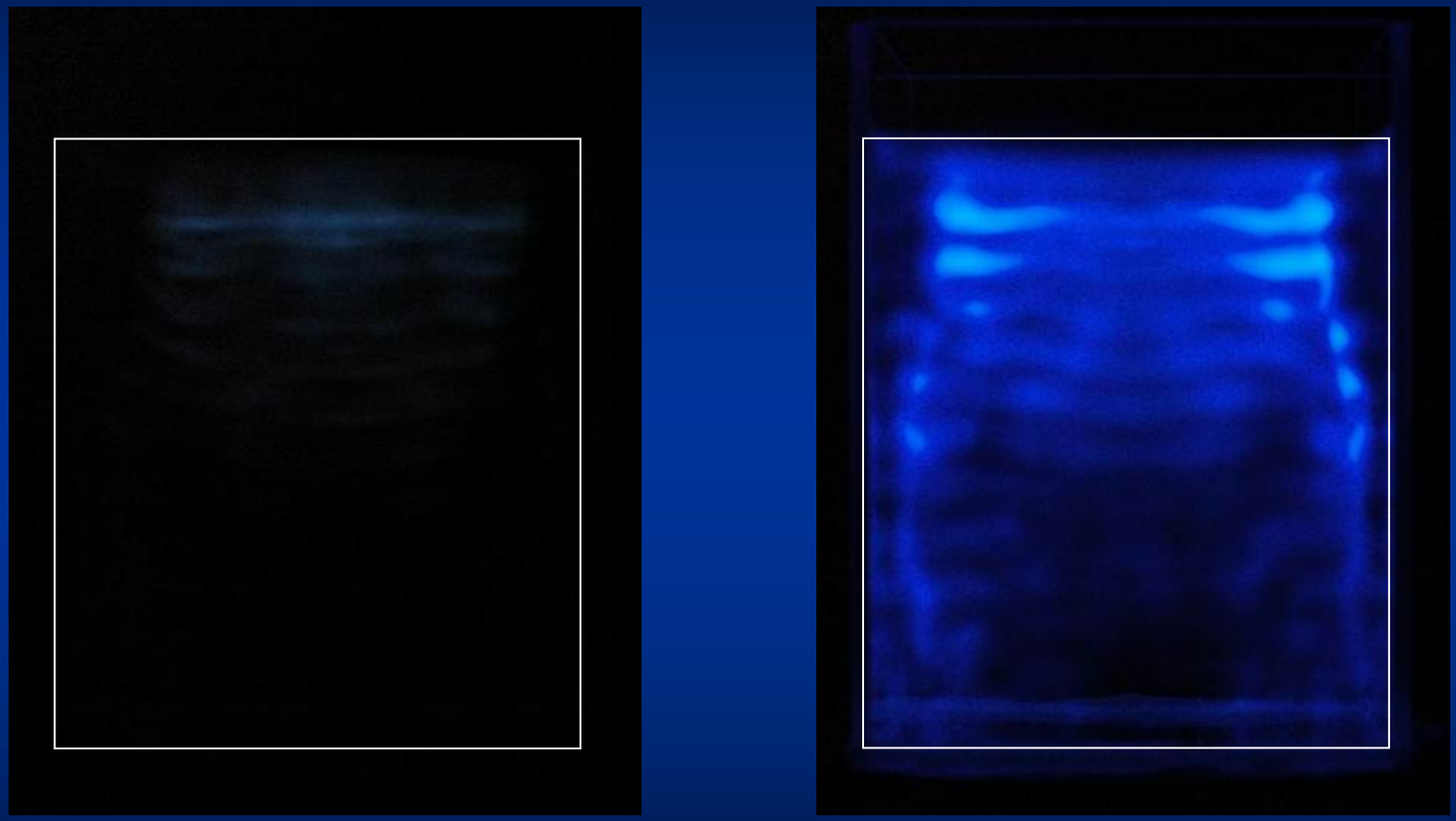




Initial Growth

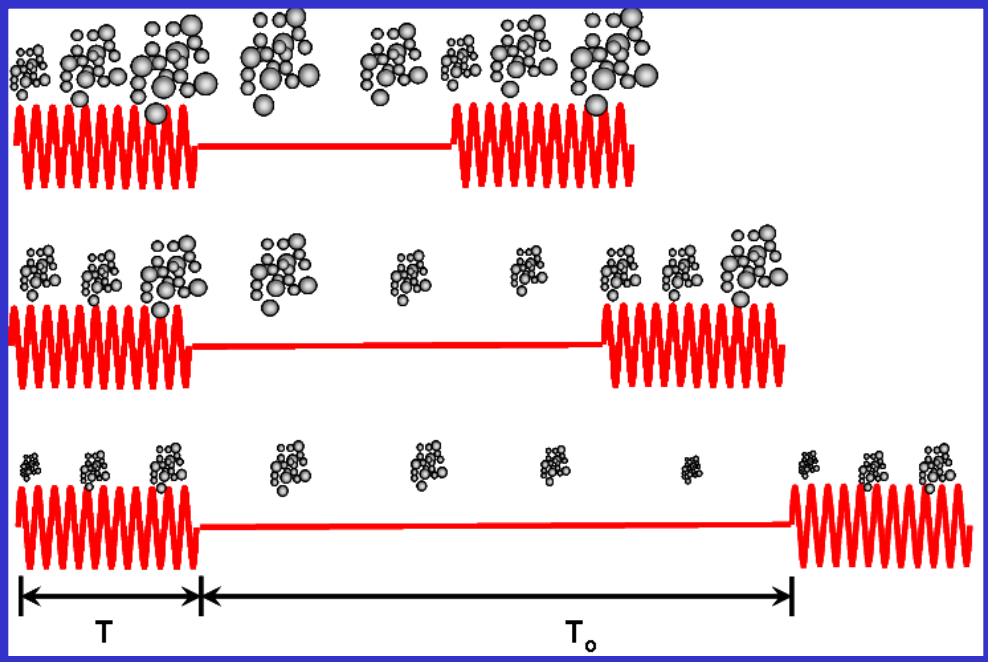


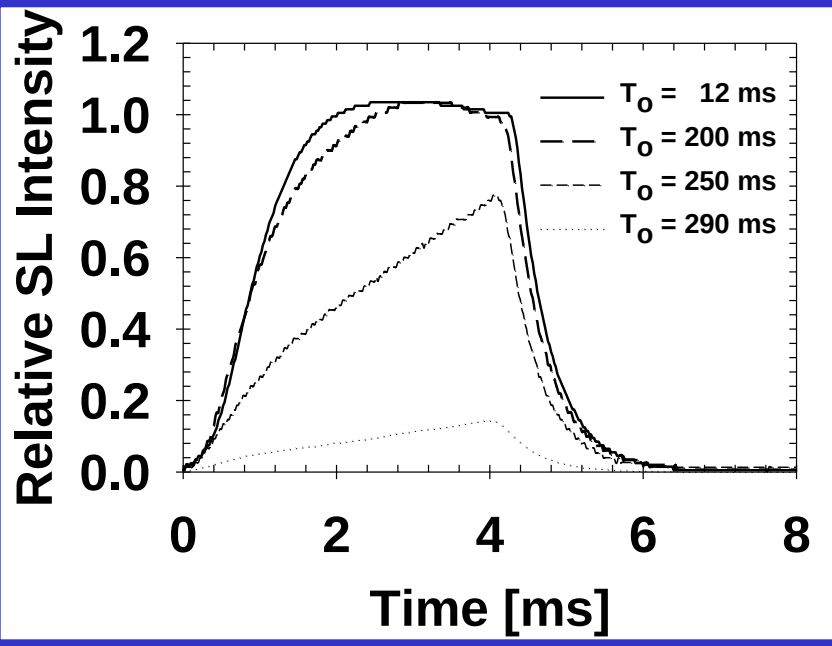
MBSL vs MBSC



Sunartio et al., ChemPhysChem, 2007, 8, 2331

Bubble Size using MBSL

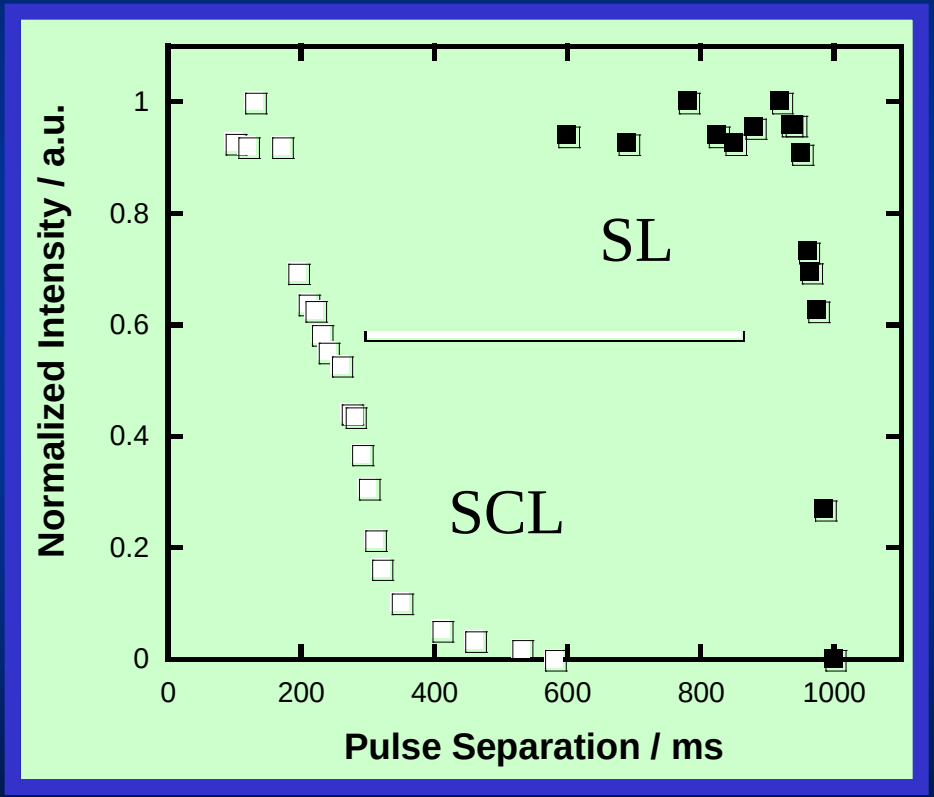


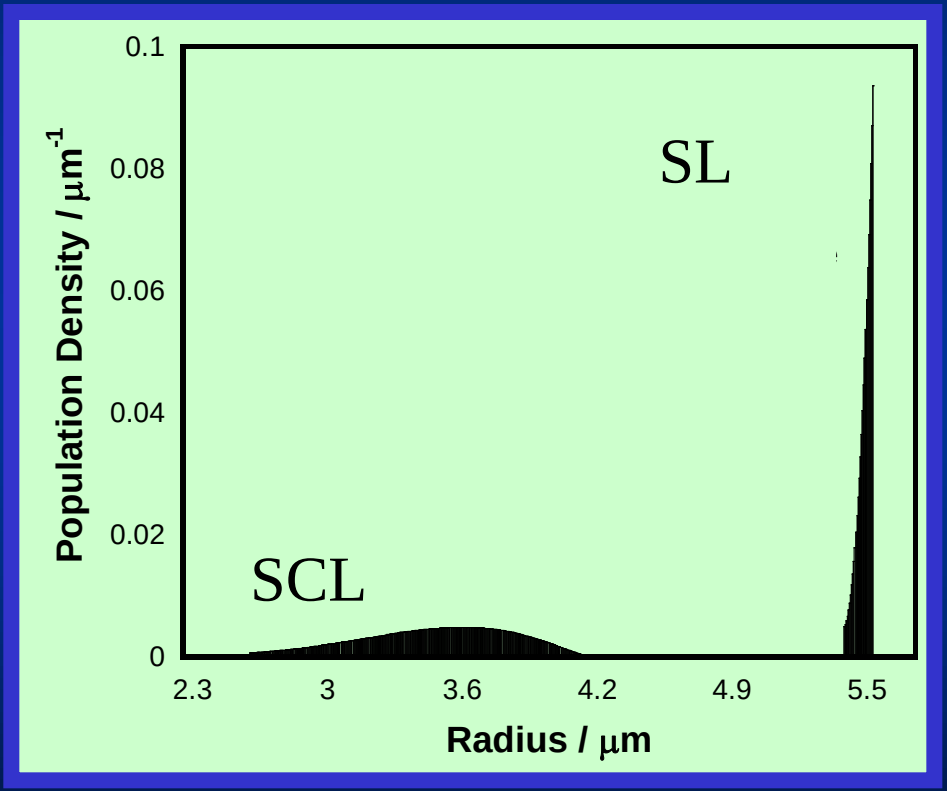


$$\left(\frac{\mathrm{D}\ C_s}{\rho_g R_o^2}\right) t = \frac{1}{3}\left(\frac{\mathcal{R}\ T\ \rho_g R_o}{2\ \mathrm{M}\ \gamma} + 1\right)$$

Lee et al., JACS, 2005, 127, 16810

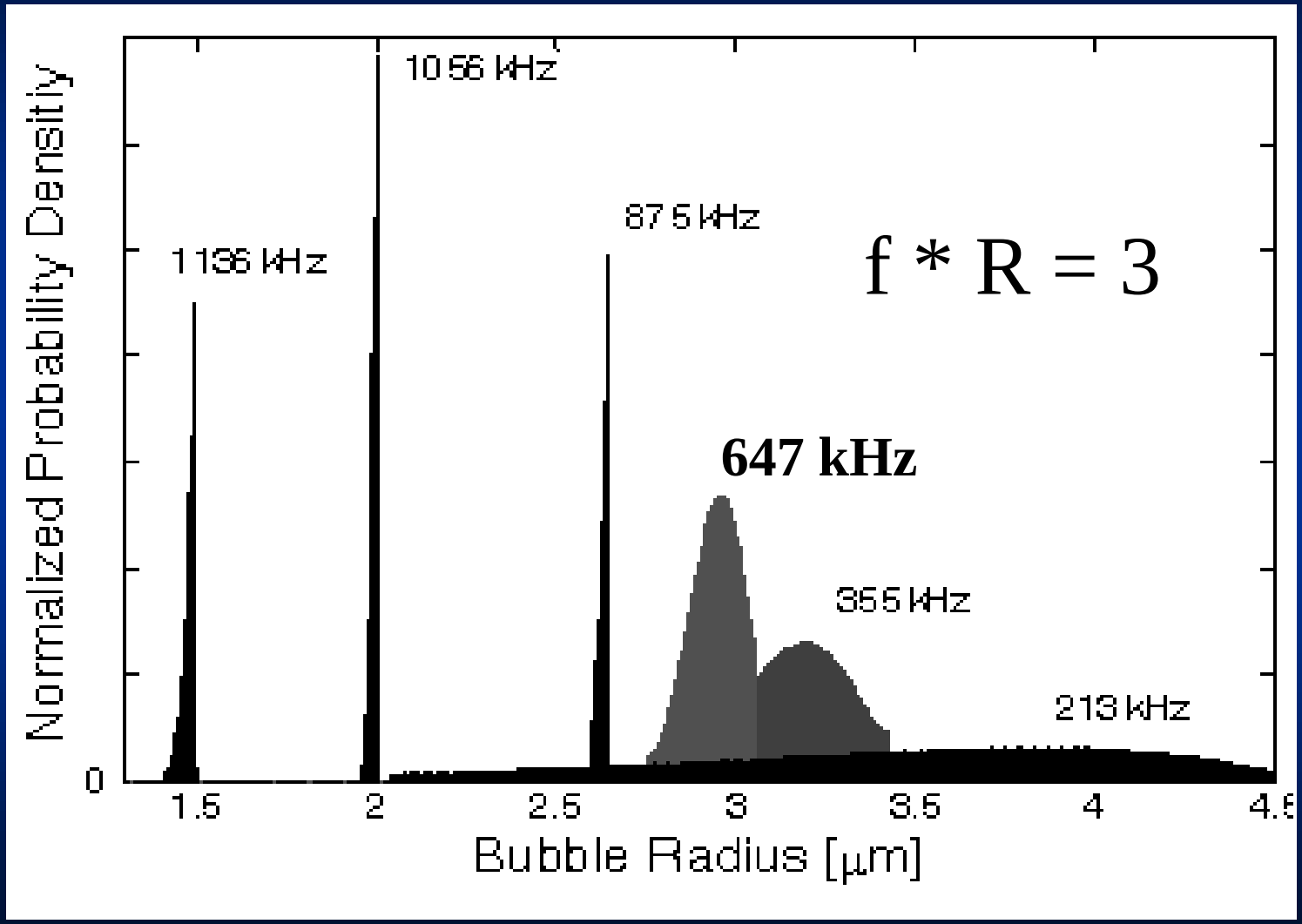
SC Bubble Size Distribution





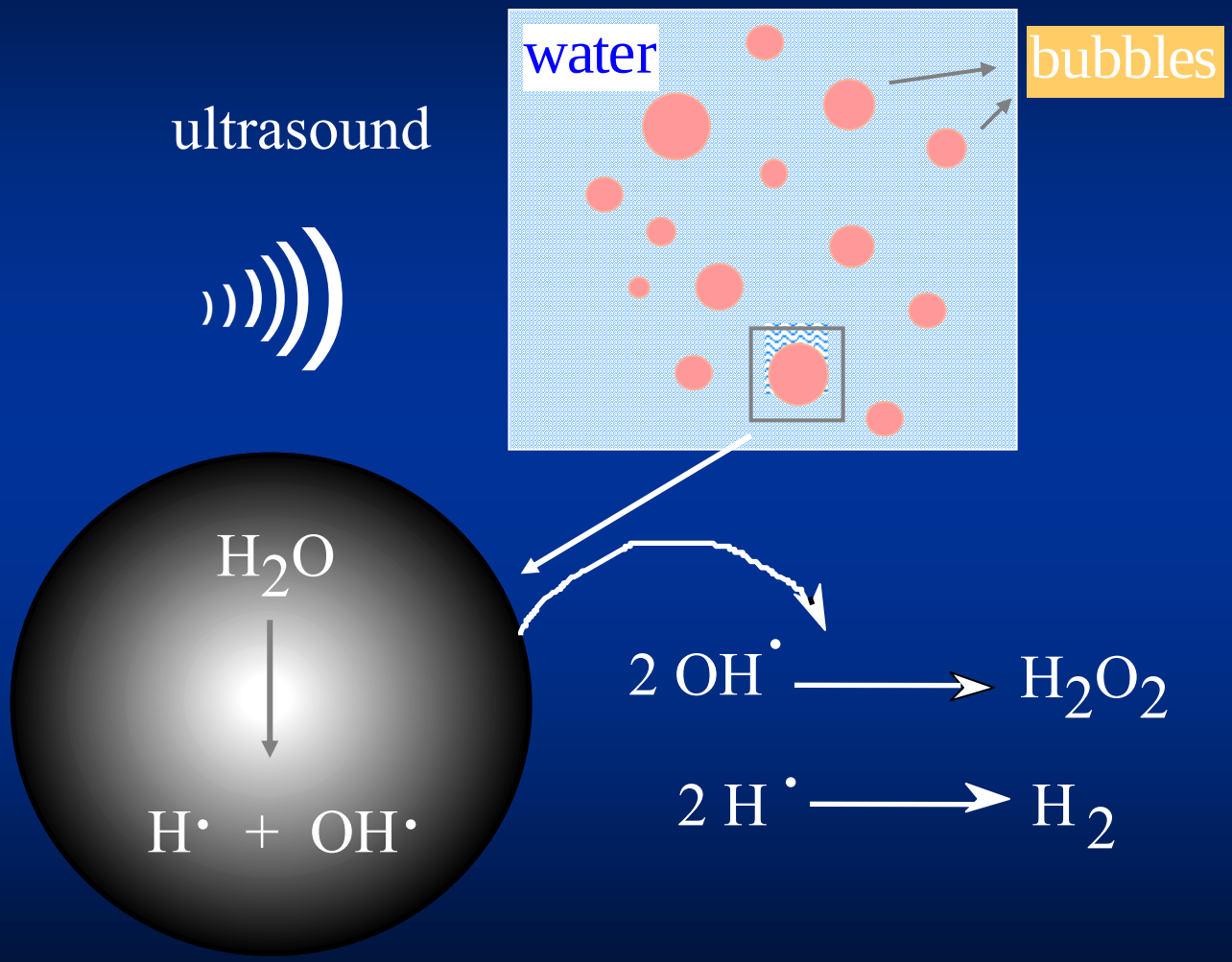
Brotchie et al., PRL, 2009

SC Bubble Size Distribution

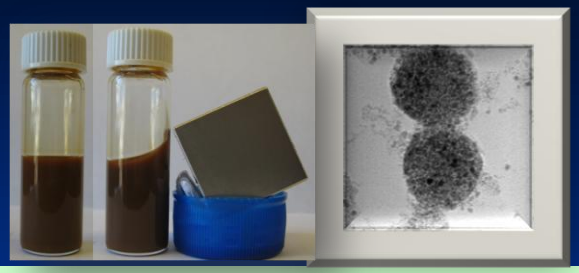


Brotchie et al., PRL, 2009

Primary Radicals

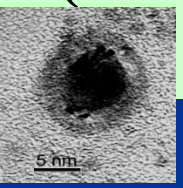


Current Research Projects



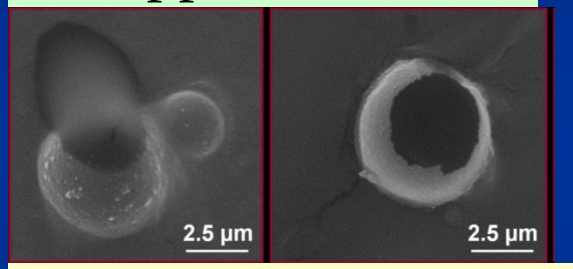
Fundamentals of acoustic cavitation

Functional microspheres for biomedical applications



Nanoparticle synthesis (catalysts, functional materials)

Energy
Environment
Health
Food

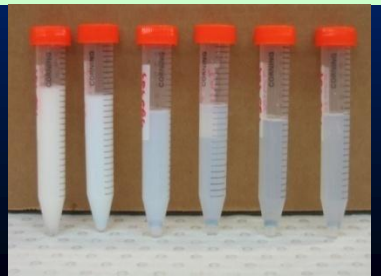


Flavor and nutrient encapsulation

Food and dairy processing

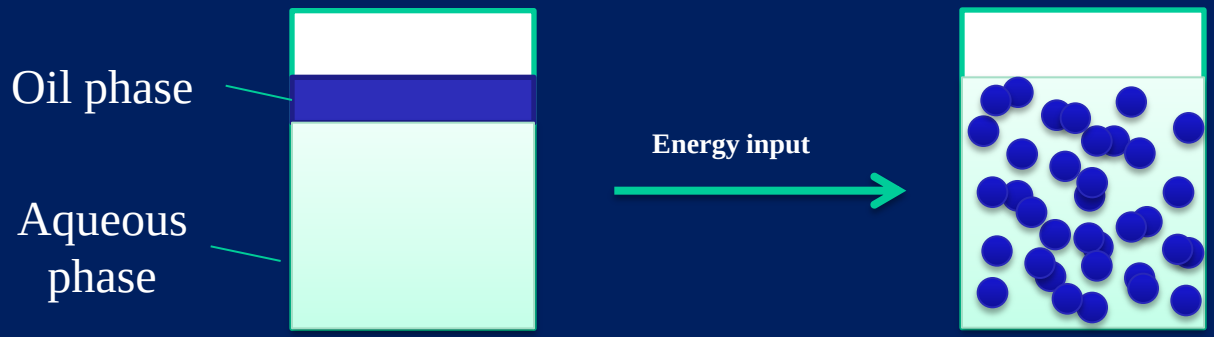
Wastewater treatment

Sonoprocessing



Physical forces

Food emulsions

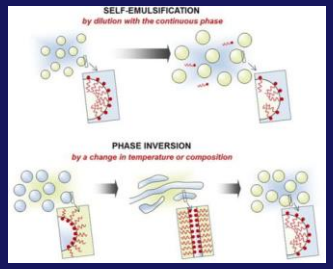


Stability dependent on

1) Emulsion droplet size
2) Surfactants
3) Storage conditions

Emulsification techniques

Rotor Stator/high speed mixing	High pressure homogenization (>300 bar)	Micro-fluidization	Phase inversion
• Cost-effective • Simple operation • Limited effectiveness in creating nanosized emulsions • High speed moving parts	• Very effective at shearing droplets • High throughput capability • Challenging to clean and maintain	• Extremely effective at shearing droplets • Medium to high throughput capability • Challenging to clean and maintain • Expensive	• Low energy process • Spontaneous formation possible • Often requires large amounts of surfactant to carry out



Creation of stable surfactant-free simple emulsions in milk

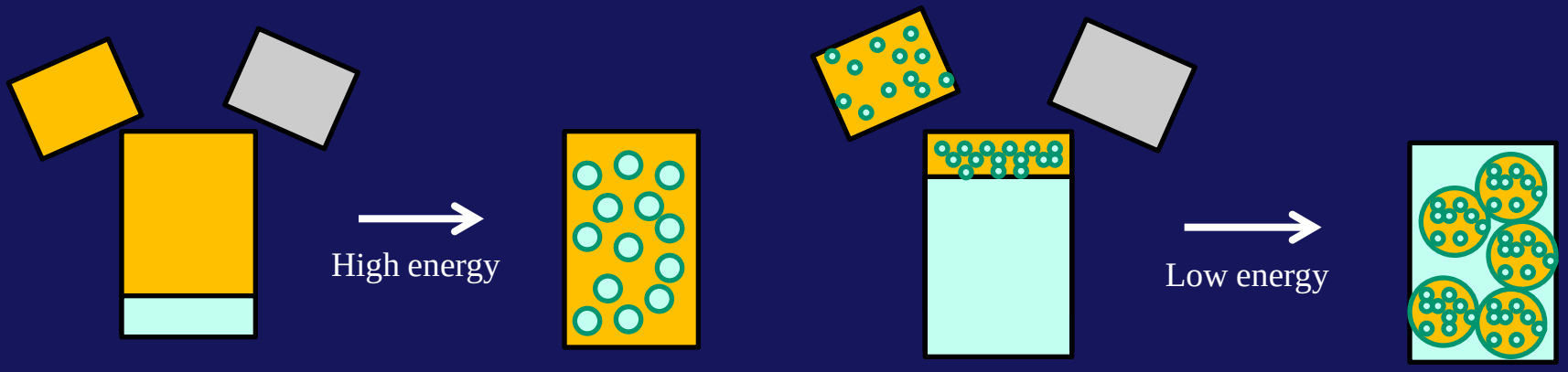
12.5 min
Ultraturrax

5 min
ultrasonication

	D (4,3) µm	Dv50 µm	Dv90 µm
U.S. 5 min	0.48	0.34	0.99
U.T. 12.5 min	1.54	1.3	3.3

Shanmugam & Ashokkumar, Food Hydrocolloids, 2014

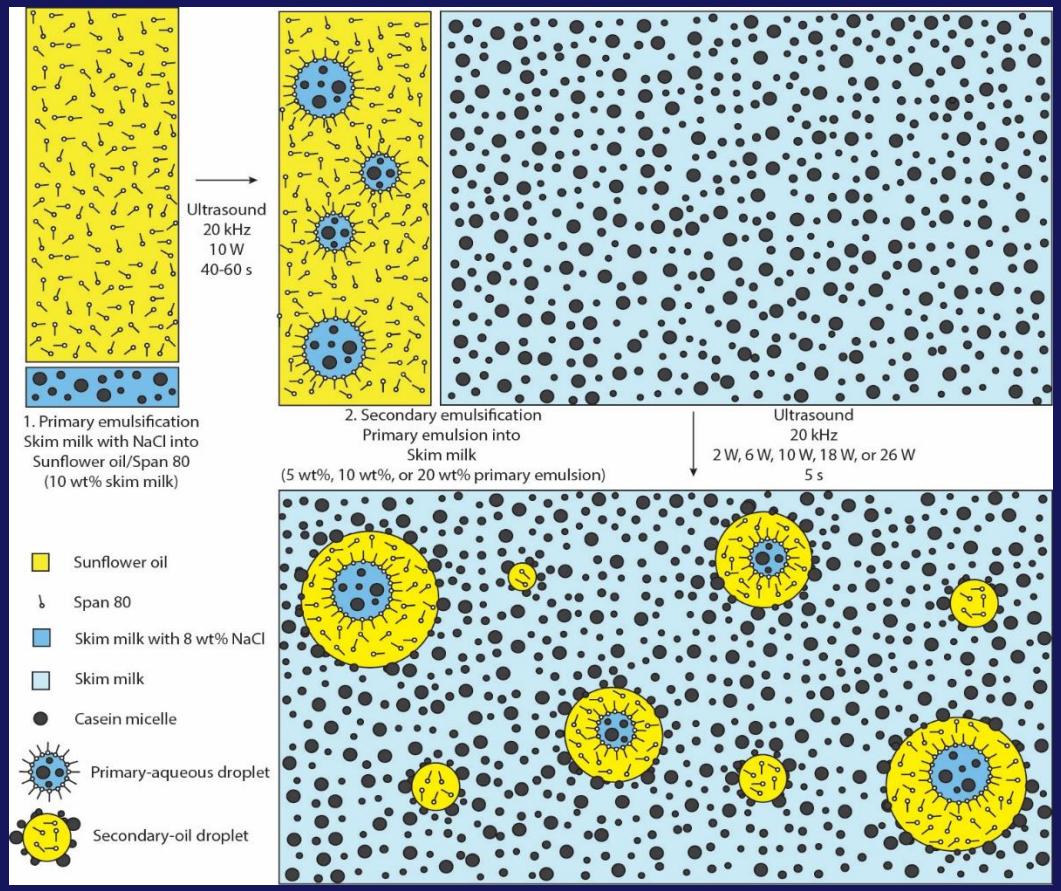
Water-in-oil-in-water double emulsions



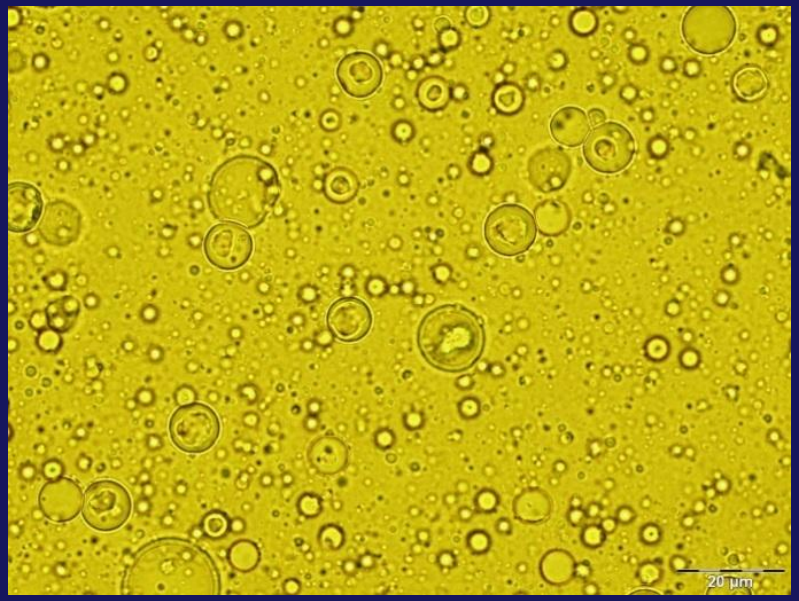
- Encapsulate water soluble bioactive materials such as flavours and nutrients
- Improve sensory properties of reduced fat products by fat displacement

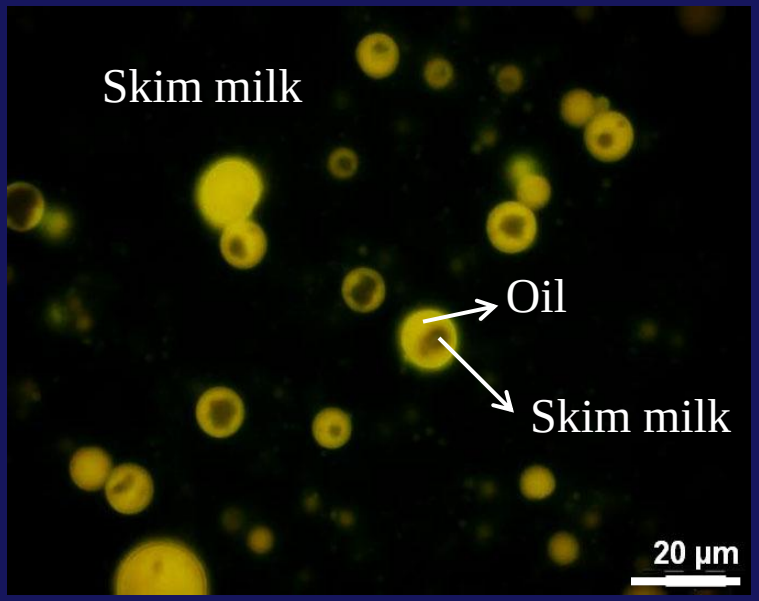
Formation of double emulsions using ultrasound

- Step 1: Formation of W/O emulsion with 10 wt% skim milk loaded into oil phase – energy requirement ~ 50 kJ/kg
  - Span 80 surfactant used at 10 wt% of oil phase
- Step 2: Formation of W/O/W emulsion with 5-20 wt% - energy requirement ~ 2-20 kJ/kg
  - No additional surfactant required in skim milk outer phase

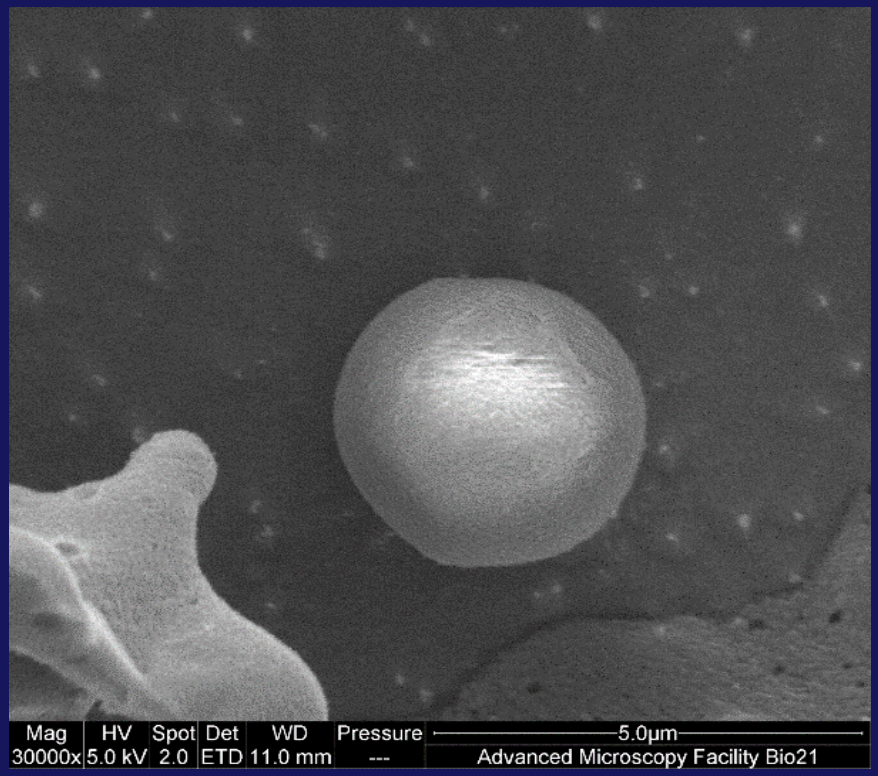


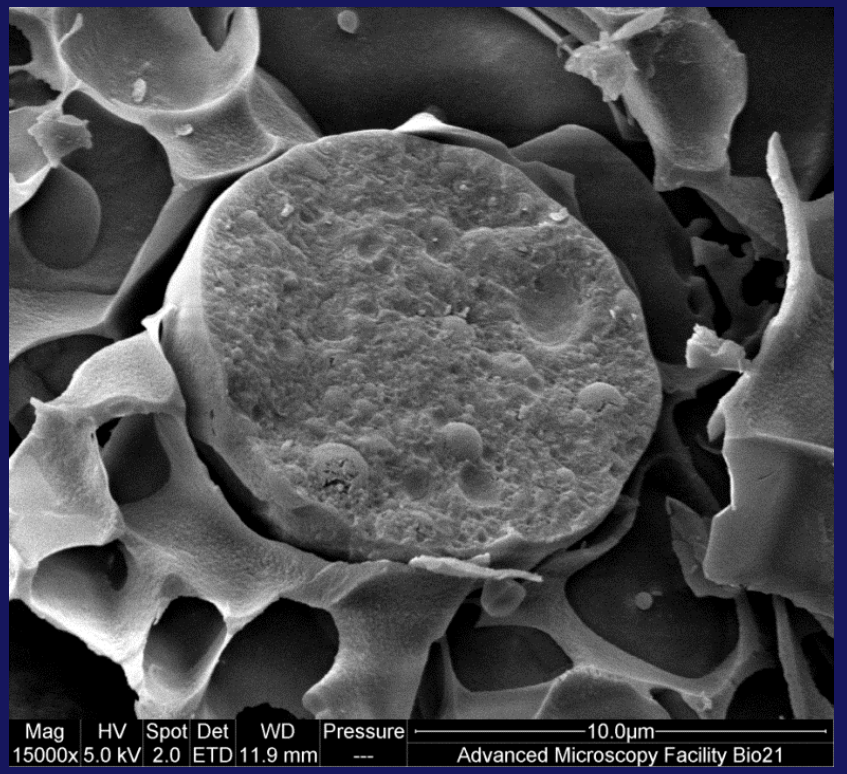
Double emulsion formed in skim milk



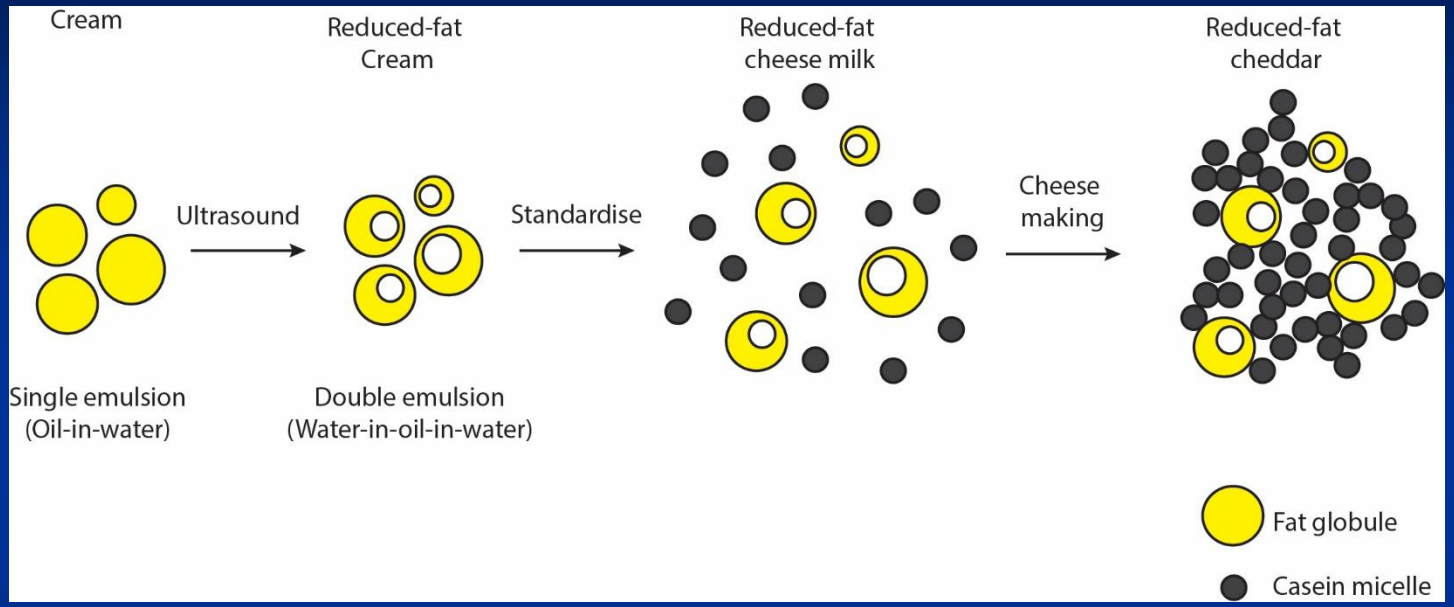


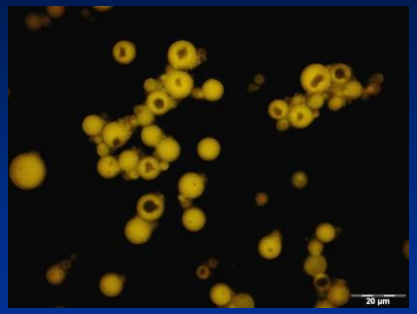
Surface and internal morphology





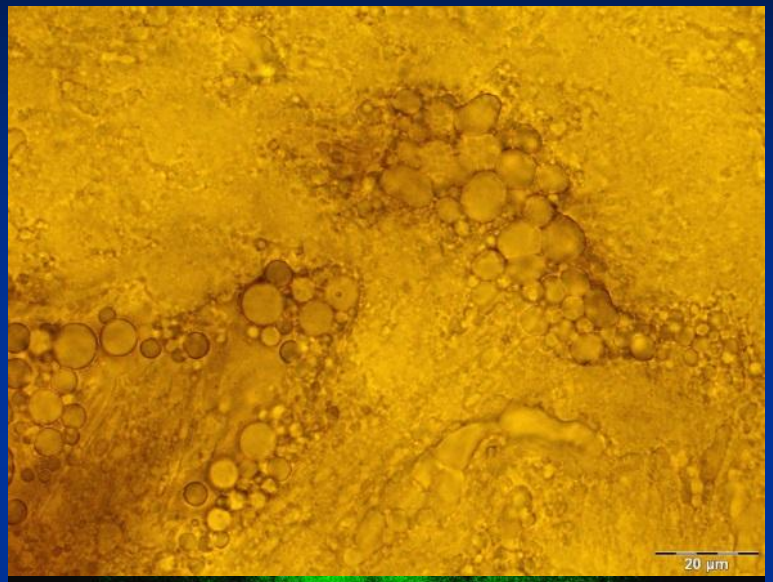
Example – Improving low fat cheddar production with double emulsions

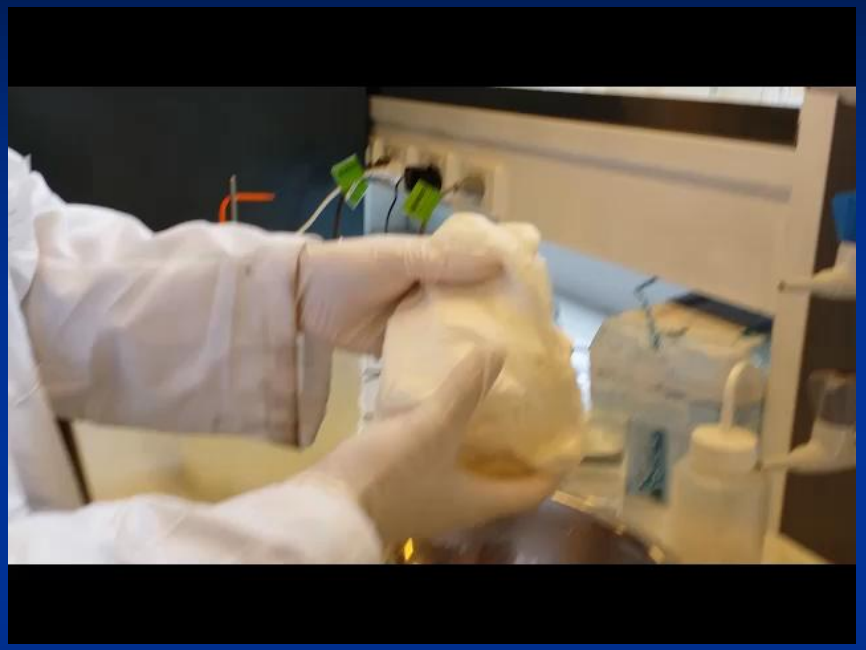




- How much fat can be displaced?
- What is the cost?
- What are the properties of the reduced fat cheddar?
- Are there any side-effects?

Sunflower oil double emulsion cheese proof of concept

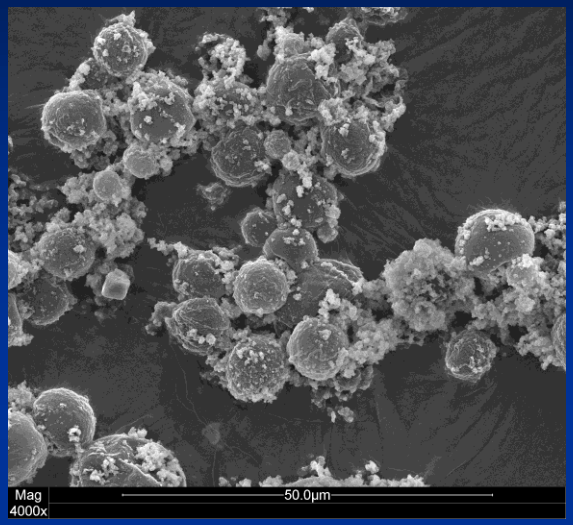


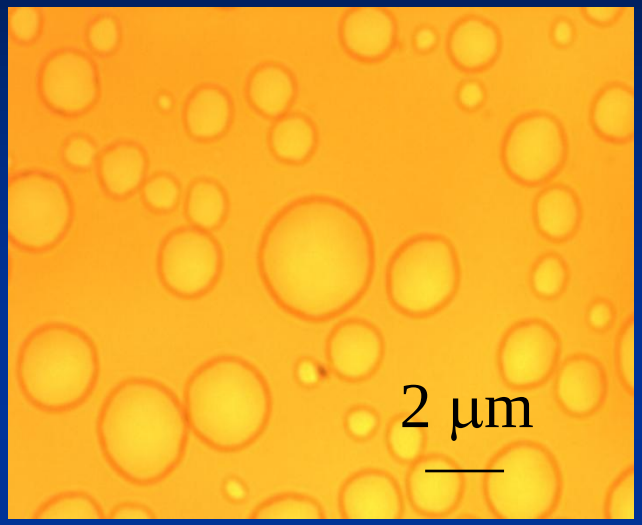


Microspheres

Potential Applications: Encapsulation/Delivery

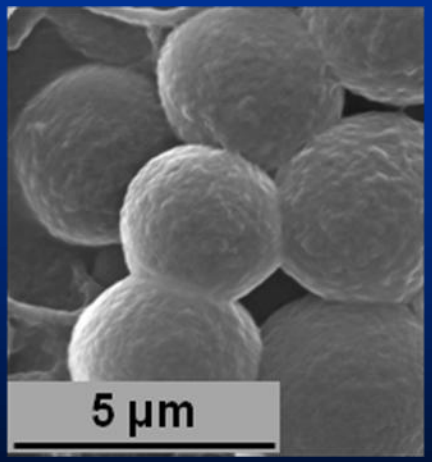
Lysozyme

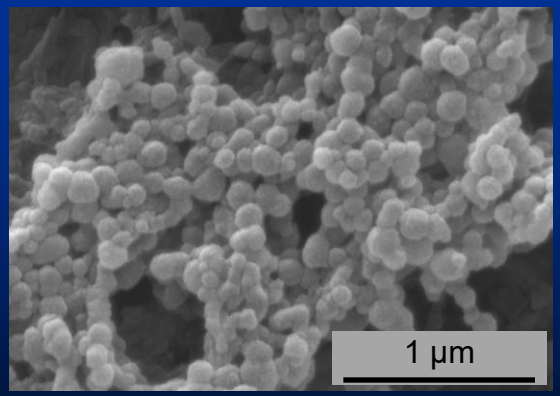




Casein

Whey protein

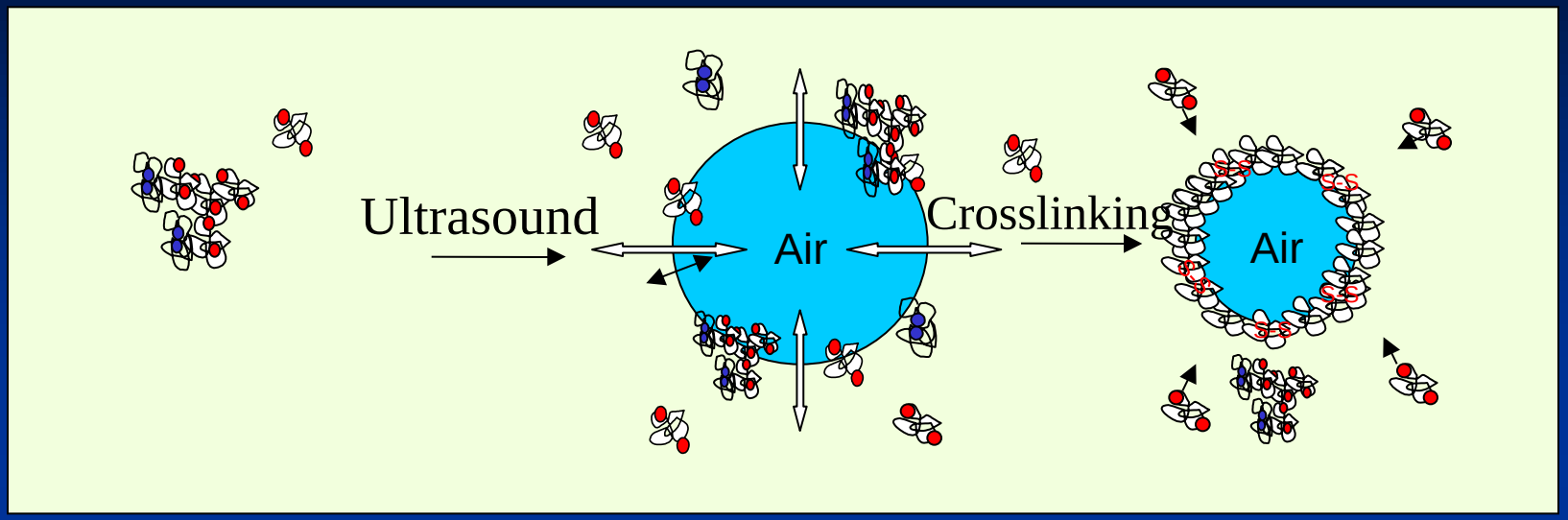




Fish oil filled Whey protein MBs

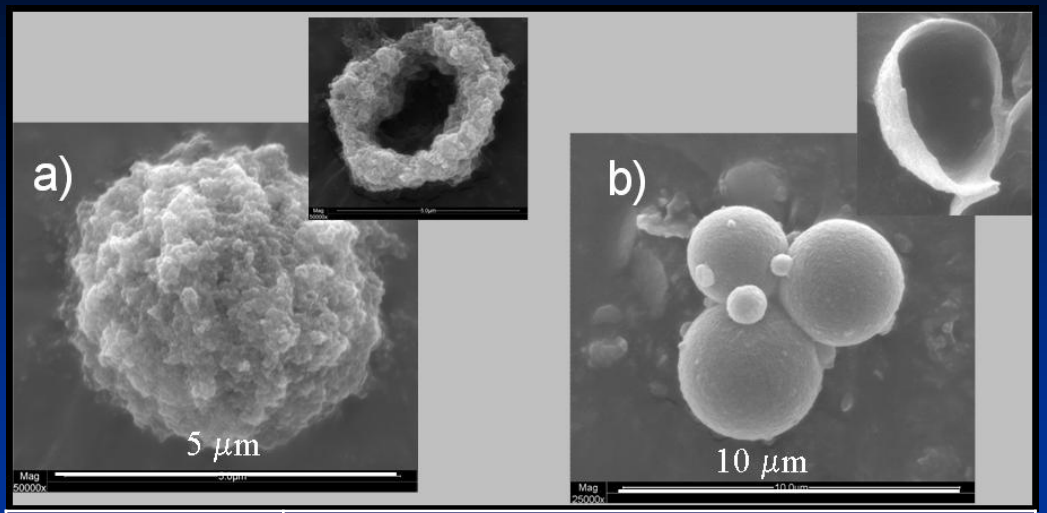
Cavalieri et al, Langmuir, 2008, 24, 10078

Mechanism of microsphere formation



- protein unfolding (increases hydrophobic attraction between molecules)
- aggregation at air/water interface generated by ultrasound treatment
- Intermolecular crosslinking

Mechanism of microsphere formation

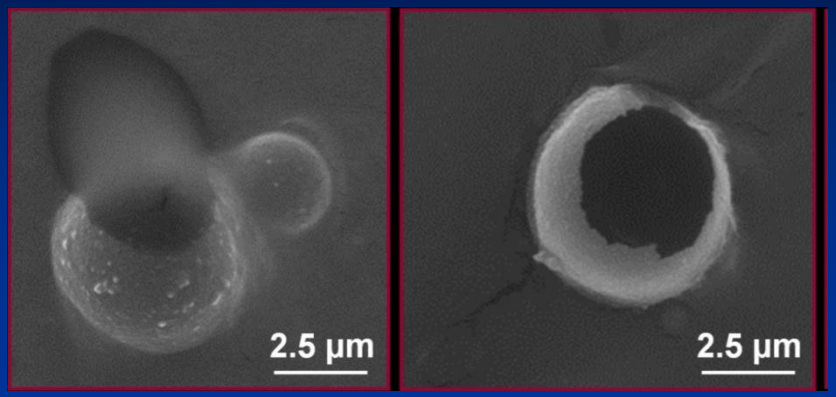


Samples	Microbubble Size (μm)
PMA	not stable
PMA_{SH5}	not stable
PMA_{SH10}	4.3 ± 0.5
PMA_{SH30}	8 ± 1

Chem Commun, 2011, 47, 4096

Microspheres

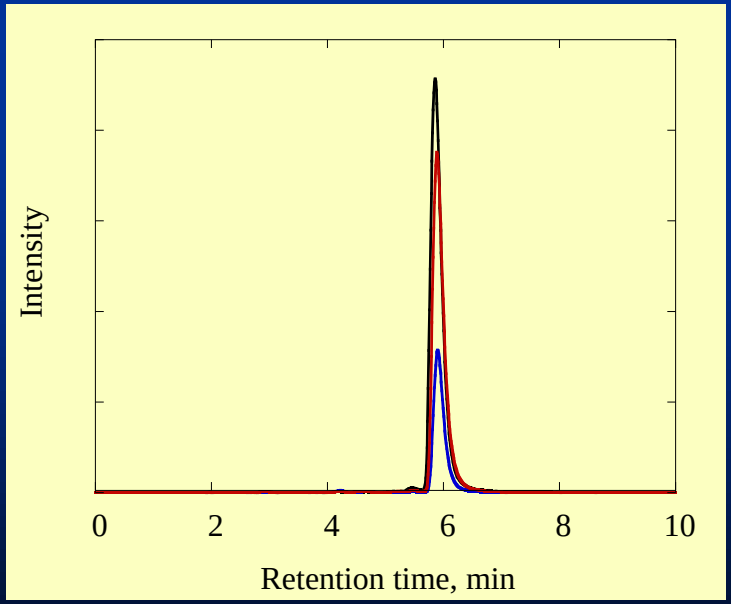
Potential Application: Nutrient encapsulation



Chitosan

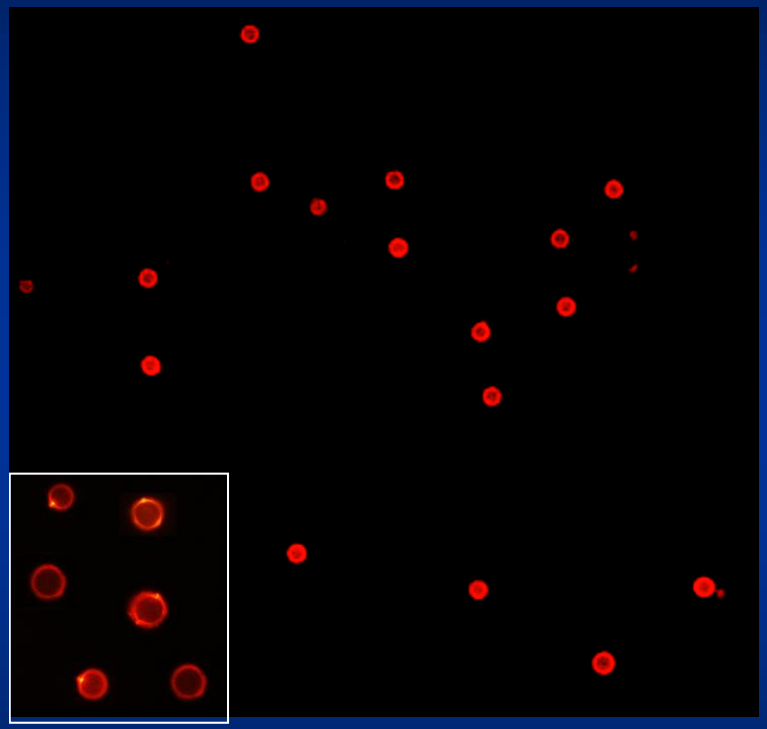
β-Lactoglobulin
Lysozyme
Whey proteins
BSA
Chitosan

Fish oil
Vitamin E
Carotene

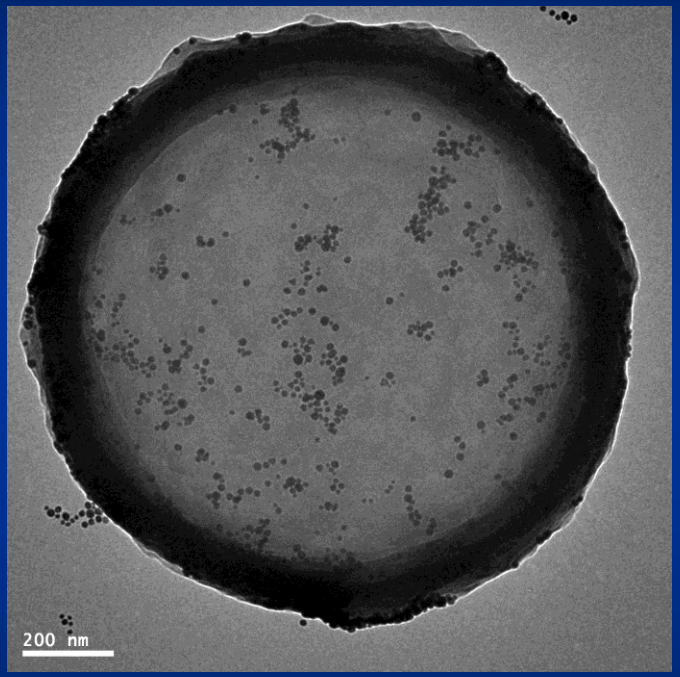


Zhou et al., manuscript in preparation

Functional Properties of Microbubbles

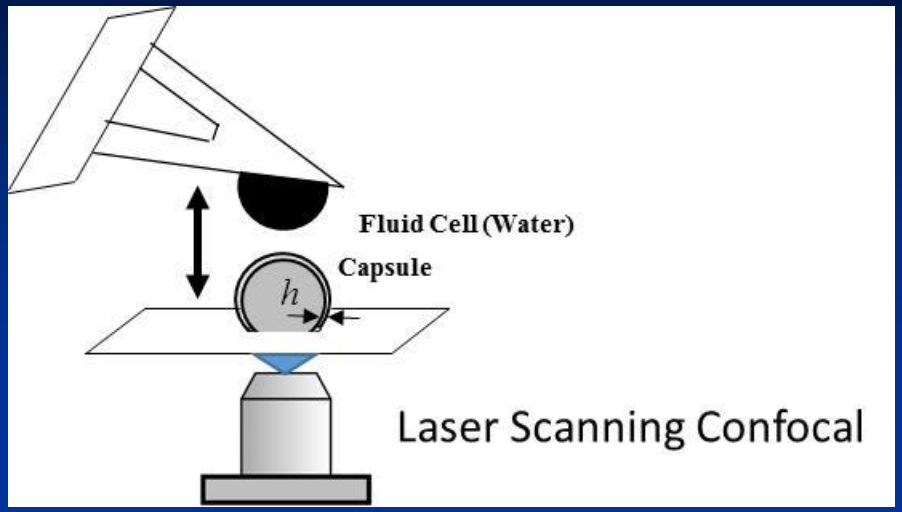


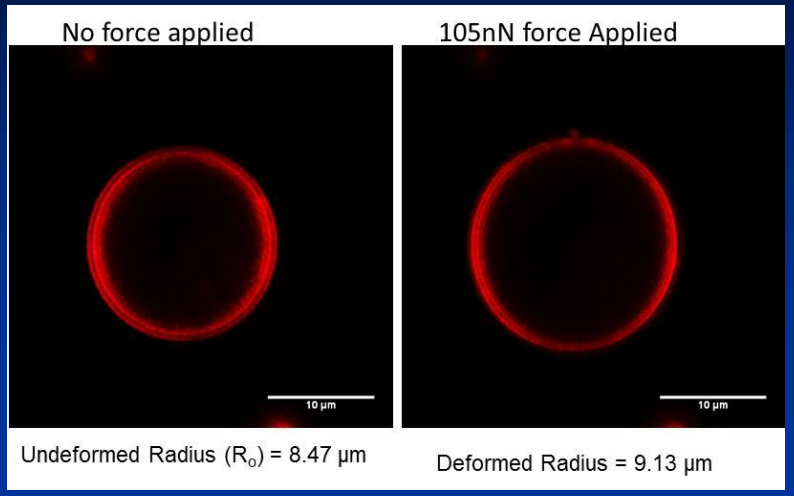
Optical fluorescence microscopy image of doxorubicin-loaded microcapsules (main figure) and microbubbles (inset).

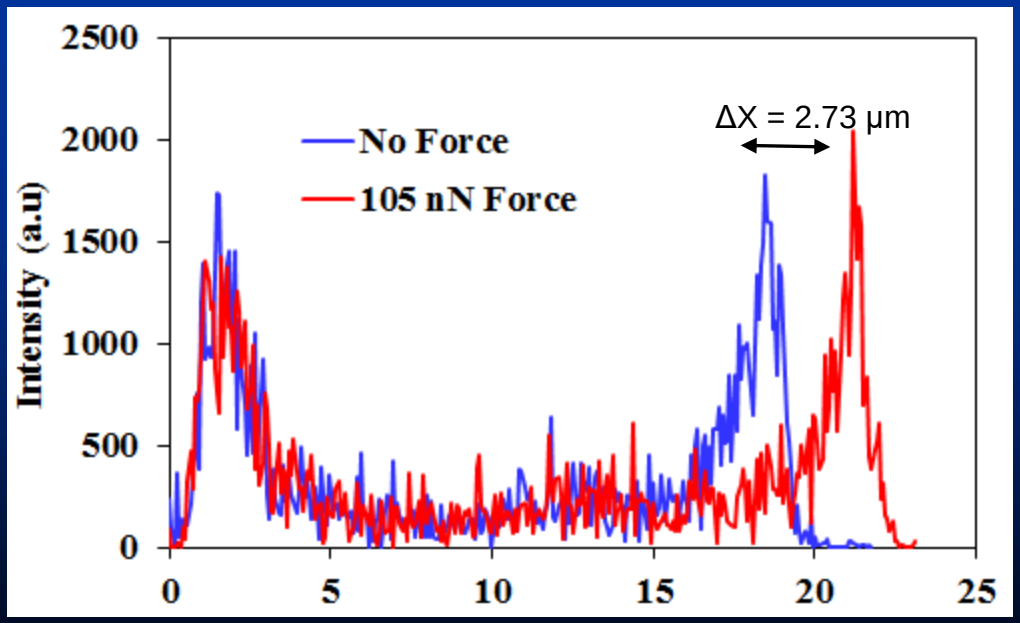


Gold particle loaded-microbubbles

Combined AFM and Laser Scanning Confocal Microscopy







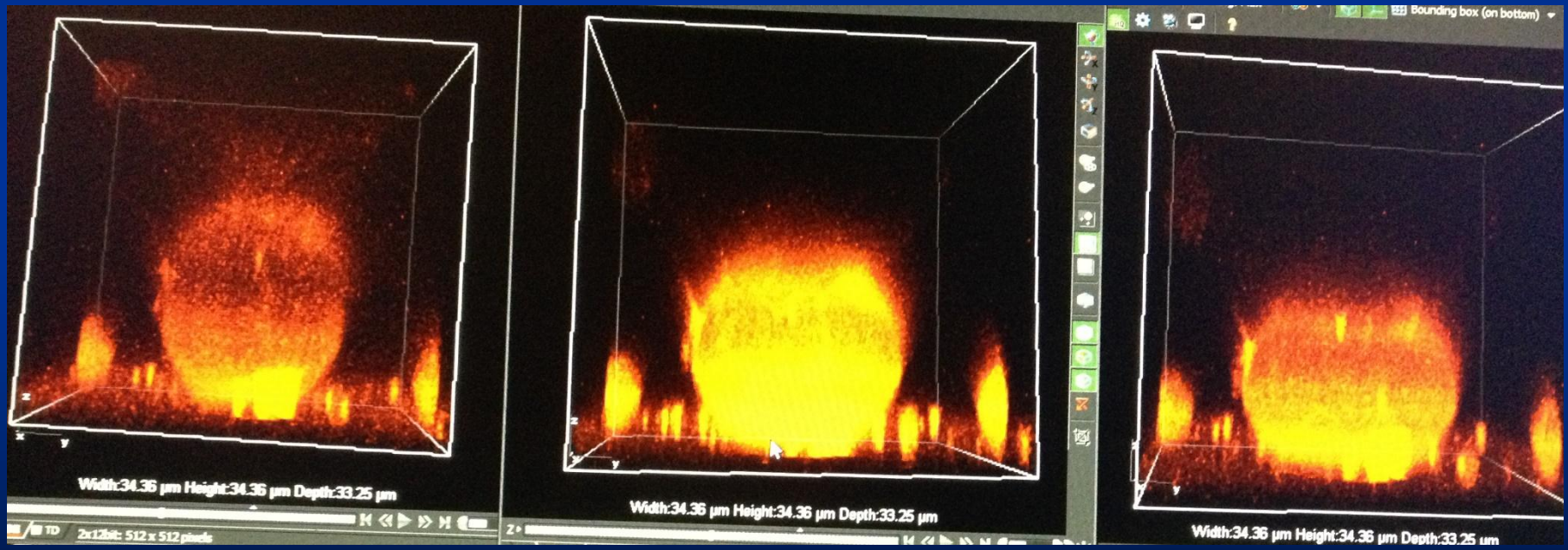
Combined AFM and Laser Scanning Confocal Microscopy

3D Scanning

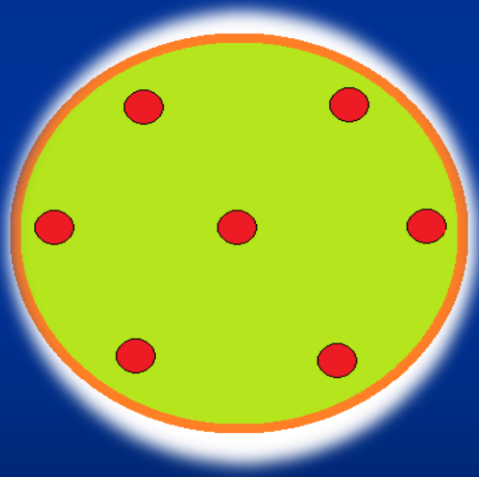
No force applied

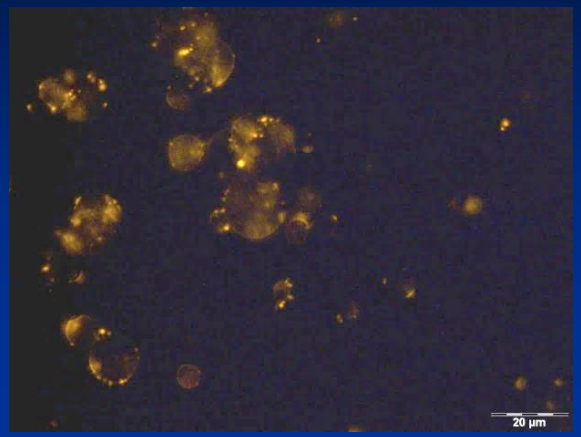
105nN force Applied

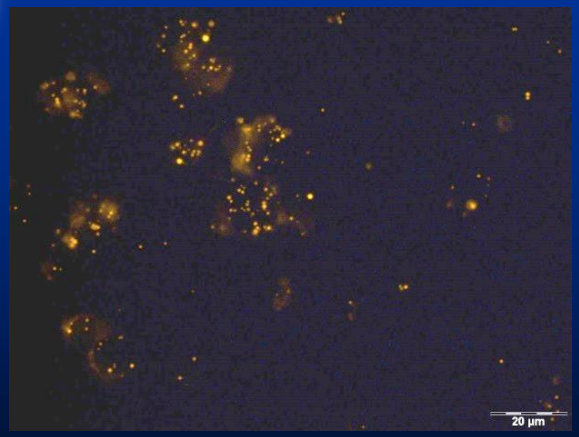
Very large force Applied

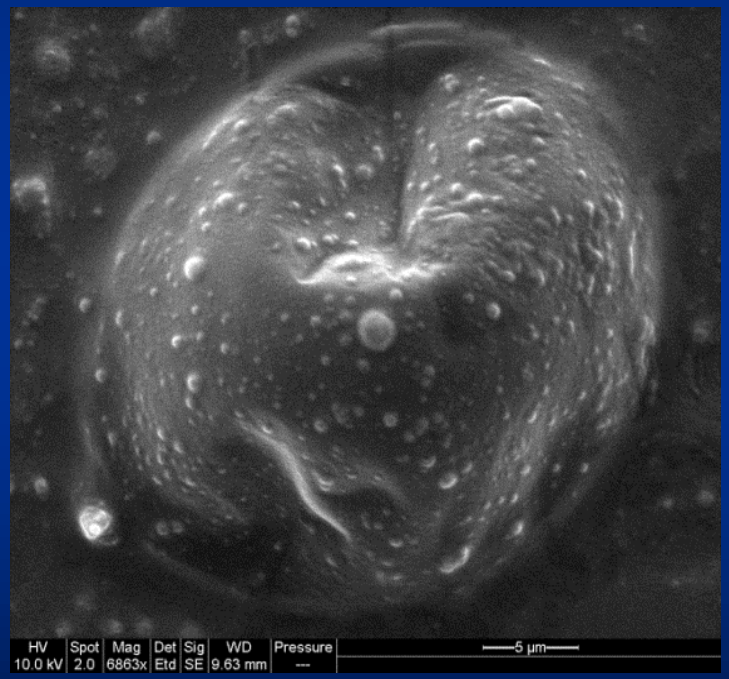


Functional Properties of Microspheres

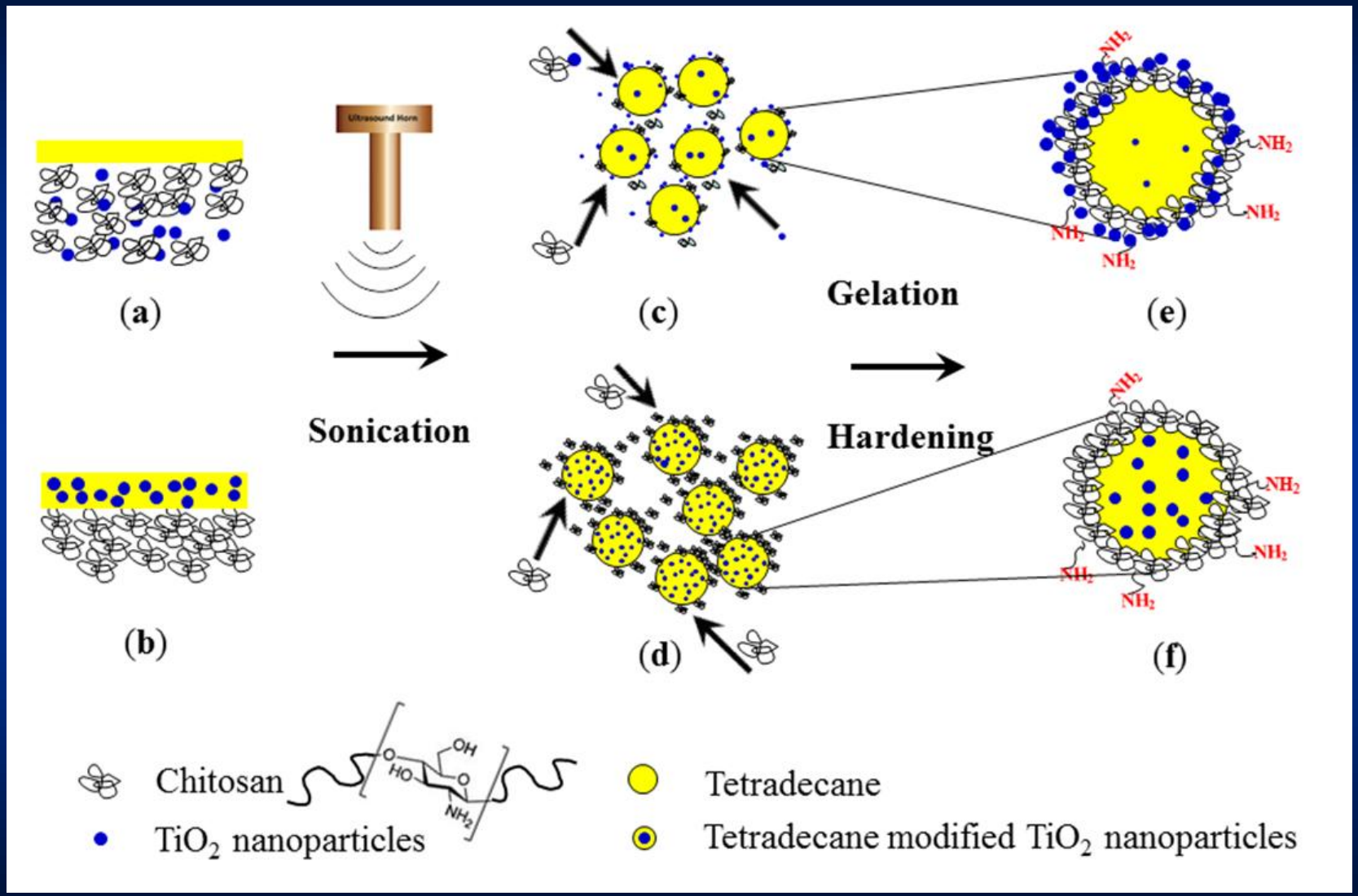




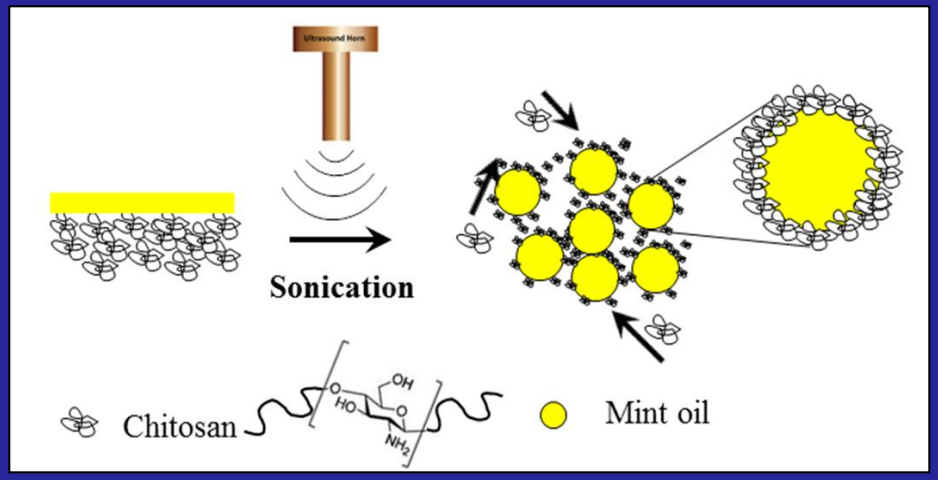


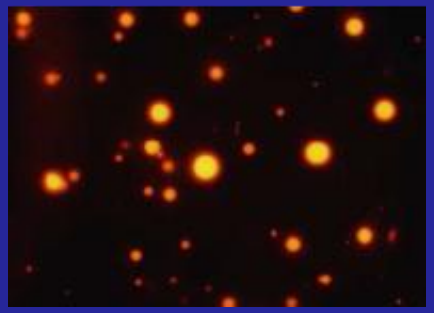


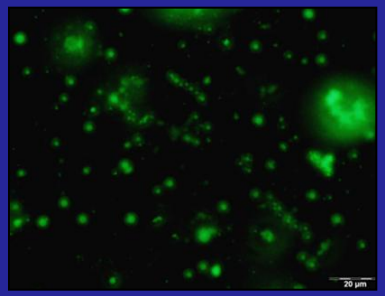
Functional Properties of Microspheres

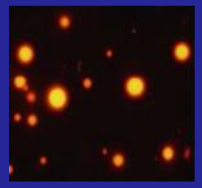


RSC Adv, 2015, 5, 20265



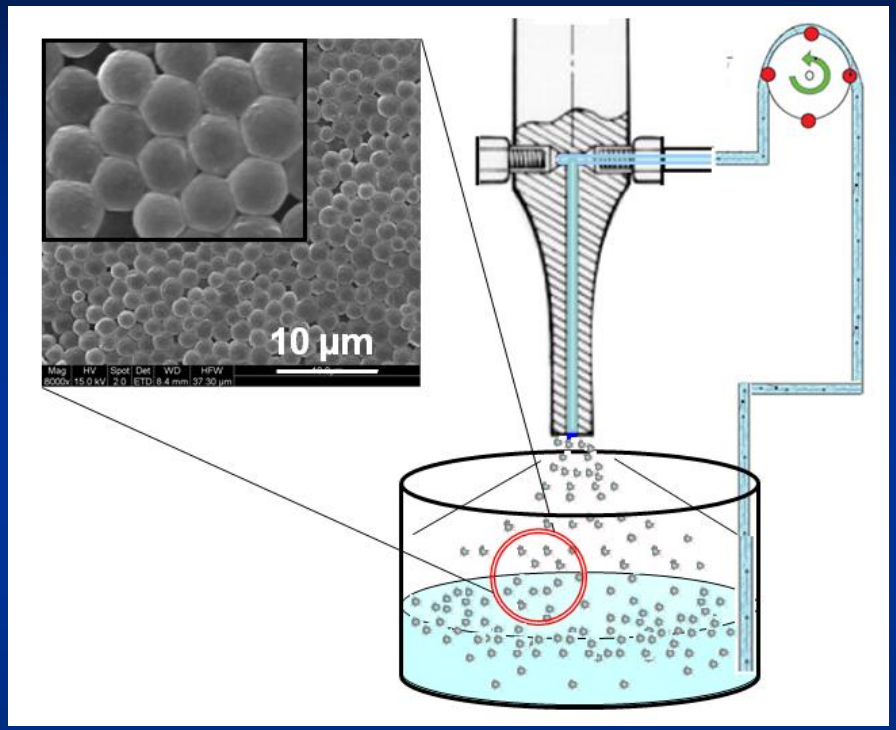


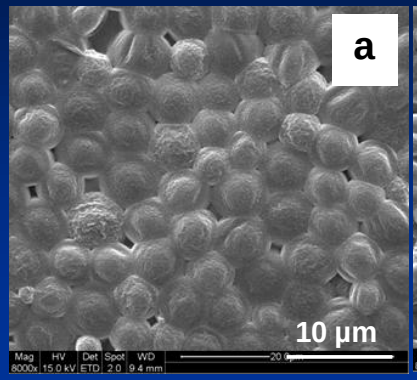


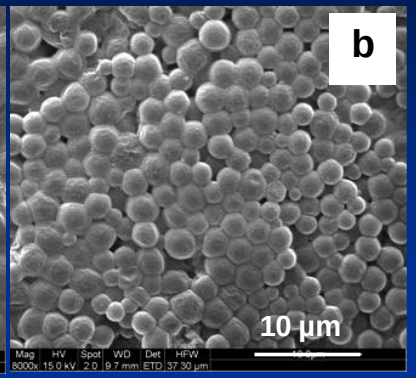


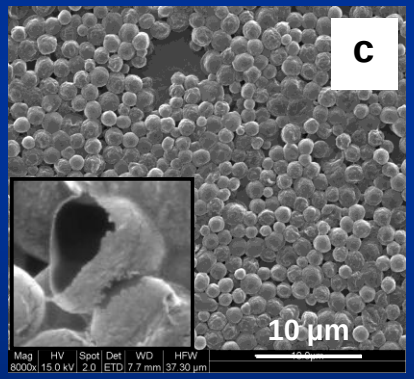
Cocoa Butter

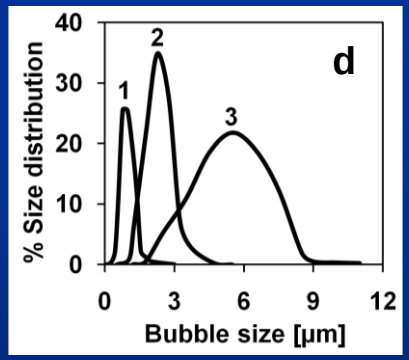
Nano- and Microbubbles

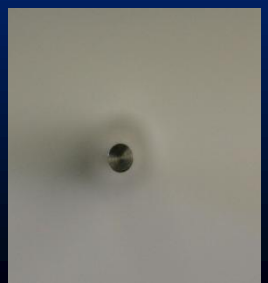




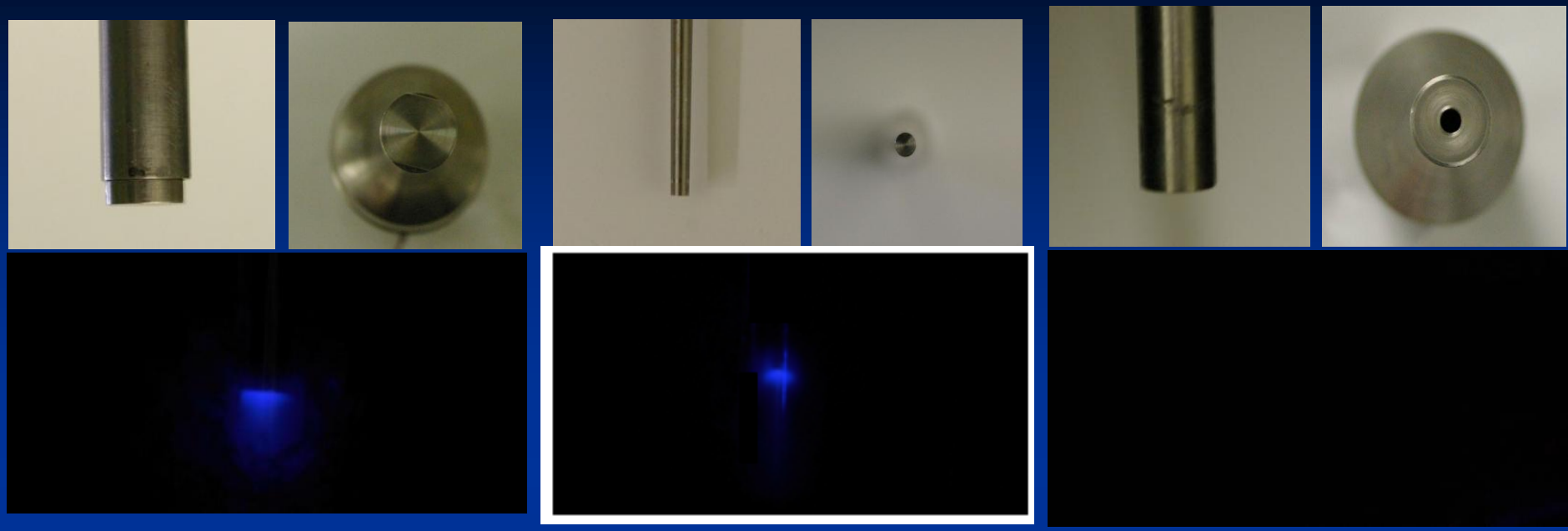


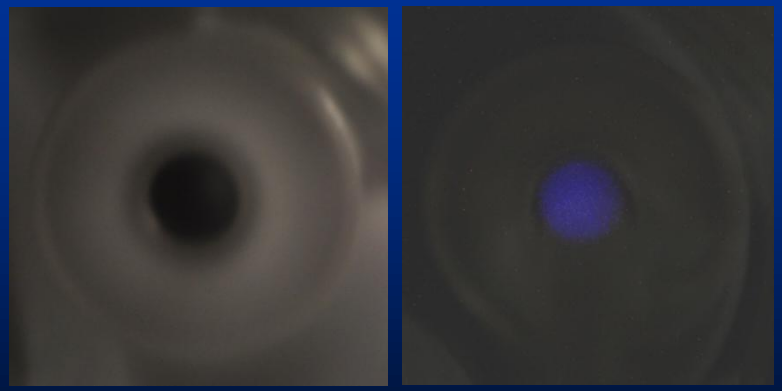




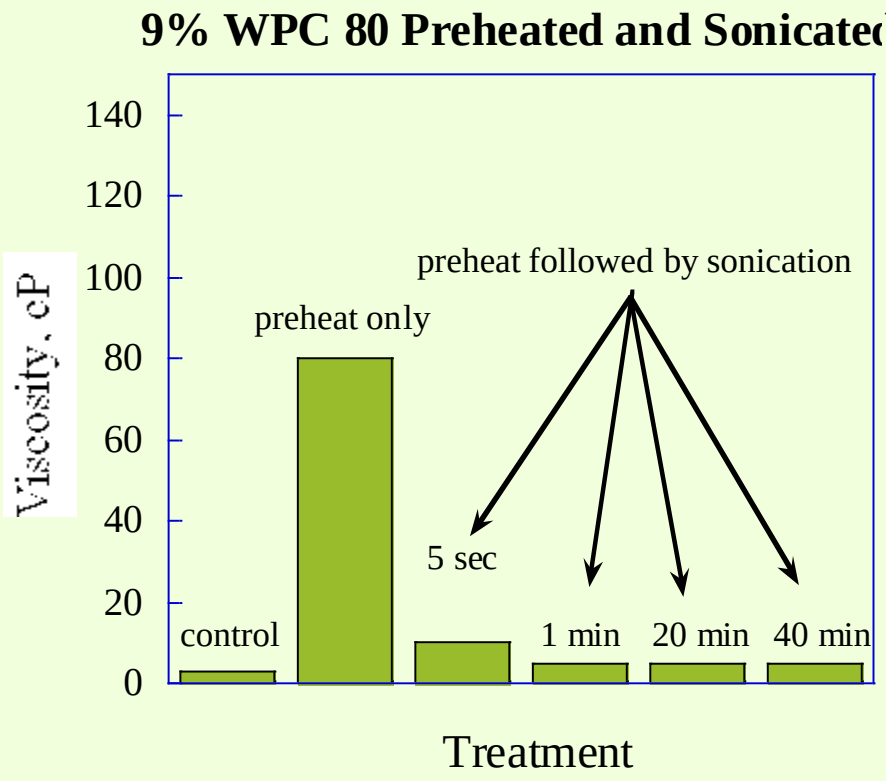


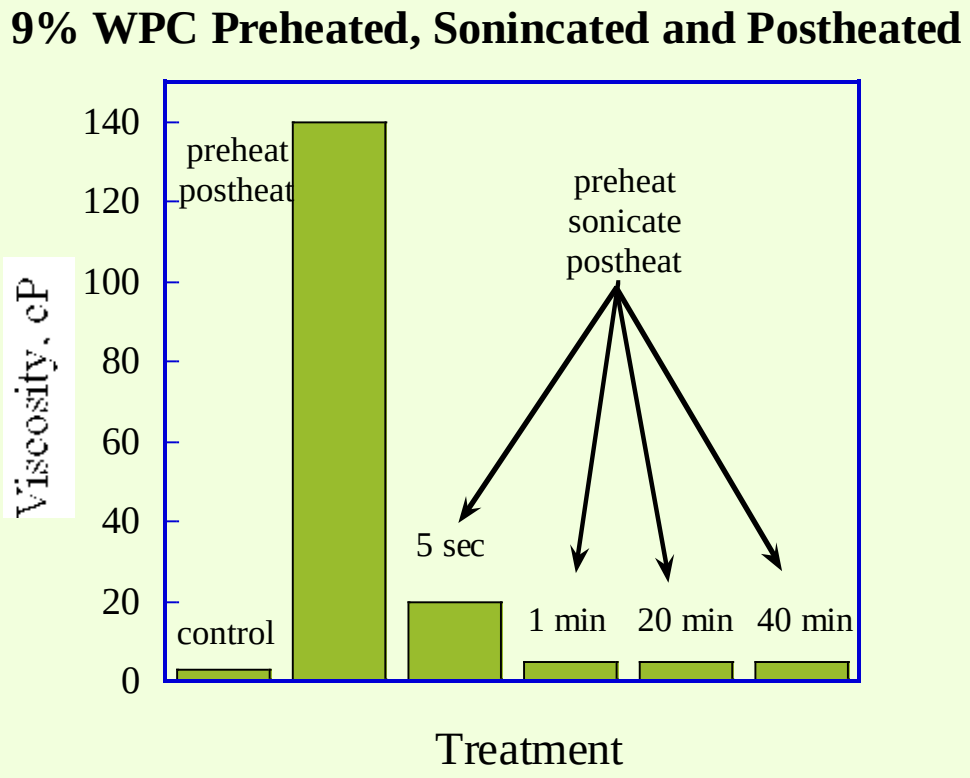
Nano- and Microbubbles





Whey Protein Concentrate
Heat Stability and Viscosity Reduction





Ashokkumar et al., J. Dairy Sci., 2009, 92, 5353

Dairy Systems

Composition of milk

Components	%	Constituents
Water	>80	
Proteins	~5%	80% Caseins (α, β, γ, κ) 20% Whey Proteins (α-lactalbumin, β-lactoglobulin – 1:3)
Carbohydrates	~5%	Lactose
Fat	~5%	
Minerals	~1%	Ca phosphate (colloidal and dissolved)

Systems Investigated

- Three broad categories
  - Whey Protein Systems
  - Casein Containing Systems
  - Fat Containing Systems

Whey Protein Systems

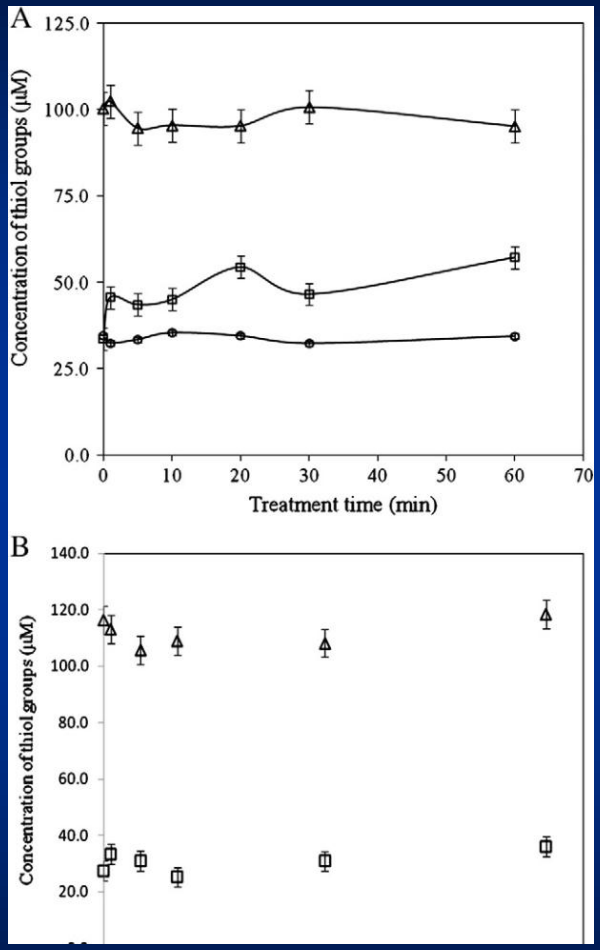
- Whey Protein Concentrate
- Whey Protein Isolate
- Pure α-LA
- Pure β-LG
- Pure protein mixtures (1:3)

Techniques

- Differential Scanning Calorimetry - DSC
- HPLC
- Circular Dichroism – CD
- Thiol group - Ellmans Assay
- Hydrophobicity of the proteins – Fluorescence Spectroscopy

Structural Changes of Sonicated Pure Protein Systems

Reactive Thiol Groups

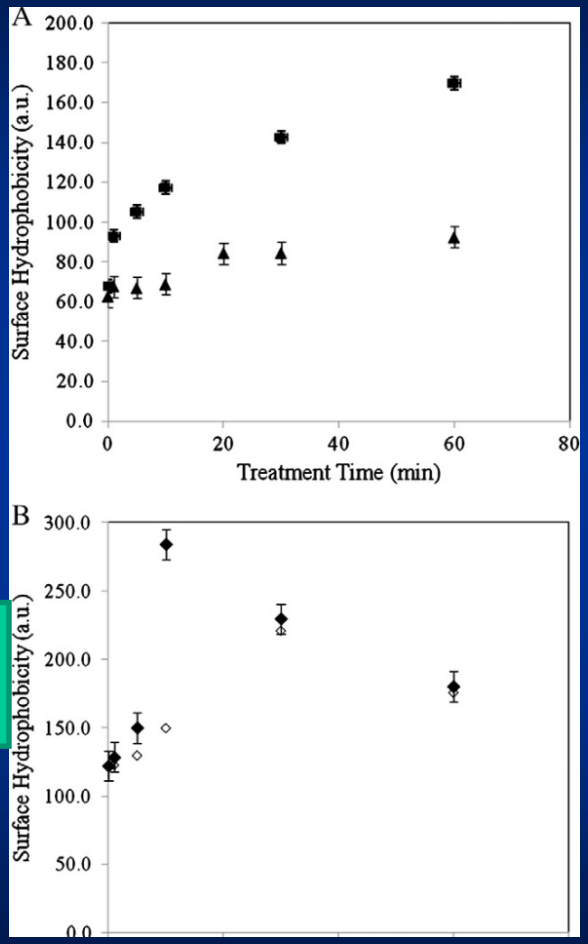


(A)Pure β-LG
(B) 3:1 β-LG/α-LA protein mixture
Reactive thiol groups (□), Total thiol groups (△)

- β-LG - increased by ~36% after 1 min US
- Continuous increase up to 60 min
- 3:1 β-LG/α-LA Protein Mixture – Slight increase up to 60 min

- Pure β-LG - no changes up to 20 min, then increased between 20 and 60 min.
- Pure α-LA - Significantly increased up to 60 min
- 3:1 β-LG/α-LA protein mixture - an increase up to 10 min US, followed by a decrease

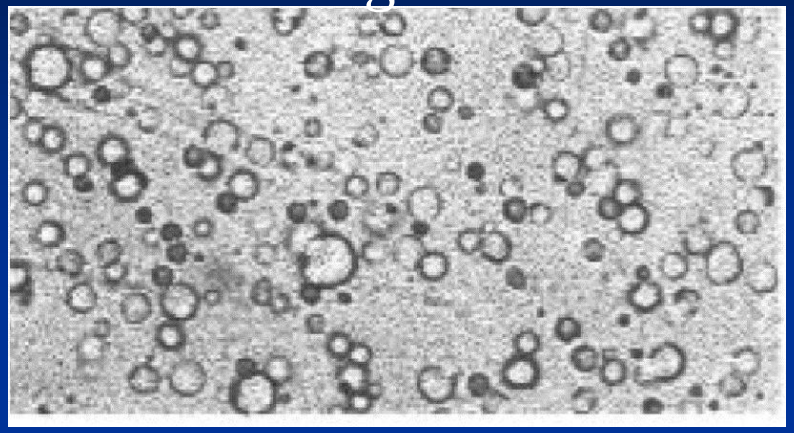
Surface Hydrophobicity



(A) β-LG (▲) , α-LA (■)
(B) 3:1 β-LG/α-LA mixture

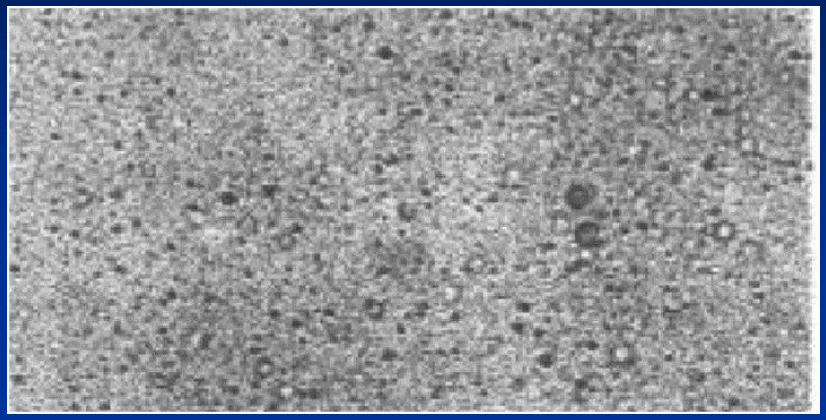
Ultrasound in Dairy Science
Homogenisation of milk fat

Non-homogenised milk



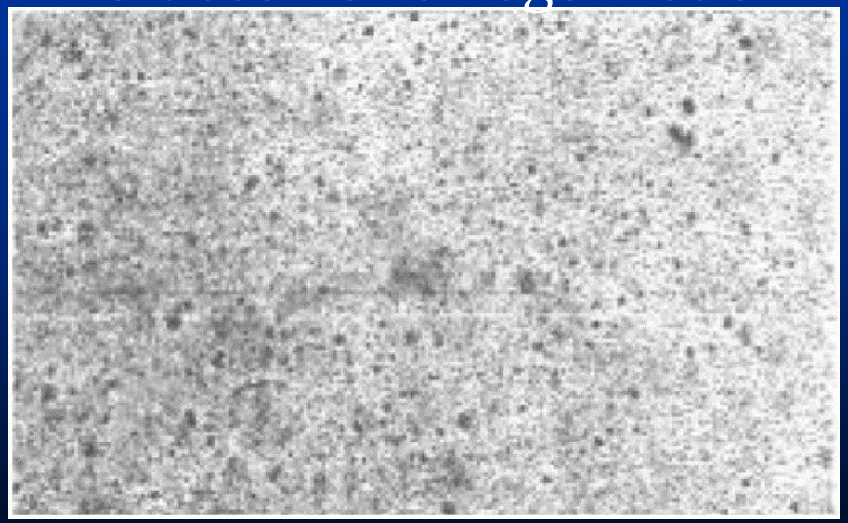
4-7 μm

Conventional homogenization



< 2 μm

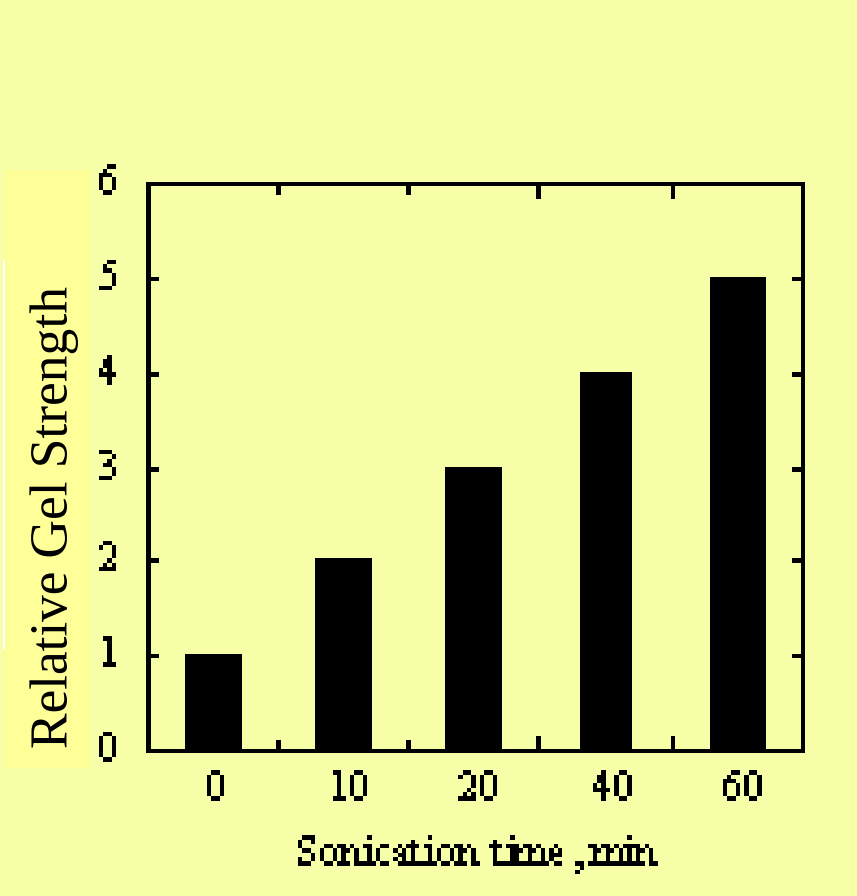
Ultrasonic homogenization

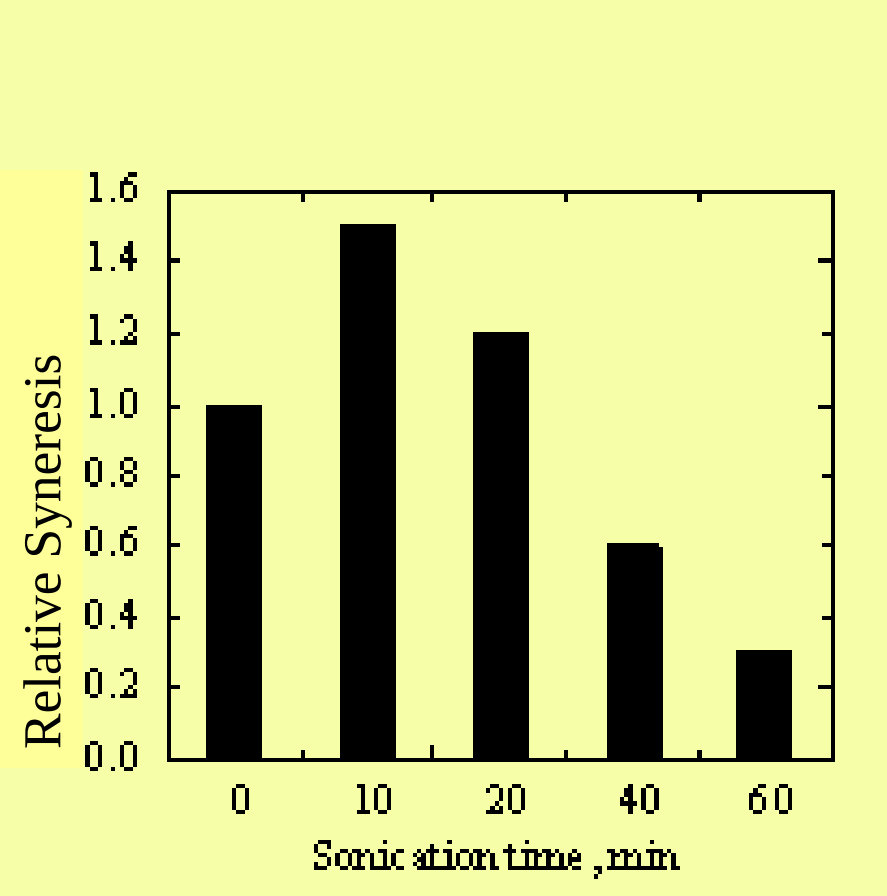


< 1 μm

Ertugay et al., Turk. J. Vet. Anim. Sci., 28, 303, 2004

Whey Protein Concentrate
Gel Strength and Syneresis

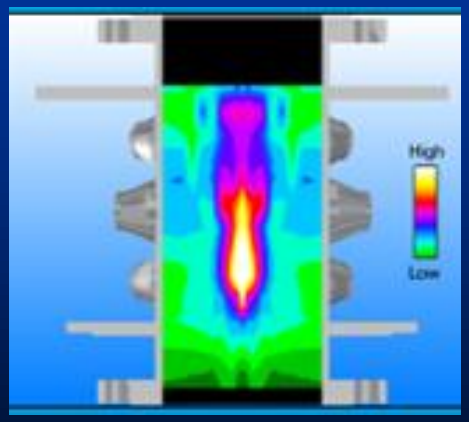




Zisu et al., J. Diary Research, 2011, 78, 226

Direct Contact Low Frequency Power Ultrasound

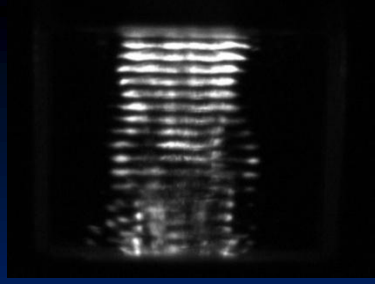
Non-Contact Low Frequency Power Ultrasound



Separation

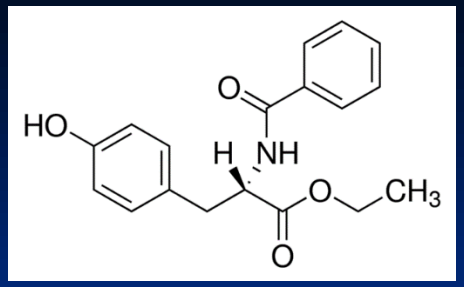
Courtesy of Dr. Kozuka, AIST, Nagoya, Japan

Separation

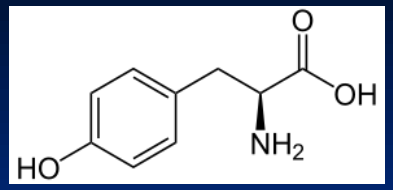


Juliano et al., Ultrasonics Sonochemistry, 2011, 18, 963

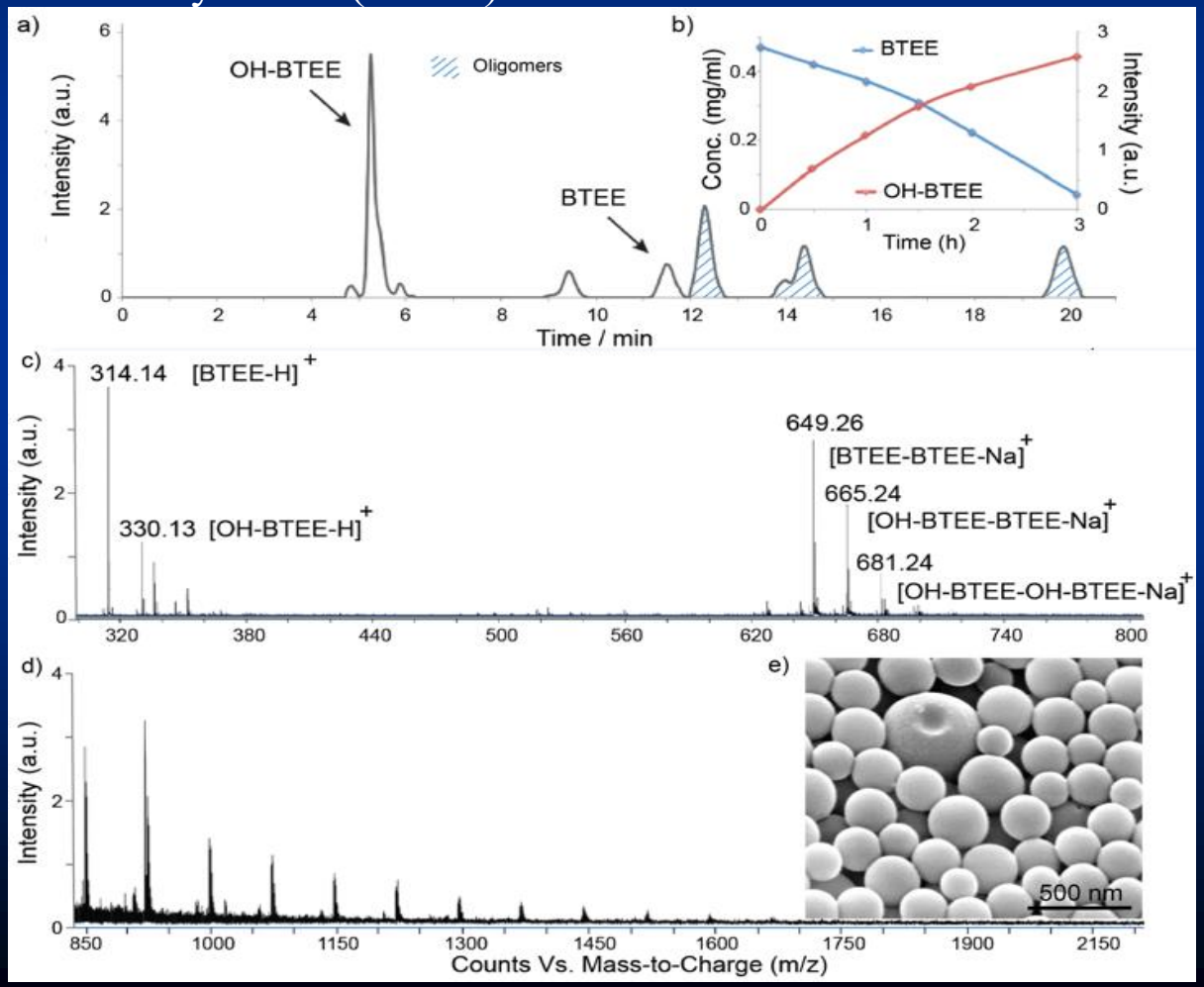
Sonoassembly of oligophenol nanostructures



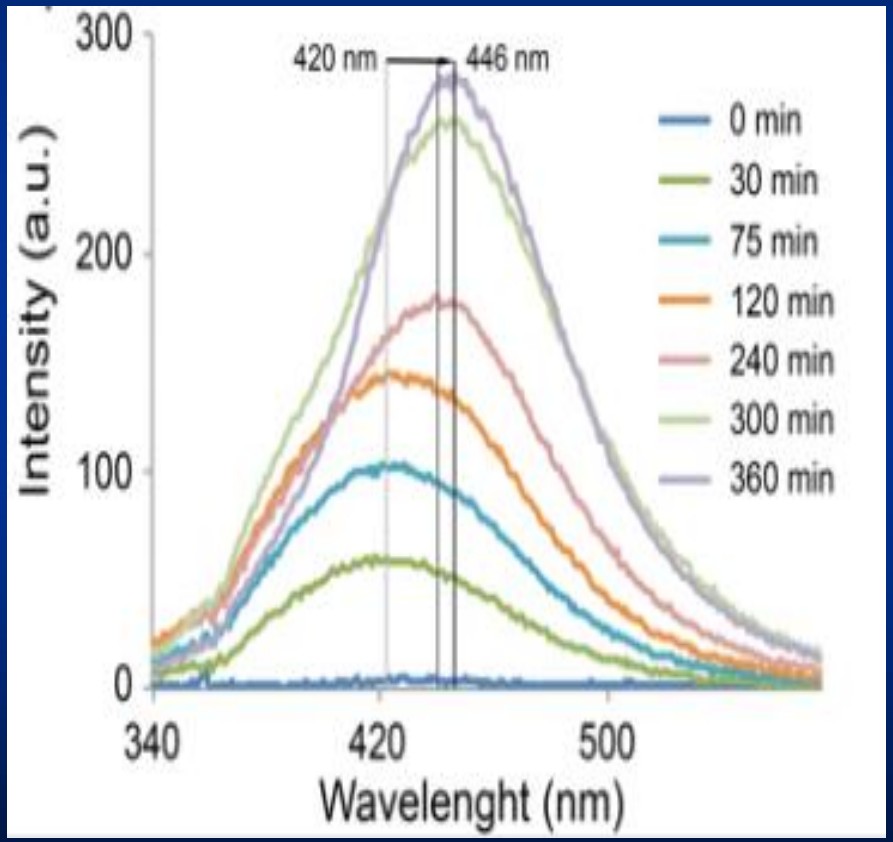
N-Benzoyl-L-Tyrosine Ethyl Ester (BTTE)

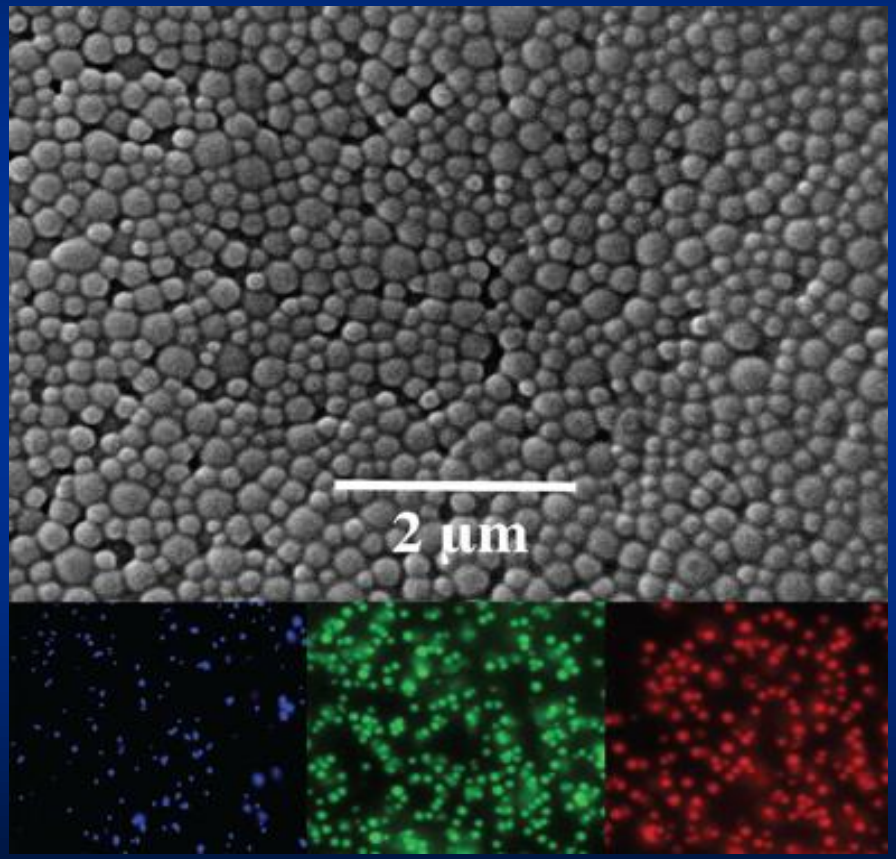


L-Tyrosine

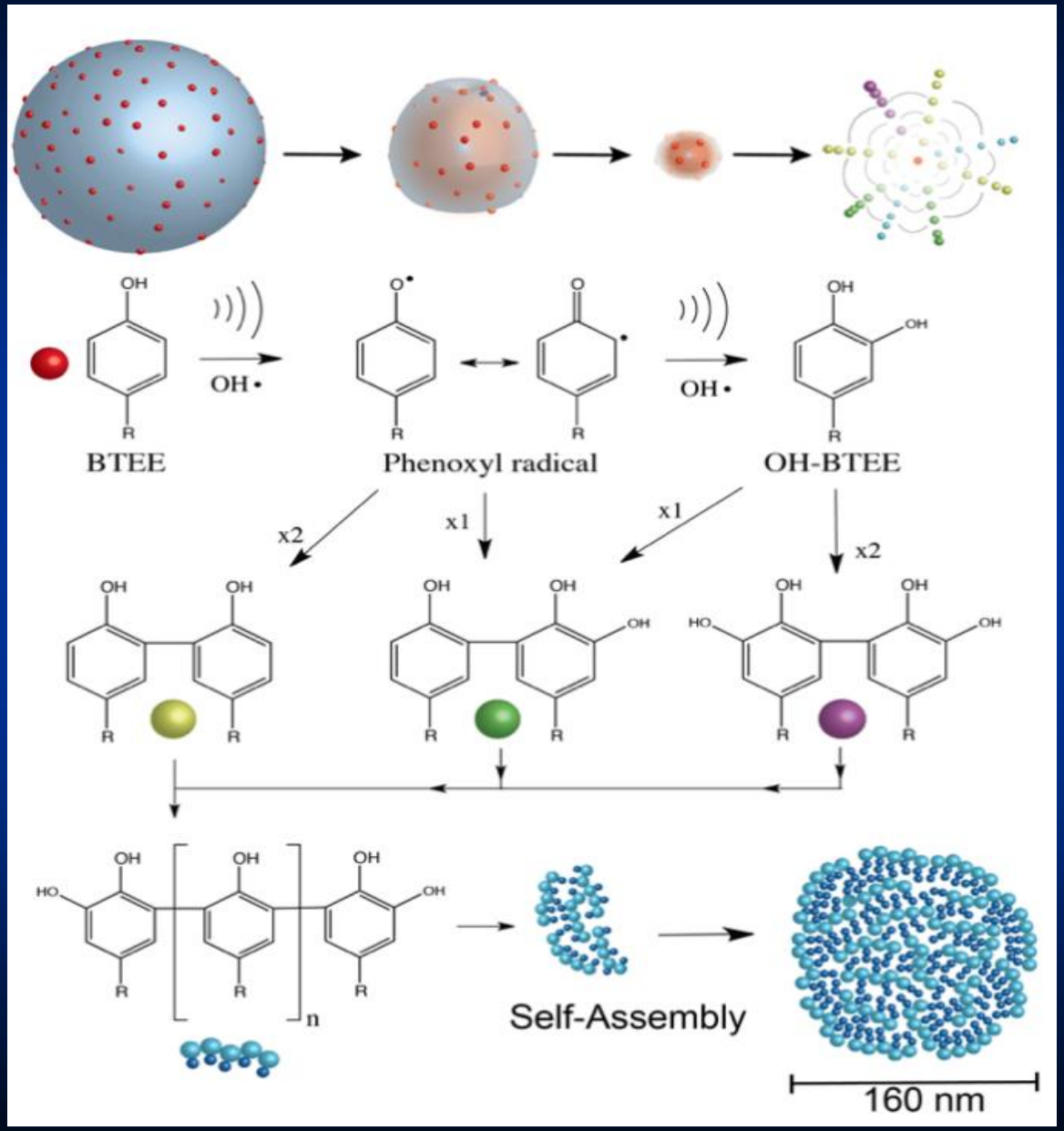


Sonoassembly of oligophenol nanostructures





Sonoassembly of oligophenol nanostructures



Summary

- Acoustic cavitation – a complex process
- The physical and chemical effects could be used for materials synthesis
- Biomedical and various applications

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Many visiting academics and students

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