Recent Advances in nanotechnology of food materials for food and non-food applications

JOZEF L. KOKINI

PURDUE UNIVERSITY

DEPARTMENT OF FOOD SCIENCE

PURDUE
UNIVERSITY

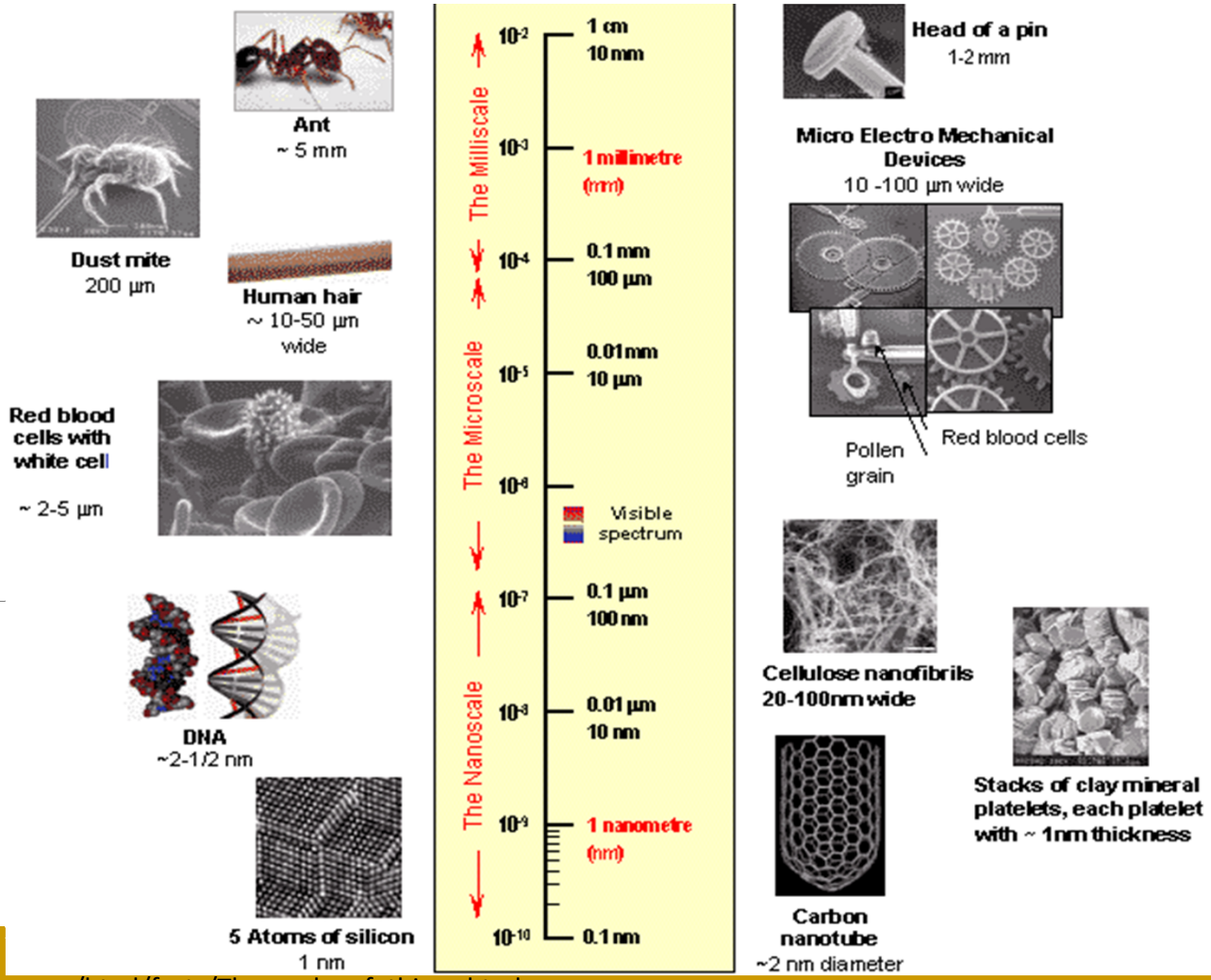
PURDUE UNIVERSITY

What is Nanotechnology?

Nanotechnology – the ability to work at the atomic, molecular, and supramolecular levels in the length scale of 1-100 nm range, in order to create materials, devices, and systems with fundamentally new properties and functions because of their small structure.

(*www.nano.gov)*

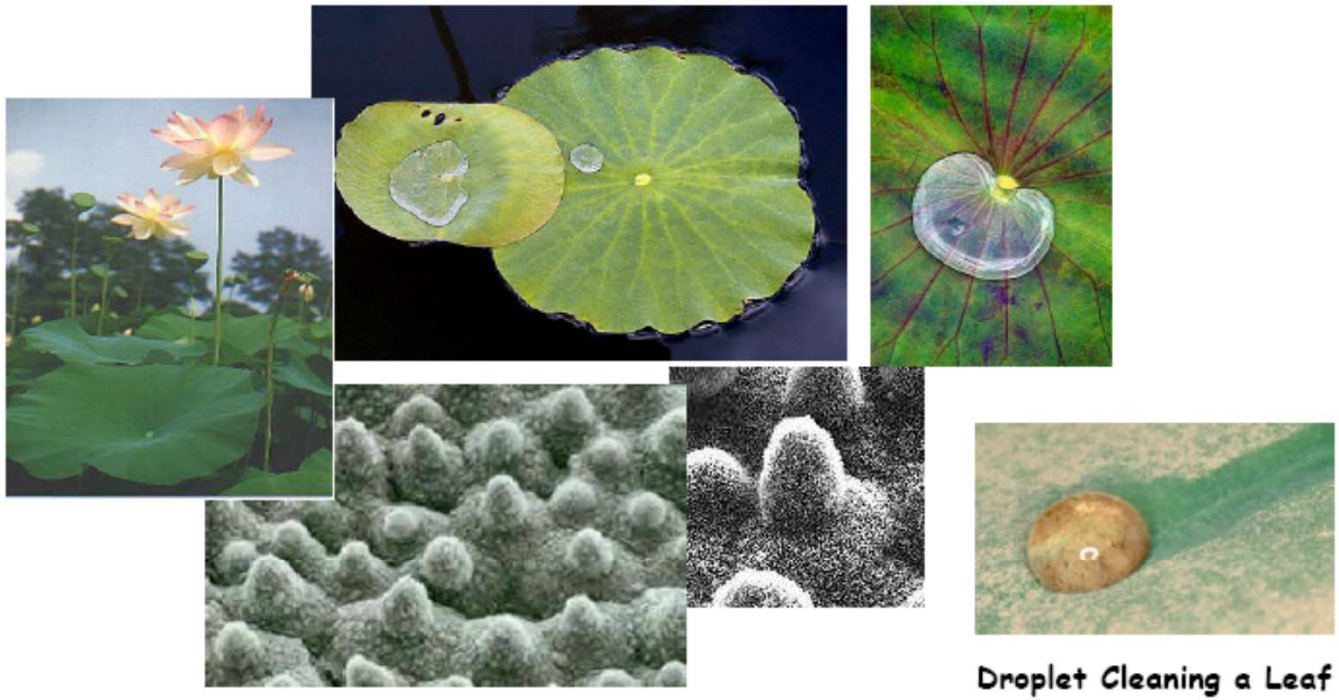
The scale of Things-Nanometers and More



http://nano.gov/html/facts/The_scale_of_things.html

The Lotus Leaf Effect

surface morphology + chemistry to control fluid interactions



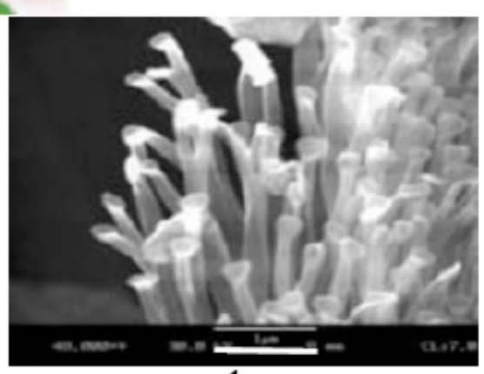
Lotus leaf

Droplet Cleaning a Leaf

Through the combination of micro- (cells) and nano-structure (wax crystals) contact areas are minimized. Any hydrophilic contamination on the leaf adheres to the water rather than the leaf itself and rolls away with the droplet.

Geckos use morphology to control surface energy for climbing **Velcro effect**

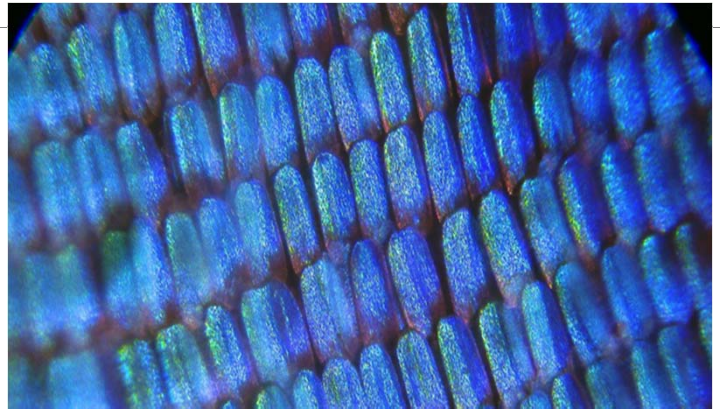
http://www.lclark.edu/~autumn/



1 μm

SEM of keratin hairs covering gecko feet

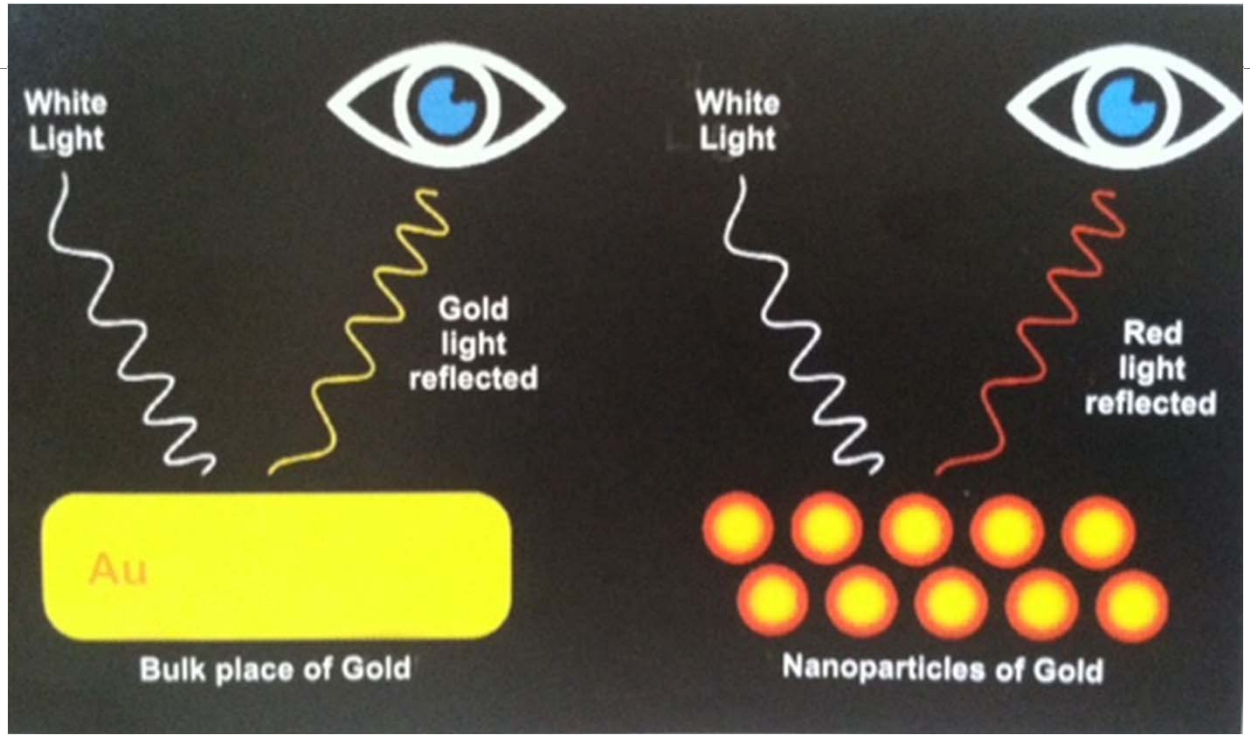
Butterfly wing scale photonics



The structures responsible for producing intense color are precise to the nanoscale, with repeating patterns of cuticle and air space.

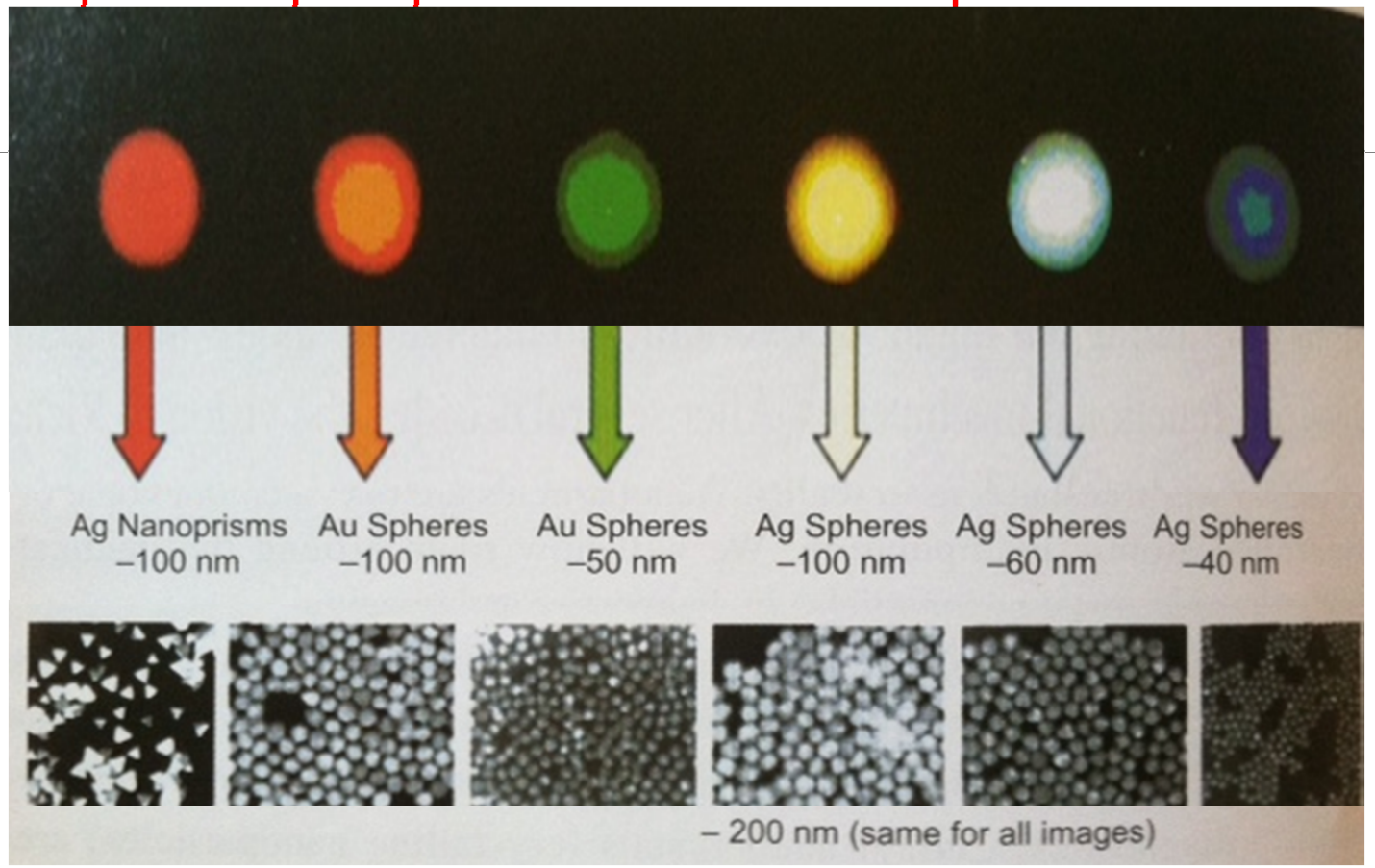
PURDUE UNIVERSITY

Light reflection by gold nano particles



Light reflections by bulk and nano gold particles

Optical properties of nanoparticles



Optical properties of the nanoparticles can be changed with different size, shapes and composition of metal nanoparticle.

Bio-Nanotechnology: Concepts & Applications , Sharon M., Sharon M., Padey S. and Oza G., CRC Press ,USA

Introduction

Food nanotechnology focuses on the fabrication of structures at the nanometer scale with unique properties that can be used for various food applications such as

- delivery systems
- improved food contact surfaces with unique

surface properties

- food characterization tools
- microfluidic devices
- sensors
- nanocomposite films

Biocompatible spherical nanoparticles and nanotubes

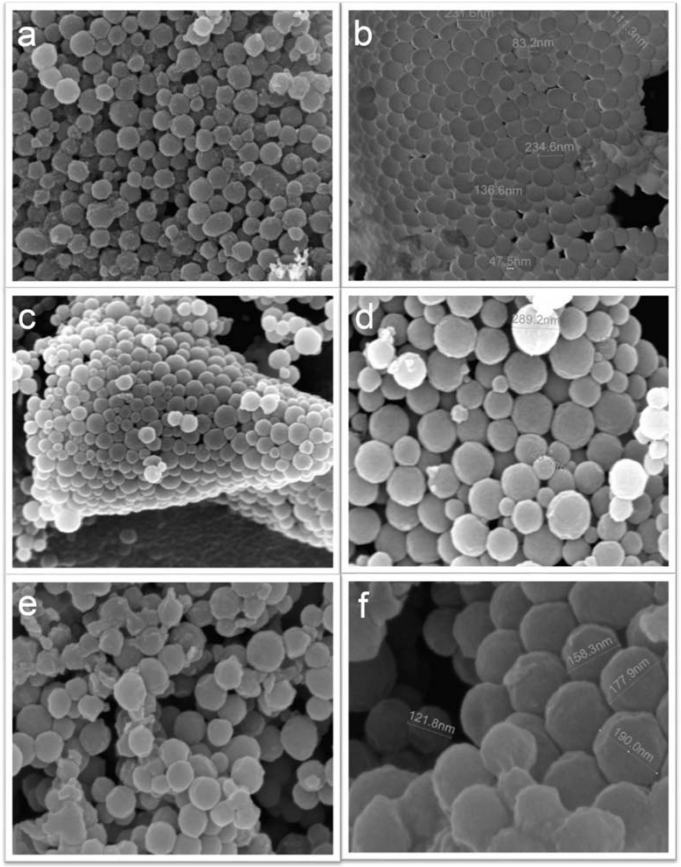
- Biocompatible nanoparticles from natural biopolymers (carbohydrates and proteins)
- protein and carbohydrate polyelectrolytes are a desirable choice
- nanoparticles with unique physical, chemical, and biological characteristics can be made.
- Methods for the preparation of bio-nanoparticles :
  - Desolvation
  - Coacervation
  - Emulsification

Nanoparticulation of α-lactalbumin (AL)

- The effect of different desolvating agents including **ethanol, methanol, and acetone,** and the **desolvating agent to protein solution ratios on the particle size, polydispersity index, and zeta potential** of AL NPs was also studied (Abdunnasir et al., 2016)

Properties of AL nanoparticles

- The **smallest nanoparticles** are sub 200 nm in size and are produced by methanol.
- In all cases, the polydispersity index points to great uniformity approaching **monodispersity.**
- The zeta potential of all the nanoparticles was higher than -30 mv at pH around 7 showing **excellent stability** in these particles.
- Ethanol and acetone prepared nanoparticles were around 200-230 nm suitable for use in food and drug formulations.
- Perfectly spherical NPs can be prepared from AL with ethanol as the desolvating agent (Abdunnasir et al., 2016).



The SEM images of α-LA nanoparticles (a, b- prepared with ethanol; c, d- prepared with acetone; e, f- prepared with methanol; all in 1:5 water: desolvating agent ratio) (Abdunnasir, 2016).

Nanoparticulation of Bovine Serum Albumin (BSA)

- Ethanol, acetone, and the mixtures of ethanol and acetone as desolvating agents
- Ethanol best desolvating agent based on the uniformity and size of manufactured nanoparticles.
- Ethanol to water in the ratio of 4:1, produced highly uniform and small nanoparticles (~90-140 nm, PDI=0.045).
- SEM images confirm spherical and uniform nanoparticles.
- The mixture of ethanol and acetone produced spherical nanoparticles; two groups of nanoparticles with different sizes (SEM pictures and DLS data).

Ovalbumin nanoparticles

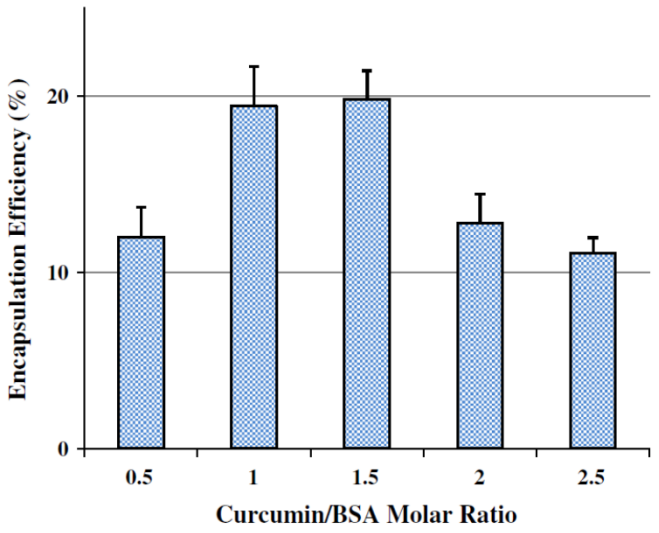
- OVA nanoparticles prepared using desolvation method and centrifugation to remove large aggregates. Abdunnasir et al., (2016)
- sub-100nm OVA nanoparticles
- Both ethanol and methanol produce NPs in the range of 60-80 nm, and the smallest nanoparticles are prepared by using methanol as the desolvating agent.

DA[a]	W:D[b]	PS[c] (nm)	PDI[d]	ZP[e] (mV)
Ethanol	1:3	69.2±7.0	0.259±0.119	-22.3±3.4
Ethanol	1:4	76.6±17.6	0.235±0.067	-23.2±1.8
Ethanol	1:5	80.3±2.8	0.231±0.010	-24.8±4.3
Ethanol	1:10	73.8±4.3	0.247±0.016	-23.3±5.2
Ethanol	1:20	75.7±4.0	0.239±0.029	-19.1±4.7
Acetone	1:3	71.6±9.6	0.259±0.001	-17.5±4.8
Acetone	1:4	84.8±7.5	0.183±0.002	-21.5±8.0
Acetone	1:5	96.8±12.2	0.196±0.034	-22.6±5.9
Acetone	1:10	136.7±7.1	0.125±0.001	-21.0±7.8
Acetone	1:20	160.7±5.2	0.127±0.001	-18.2±4.3
Methanol	1:3	66.4±30.1	0.381±0.242	-14.8±7.3
Methanol	1:4	75.9±10.1	0.285±0.039	-18.9±3.0
Methanol	1:5	60.5±7.8	0.224±0.004	-15.8±7.1
Methanol	1:10	72.8±15.0	0.313±0.021	-18.0±8.8
Methanol	1:20	61.2±5.4	0.246±0.055	-18.8±7.6

Desolvating agent type, [b]water: desolvating agent volume ratio, [c]nanoparticle particle size, [d]polydispersity index, [e]nanoparticle zeta potential. Each data point represents the average from three replicates.

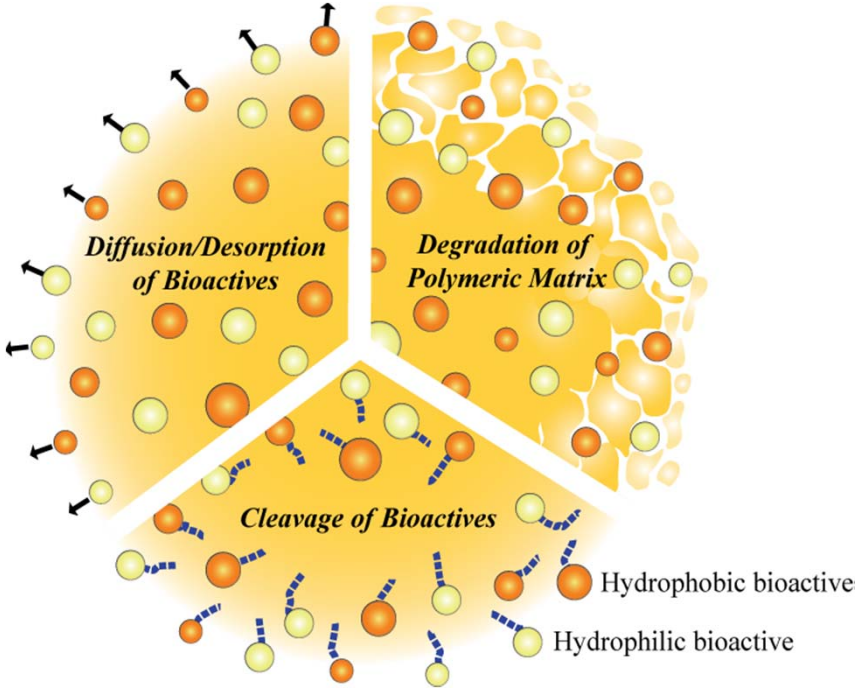
Curcumin loaded BSA nanoparticles

- Curcumin loaded BSA nanoparticles were prepared by the desolvation method (Sadeghi et al., 2014).
- Different molar ratios of curcumin were added to BSA aqueous solutions and ethanol was used as the desolvating agent.
- After particulation freeze drying was done to prepare curcumin loaded BSA NPs as a powder. In this powder form, curcumin was attached to the surface of BSA nanoparticles or encapsulated in them.
- Both free and curcumin loaded BSA NPs were stable after glutaraldehyde treatment.
- Re-dispersibility study showed that the particle size was higher for redispersion of freeze dried sample.
- Applying sonication dramatically improved redispersibility of the curcumin loaded NPs (Sadeghi et al., 2014).



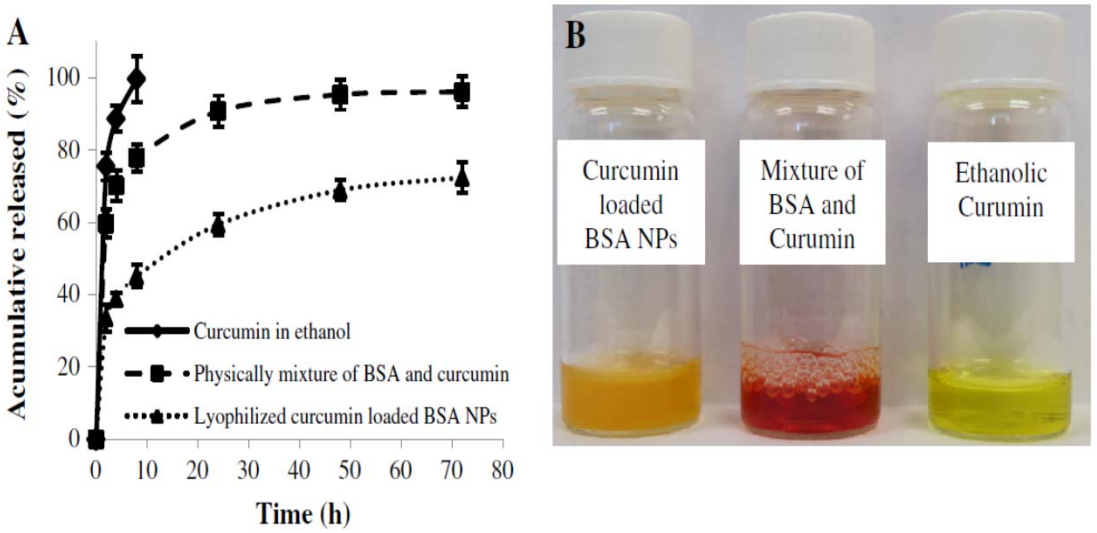
Encapsulation efficiency of curcumin-loaded BSA NPs with the use of ethanol as a desolvating agent (Sadeghi et al. 2014).

Three possible release mechanisms for entrapped bioactive compounds



The types of interaction between bioactive components and nanoparticles play an important role in release profiles.

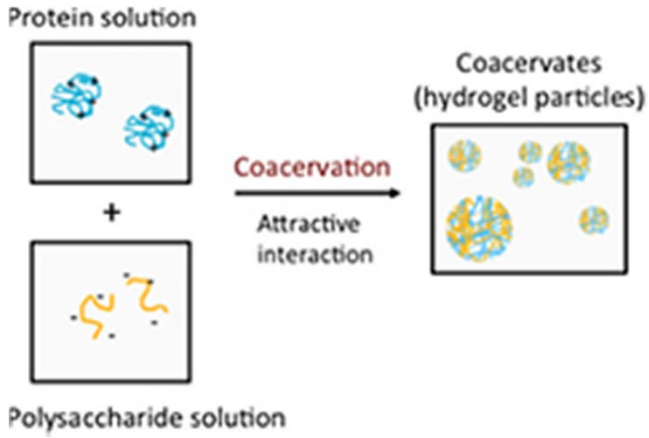
In vitro release profile of curcumin from curcumin loaded BSA NPs showed three different regions: fast release, gradual release and strongly bound part after 72 hours.



In vitro curcumin release profiles (A), and photograph of solutions (B), curcumin and BSA concentrations were the same (Sadeghi et al. 2014).

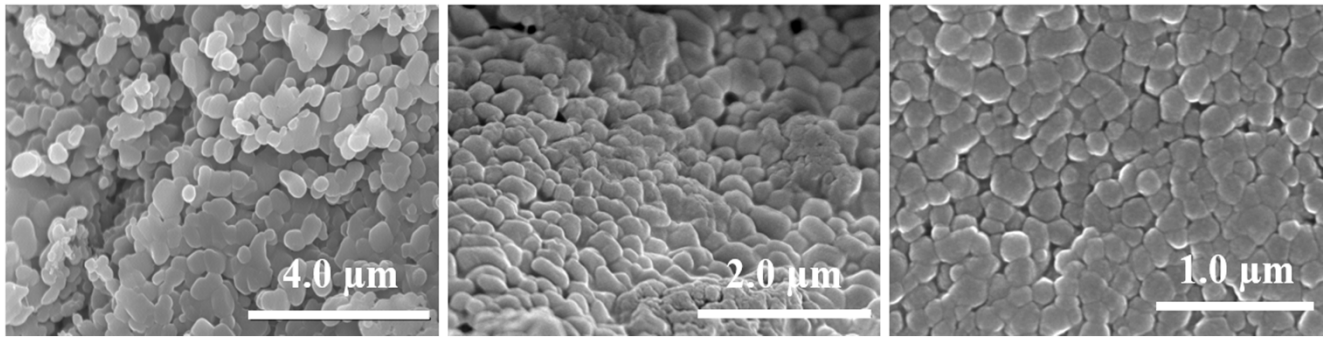
Complex Coacervation

- Two or more oppositely charged biopolymers form a Polyelectrolyte complex (PEC) to prepare nanoparticles.
- in a mixture of protein and polysaccharide, three equilibrium processes can exist including miscibility, thermodynamic incompatibility and complex coacervation.
- Control of concentration, ratio of the polymers, pH ionic strength, etc., during interaction of proteins and polysaccharides is important for successful nanoparticulation through complex coacervation.



Coacervation Method

- Oppositely charged polypeptide (protein)-protein coacervate nanoparticles.
- Poly-D-lysine, a positively charged polypeptide and BSA a negatively charged protein resulted in nanoparticles in the range of 200-500 nm in diameter (Maldonado, Sadeghi, Kokini, 2017).

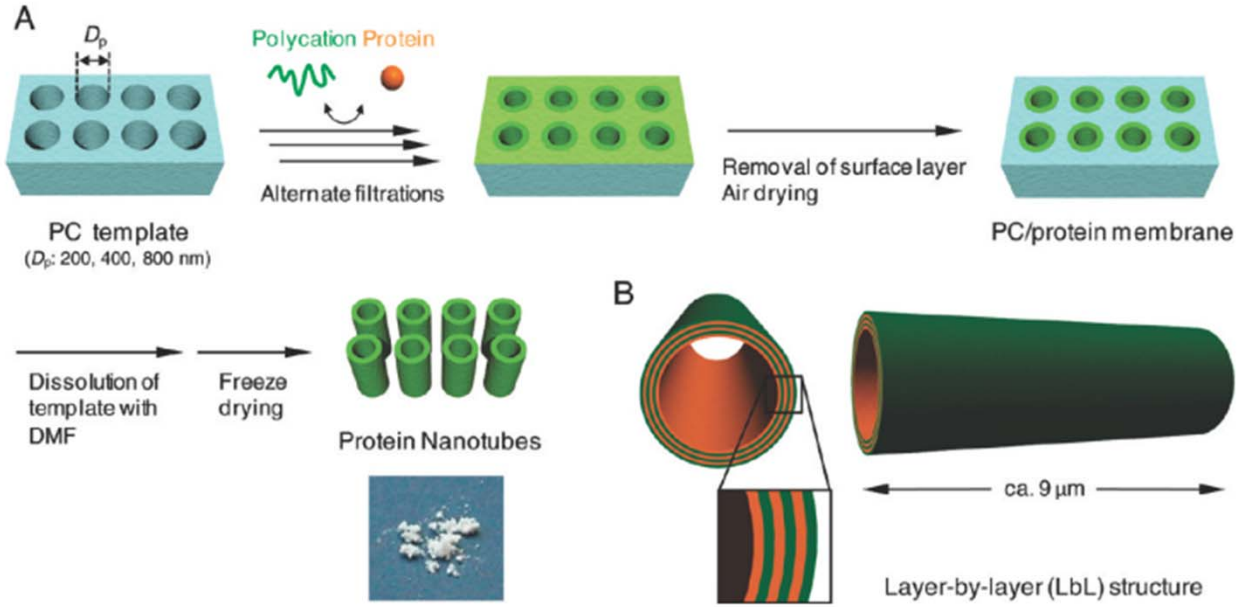


SEM images of complex-coacervation nanoparticles from PDL and BSA (Maldonado, Sadeghi, Kokini, 2017).

Biocompatible nanotubes (BNTs)

- Several advantages when compared to other nanostructures including different interior and exterior surface properties, two open ends, high loading capacity, and good stability (Sadeghi et al., 2013).
- They are fabricated from biopolymers (preferably GRAS) and may be safer than inorganic nanotubes.
- The layer-by-layer deposition method using templates with nanopores is one of the best methods to manufacture bionanotubes.
- Electrostatic interactions, hydrogen bonds, and covalent bonding are the possible mechanisms to deposit different layers of biopolymers inside template pores. Electrostatic interaction is the common binding force for layer-by-layer deposition when charged biopolymers are used.
- Surface charges of biopolymers can be controlled by solution properties such as pH and ionic strength.

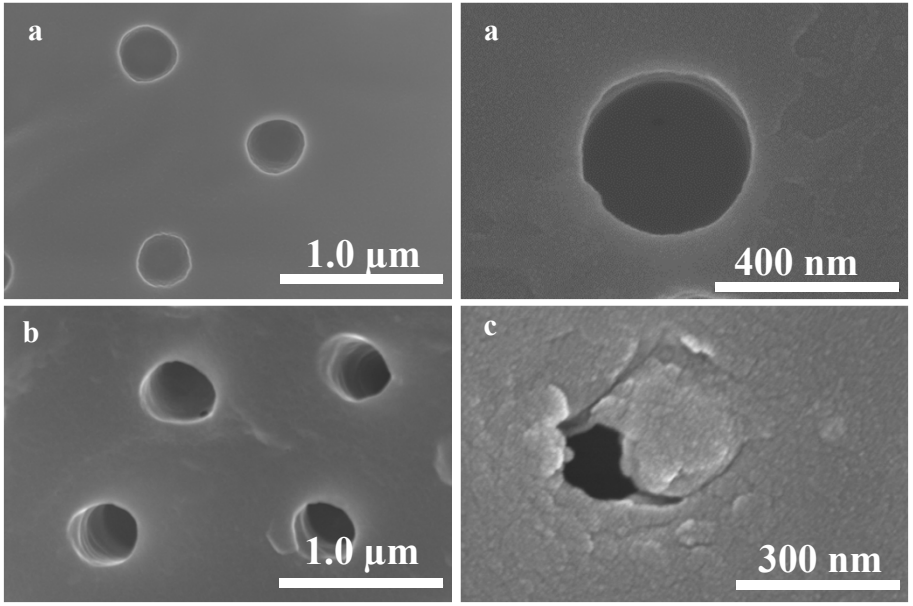
Designing bio-based nanotubes
Layer-by-Layer Deposition, LbL



Synthesis of protein nanotubes by LbL deposition technique using nanoporous PC membrane, Qu and Komatsu, 2010.

Designing bio-based nanotubes

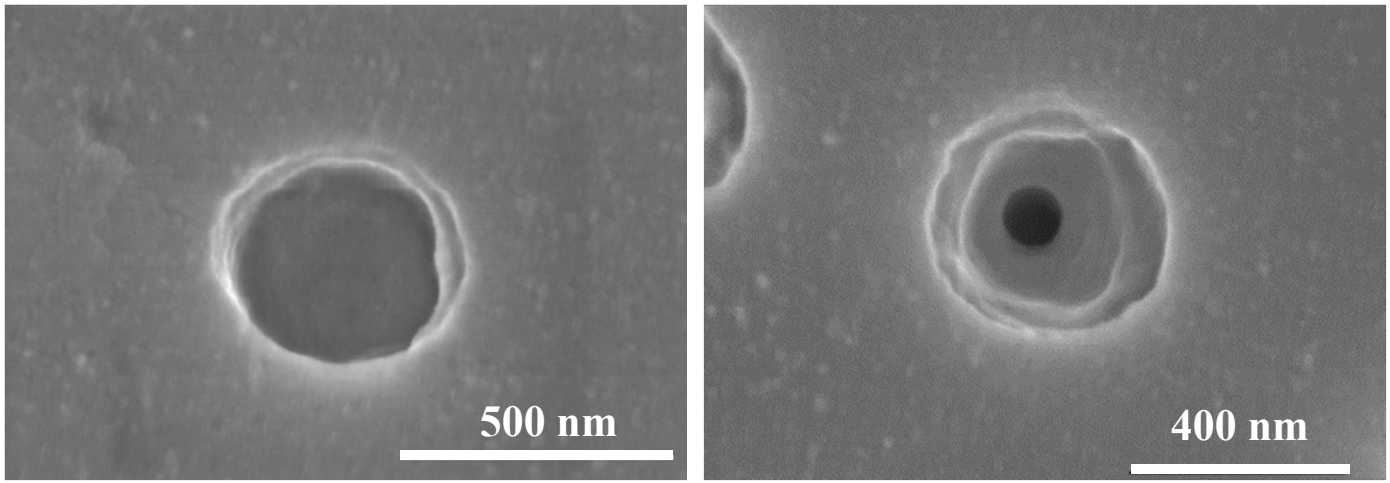
Layer-by-Layer Deposition, LbL



SEM images of one (a), two (b), and three (c) bilayers of PDL (poly-D-Lysine) and BSA (Bovine Serum Albumin) on the surface of PC template, before cleaning

Designing bio-based nanotubes

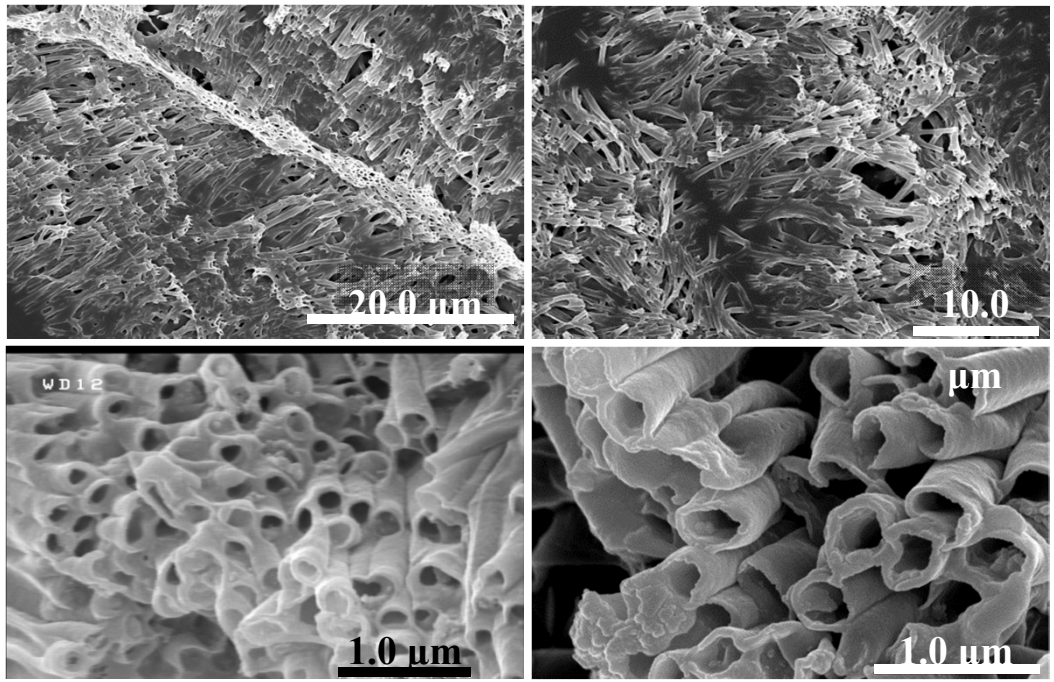
Layer-by-Layer Deposition, LbL



SEM images of deposition of three bilayers of PDL and BSA on the surface of PC template after surface cleaning

Designing bio-based nanotubes

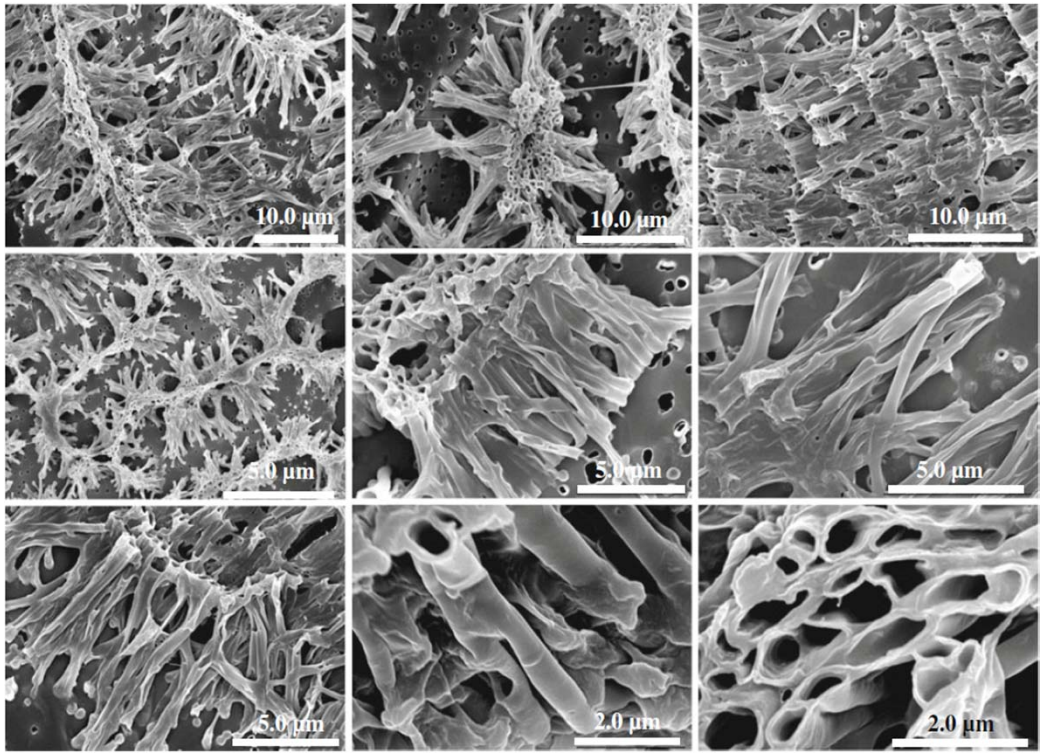
Layer-by-Layer Deposition, LbL



SEM images of $(PDL/BSA)_3$ bio-nanotubes

PURDUE UNIVERSITY

Deposition of 3 bilayers of poly-D-lysine (PDL) and bovine serum albumin (BSA)

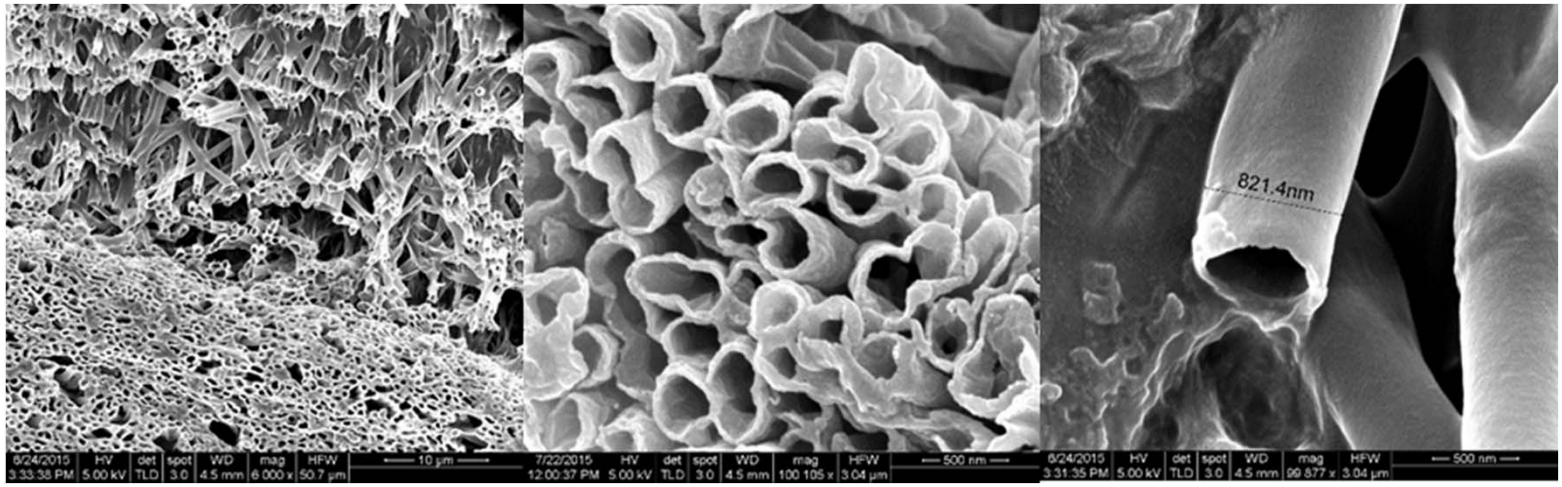


- The structure of BNTs fabricated from one, two and three bilayers of PDL/BSA inside a polycarbonate template with pore size of 400 nm.
- The SEM images of BNTs which produced by deposition of one BL (a–c), two BL (d–f) and three BL (g–l) (Sadeghi et al., 2013).

Loading capacity of BNTs

- Curcumin was used to evaluate the loading capacity of BNTs
- If the interior layer is made from BSA, which has high affinity to curcumin, the concentration of free curcumin increases in a dispersion of BNTs (Sadeghi et al., 2013).
- $(PDL/BSA)_3$ nanotubes, have a free-loading capacity of 270 µg curcumin per 1 mg BNTs. The manner in which nanotubes are designed leads to loading of specific biomolecules, a unique property of BNTs.

Alginate/BSA nanotubes



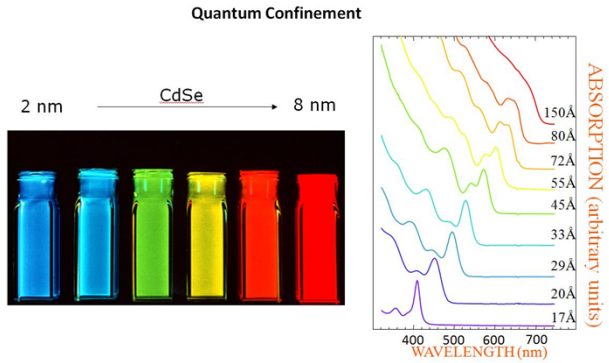
Layer-by-layer fabrication of GRAS nanotubes include alginate and BSA on polycarbonate template. SEM images of alginate/BSA nanotubes with diameter of 800 nm (left and right), and 400 nm (center) (Maldonado et al., 2017)

Using Quantum dots as imaging tools for cereal proteins in bread

Quantum dots nano-crystals were used to understand the molecular organization of food proteins in flat bread in order to assess their impact on morphological and structure characteristics.

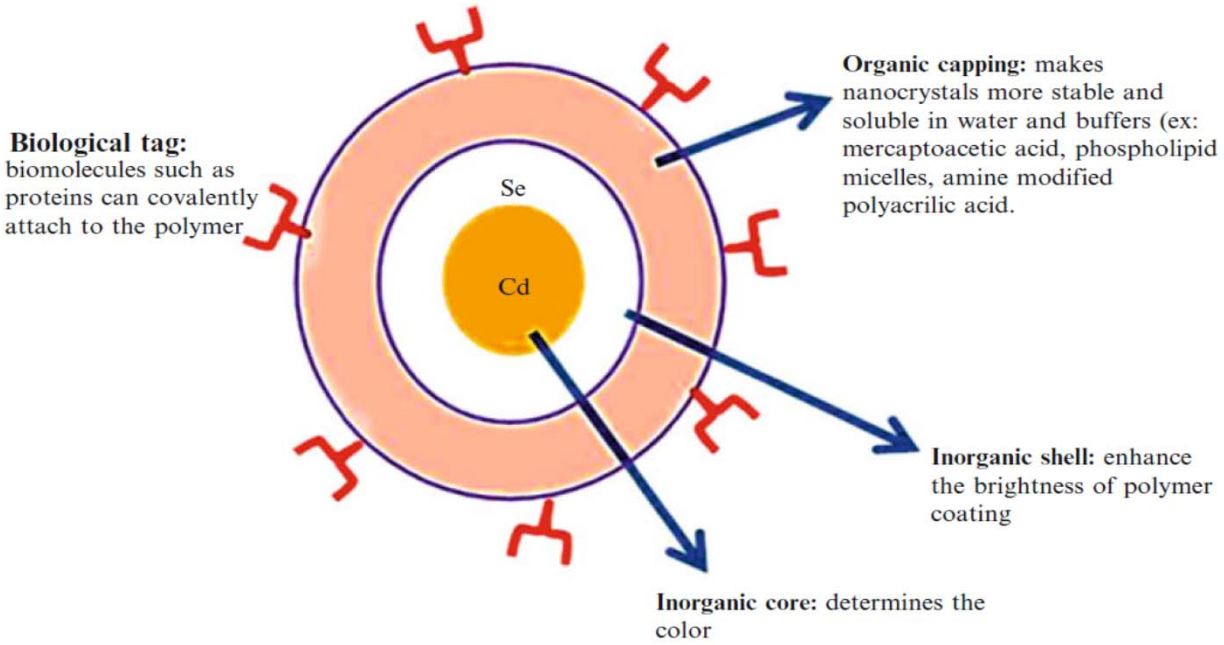
The protocols in sample preparation and labeling process were developed

(Courtesy of N. Sozer)



Quantum dots: application as food structure and microorganisms detection tools

QDs are semi-conductor materials that have a core-shell structure



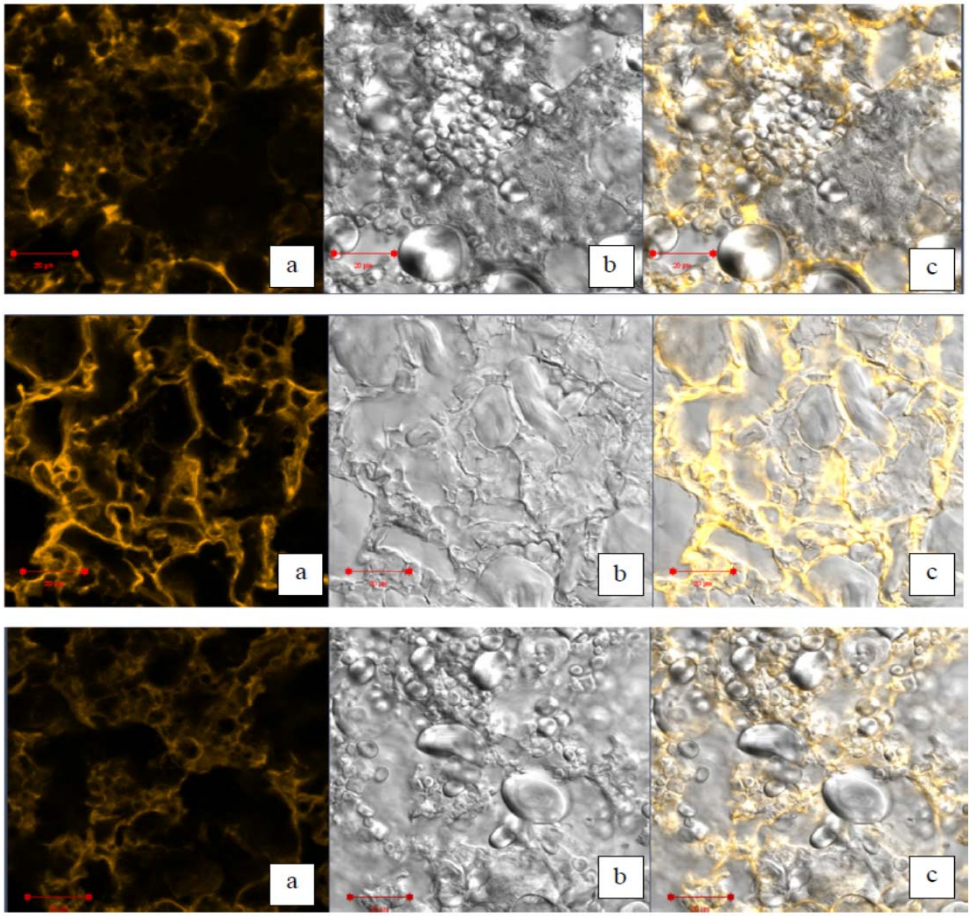
Multifunctional quantum dots can be used to target and image complex systems simultaneously (Luecha, J., Sozer, N., Kokini, J. L. 2013)

Quantum Dots (QDs)

- The size of the inorganic core can lead to different colors from red (large QDs with a size of 5-6 nm) to blue (small QDs with a size of 2-3 nm).
- The inorganic ZnS shell improves the brightness of QDs, while the organic capping with typically a polymer can make them water soluble and stable in aqueous media such as buffers.
- The most prevalent QDs are composed of atoms from group II and VI (e.g. CdSe, CdS, CdTe) elements or III–V elements of the periodic table.
- QD diameters are very small **(2–8 nm)** and the confinement of electrons and holes (an empty state which is previously occupied by an electron) becomes significant between sequential energy levels, and such characteristics leads to their unique spectroscopic properties.

Distribution of gluten in baked bread

- Merged fluorescence and differential interference contrast (DIC) images of QD labeled gluten proteins from top–center–bottom sections of baked bread samples are shown.
- The top and bottom sections showed a great deal of ungelatinized starch granules surrounded by an uneven gluten network.
- Parts of the gluten network was dense, as evidenced by the darker yellow color observed.



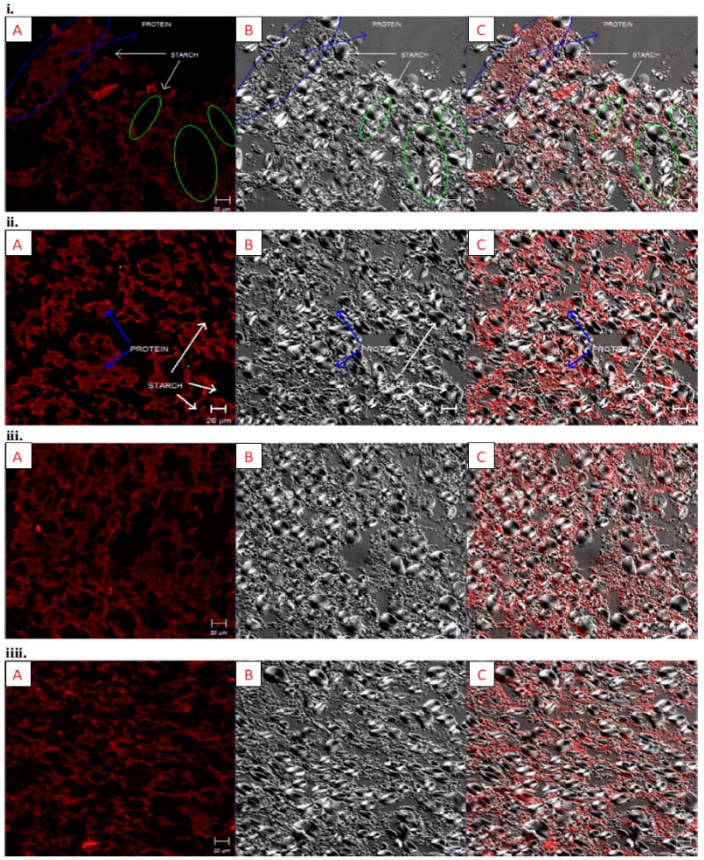
Bread sample (top–center–bottom cross-sections of bread) labeled with QDs 40× objective. a) Fluorescence image; b) DIC image; c) merged fluorescence–DIC image (Sozer and kokini, 2014).

Distribution of gliadin in dough using QDs and CLSM in reflection, transmission, and the overlay of transmission and reflection.

Microstructure of dough sections at i) arrival time, ii) peak time, iii) departure time, and iv) 10min after departure time. A (left), protein molecules bound to quantum dots.

B (middle), starch granules under polarized light

C (right), overlay of A and B showing the distribution of gliadin in the dough matrix in red and around starch. (Bozkurt et al., 2014).

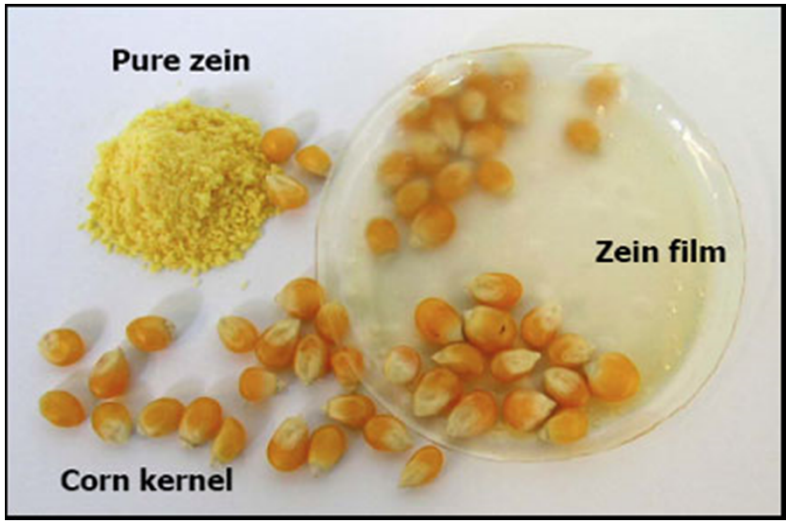


Zein biofilms

Many factors such as formulation, processing techniques, the state of film forming polymer at a particular temperature, and the water activity level may affect the mechanical and barrier properties of biofilms, such as zein films.

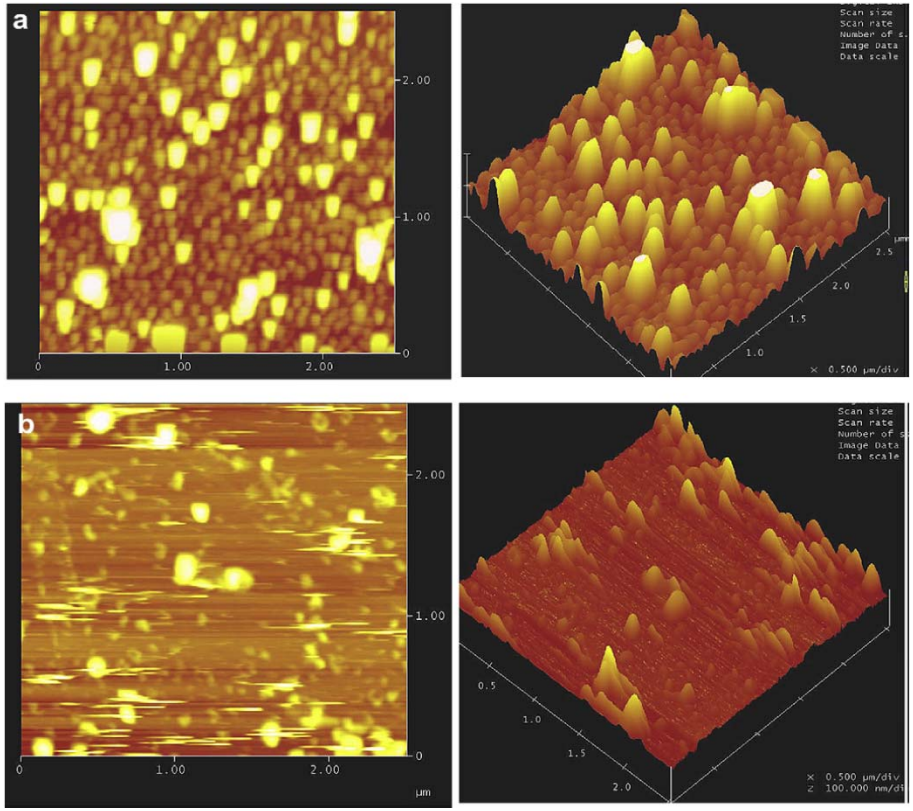
Zein isolate contains four major fractions including α, β, γ, and δ molecules with different molecular weights and characteristics.

Evaluation of the effect of these fractions on the film surface topography helps us to understand more about the prepared zein films



Effect of two zein fractions on the film surface properties

- Two fractions of unpurified zein are studied as fraction 1, which is rich in α-zein, and fraction 2, which is rich in β-zein.
- The alpha zein rich fraction results in a zein film which is less smooth than the beta-zein rich fraction. Measuring the roughness of the zein films from unpurified, α, and β zein shows that (25 images for each sample), the minimum and the maximum roughness comes from unpurified and α zein, respectively.
- These results suggest that α-zein fraction can self-assemble to form rougher but more homogeneous films compared to β-zein fraction because of higher hydrophobicity (Panchapakesan et al. 2012).

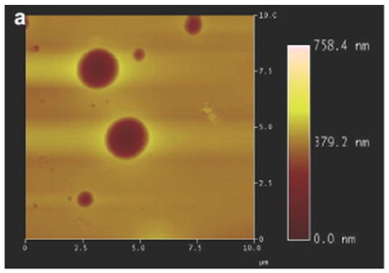


. Fraction 1 (a) and fraction 2 (b) (0.1% in 70% ethanol) as imaged by the AFM; top view (left), 3D view (right), scan size=2.5 mm, data scale=100 nm (Panchapakesan et al. 2012).

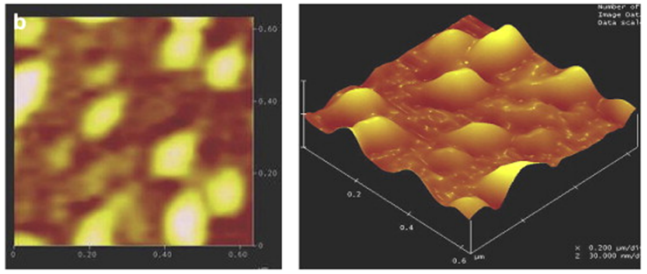
Zein topography: Effect of methods of film preparation

The topology of zein films was influenced greatly by the film fabrication methods.

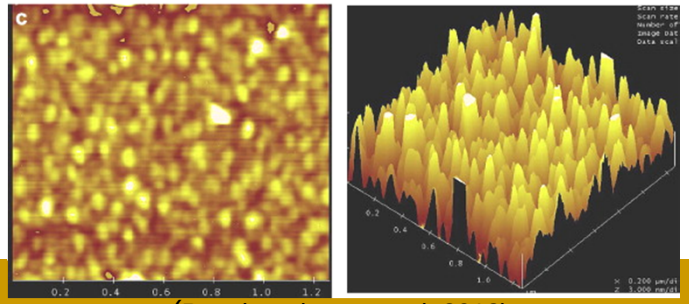
Spin casting resulted in more controllable nanostructure on zein surface.



zein film cast from solution on a petri dish



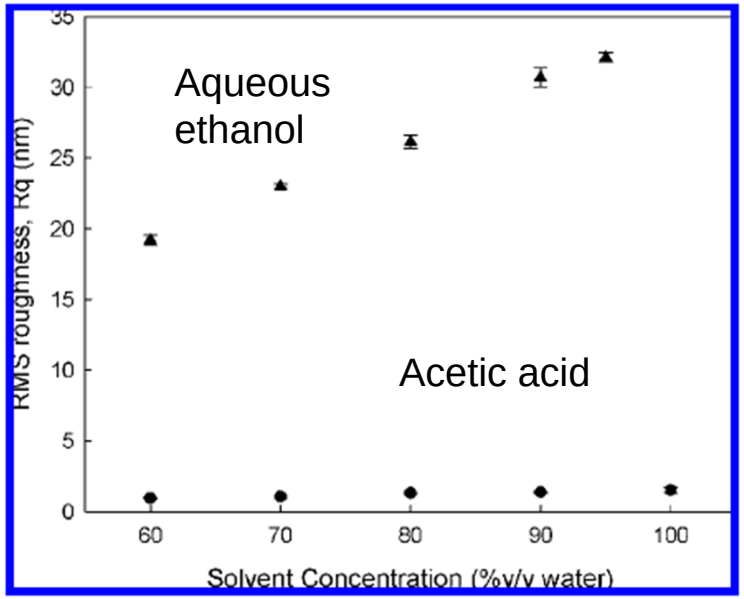
zein solution dropped on a silicon wafer.



zein film spin cast on a silicon wafer.

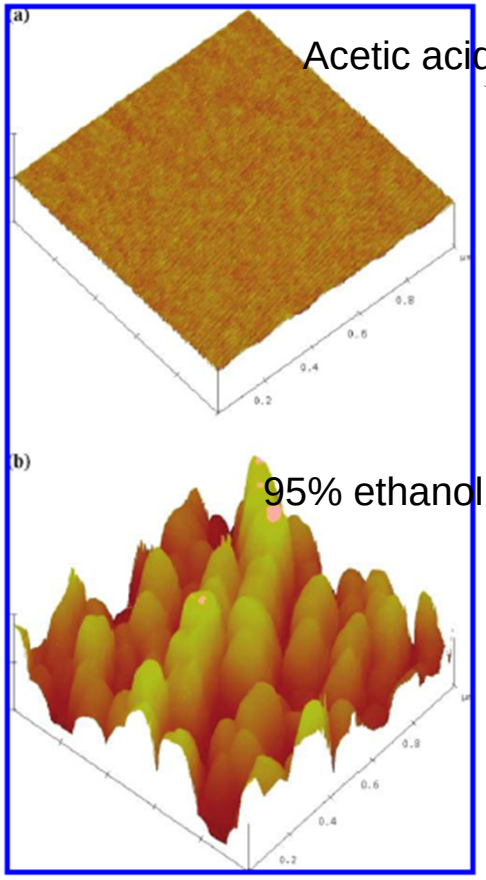
(Panchapakesan et al, 2012)

Zein topography :Effect of solvents



Zein films prepared from acetic acid showed smoother surface than ones from aqueous ethanol.

Increasing ethanol concentration resulted in rougher surface.



(Ke, Kokini, and Huang, 2009)

UV/Ozone treatment on zein surfaces

UV/Ozone treated, oxidized zein surface resulted in decreases of water contact angle as exposure time increased.

The UV/Ozone converted some of the C-H groups to $-COO^-$ groups as quantified by X-ray photoelectron spectroscopy (XPS).

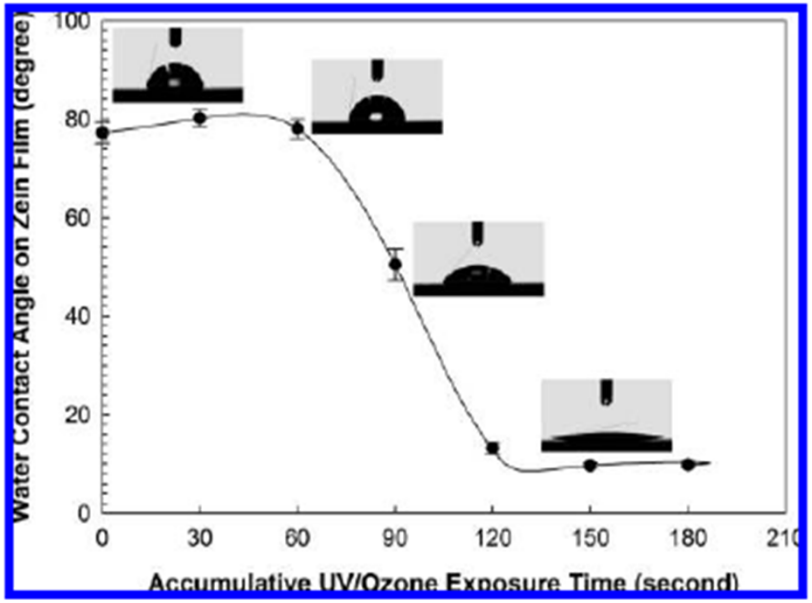


Table 2. Surface Elemental Compositions of Zein Films Prepared from Pure AcOH during Different UVO Exposure Times Obtained from XPS Survey Scans[a]

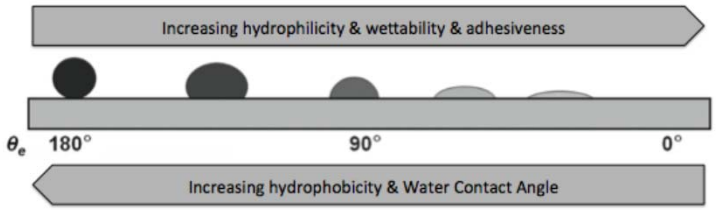
UVO exposure time (s)	elemental composition				water contact angle (degree)
	C (%)	N (%)	O (%)	O/C	
0	71.4	12.0	16.6	0.23	82.4
90	65.1	12.7	22.2	0.34	41.9
180	62.2	14.2	23.6	0.38	11.1

[a] For comparison, the water contact angles from the contact angle measurements were also provided.

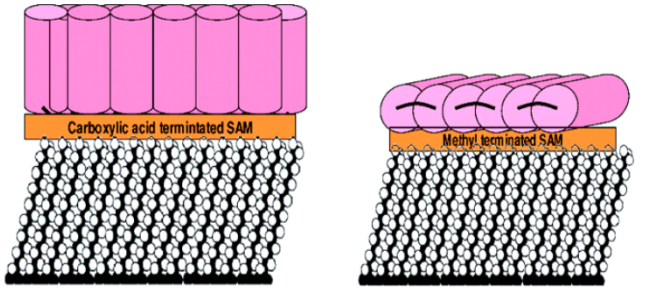
(Shi, Kokini, and Huang, 2009)

Zein self-assembles depending on the properties of the surface it comes in contact with during film formation.

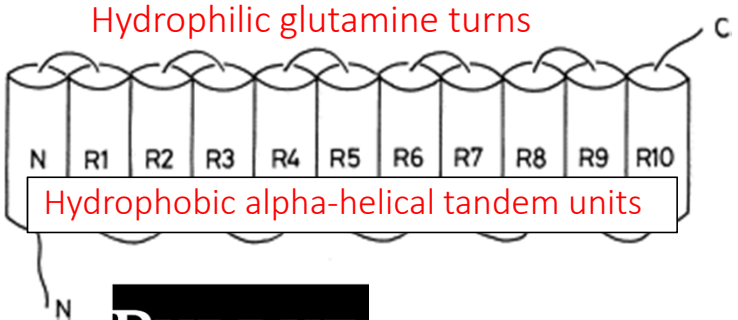
Water contact angle method to assess surface hydrophilicity

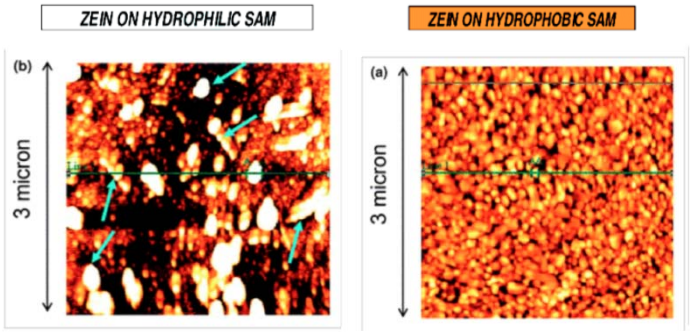


Zein self-assembly on hydrophilic vs hydrophobic surfaces



Conformational model of zein



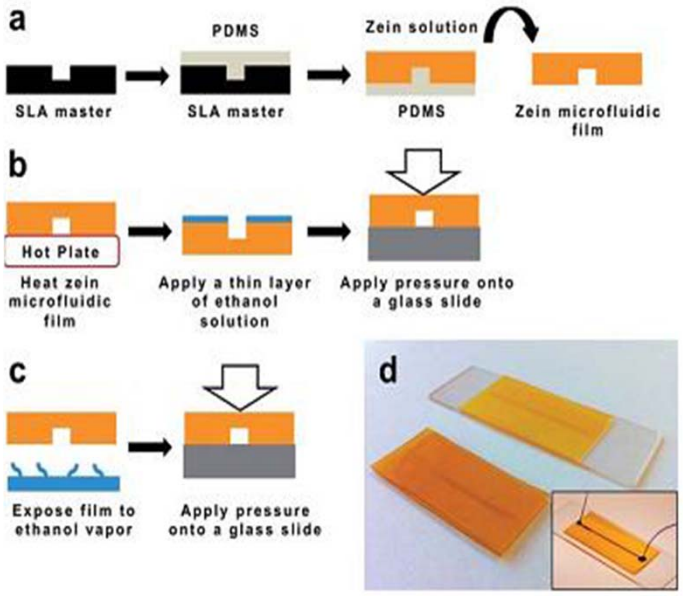


Atomic Force microscopy images

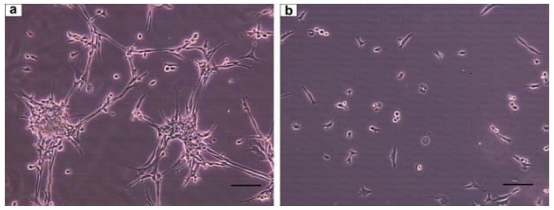
Matsushima *et al. 1997*
Subramanian and Sampath 2007

Surface hydrophilicity of the films are critical in microfluidic and tissue engineering applications

Zein microfluidics



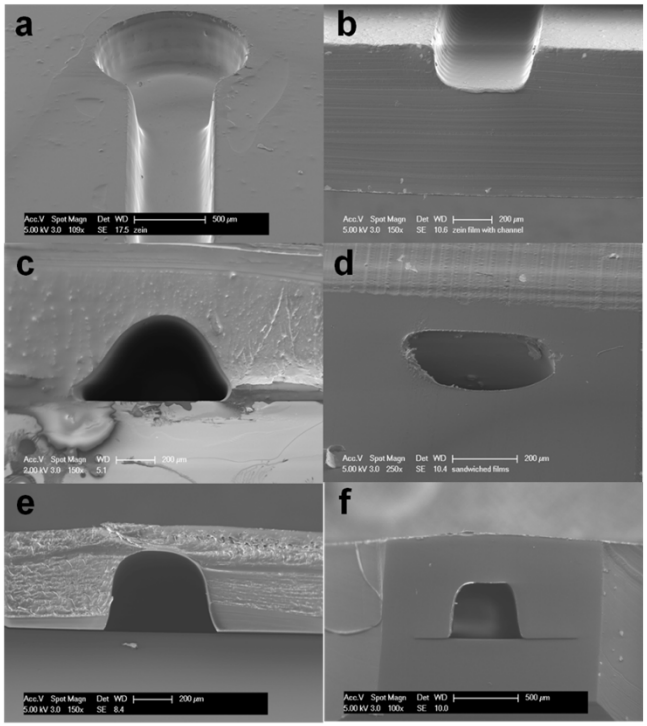
Zein as a biomaterial in Cell and/or tissue engineering



Zein as a food packaging/coating material

Luecha *et al.* 2011, Wang et al 2008

Scanning Electron Microscopy

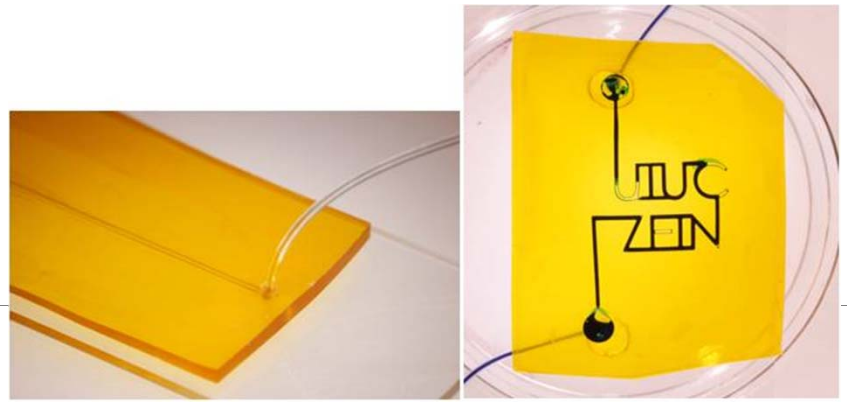


Zein film can accurately replicate the microfluidic features

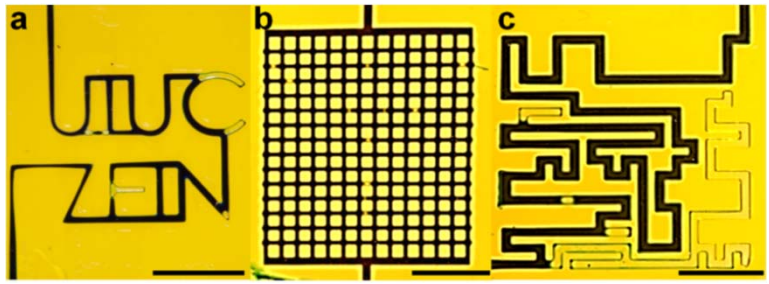
Microfluidic channels on the zein film retains the original geometry and with negligible distortion

The cross-sectional SEM images of zein-glass and zein-zein microfluidic devices bonded by solvent bonding method are shown in **(c)** and **(d)**. The images of the devices bonded by vapor deposition, zein-glass **(e)** and zein-zein **(f).**

(Luecha, Hsiao, Brodsky, Liu and Kokini, 2011)



The zein-glass microfluidic device with tubing at the inlet (Left). The bottom of zein-glass microfluidic device with channel written as UIUC ZEIN filled with blue fluid (Right)



Visualization of zein-glass microfluidic devices with complex fluidic pathways. (a) Interconnected letters composed of continuous microfluidic channels, (b) a microfluidic network with channels and chambers, (c) a solved microfluidic maize maze with multiple false paths. Blue food dye was used for visual aid. All scale bars are 5 mm.

(Luecha, Hsiao, Brodsky, Liu and Kokini, 2011)

Functionalization of materials at the nanoscale: nanocomposites

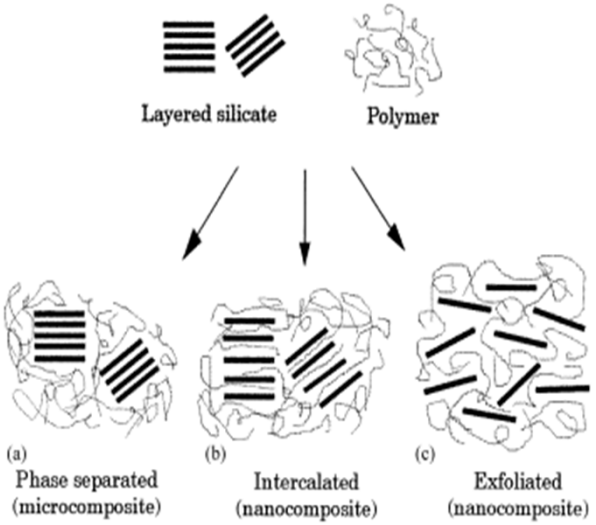
Nanoclays

- Biocompatible composites are interesting because of their renewability and environmental friendly properties.
- Zein is one of the best proteins for preparation of biocompatible films, however the films from pure zein are very brittle.
- Zein film properties can be improved by using plasticizers, chemical cross-linking, laminating, etc.
- Use of polymer layered silicate nanocomposites (PLSN) is an option to improve the properties of natural films such as zein.

Fabrication of zein nanosilicate nanocomposite films

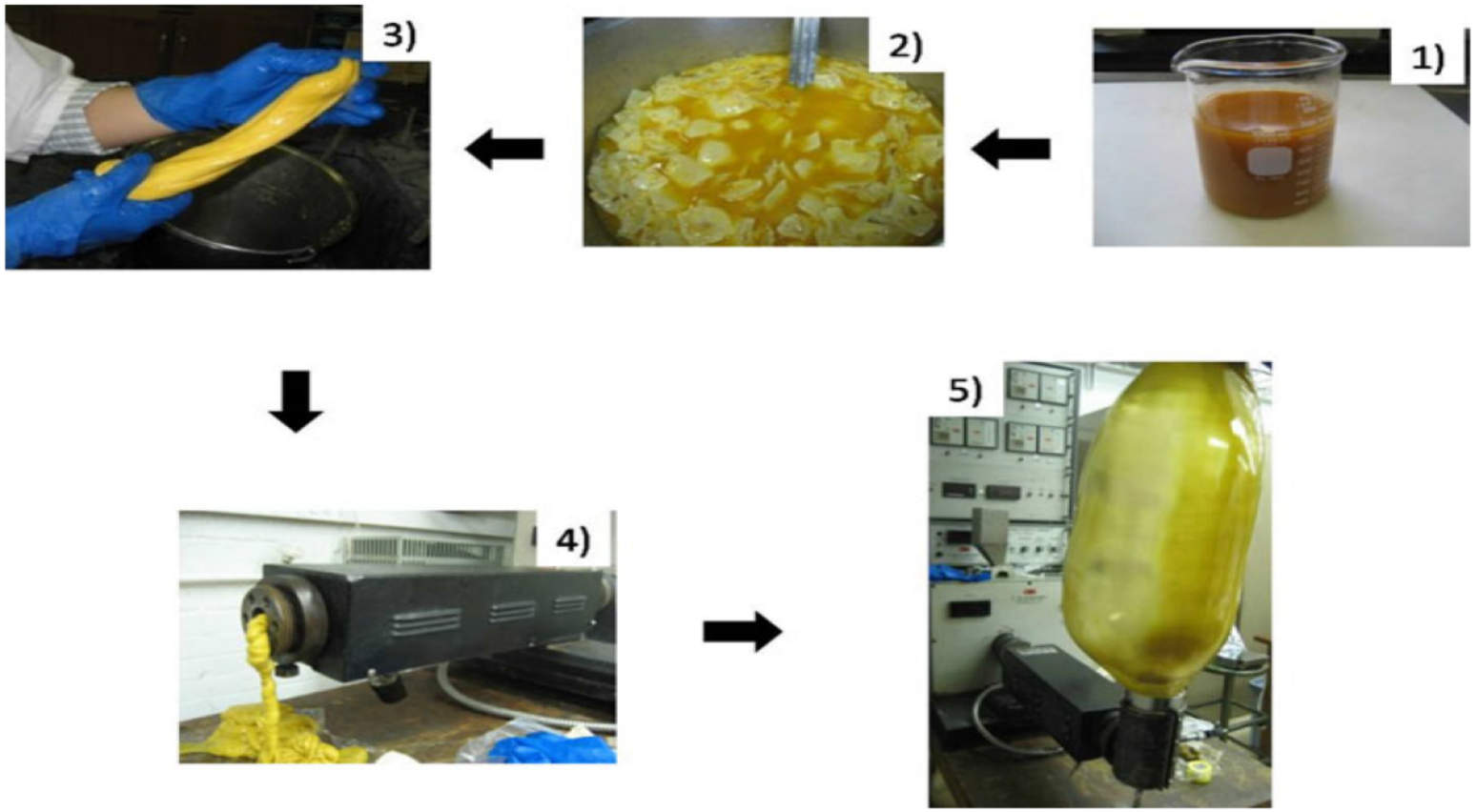
Zein nanoclay nanocomposite films were fabricated at different nanoclay contents using two methods: solvent casting and extrusion blowing methods.

The microstructures, mechanical properties and barrier properties were characterized.



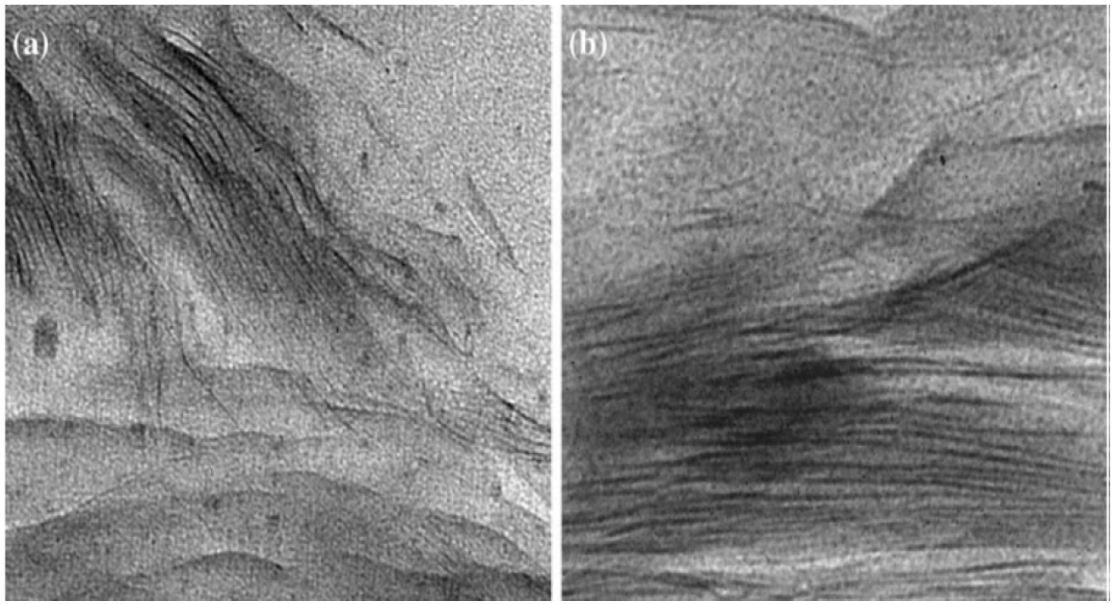
Polymer nanocomposite structure

(Luecha , Sozer and Kokini, 2010, J. Mater Sci. 45:3529-3537.)



The steps of the blown extrusion technique including 1 zein MMT solution, 2 precipitation, 3 resin formation, 4 cold extrusion, and 5 balloon formation (Luecha, Sozer, and Kokini, 2010).

The results from XRD and TEM suggested that both nanocomposite preparation methods are able to form partially exfoliated nanocomposite structures from montmorillonite and zein.



TEM images of a solvent cast zein MMT nanocomposite films with 5 wt% of MMT, and b blown extrusion zein MMT nanocomposite films with 5 wt% of MMT (Luecha, Sozer, and Kokini, 2010).

Zein films mimick the surface properties of PDMS after oxygen plasma treatment

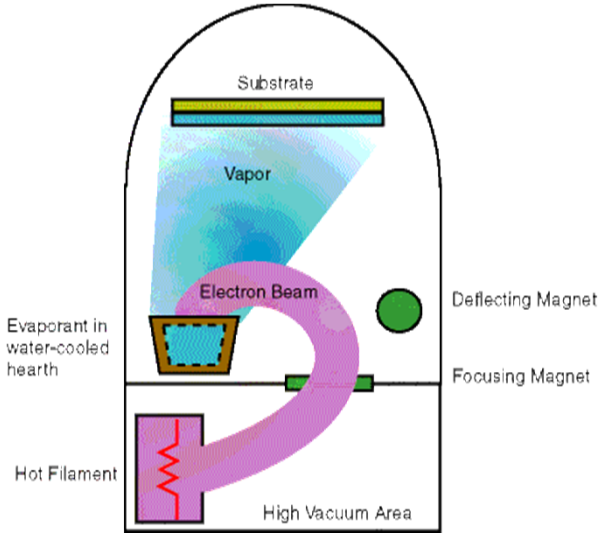
- Zein films mimick surface properties when they come in contact with PDMS
- Modification of the surface properties of zein films which are in contact with PDMS (polydimethylsiloxane) and exposed to oxygen plasma might be an interesting technique to control hydrophilic/hydrophobic properties balances on the surface of zein films.
- Applying oxygen plasma exposure on PDMS increases hydrophilic properties of the PDMS surfaces and the water contact angle .
- Water contact angle of zein shows higher hydrophilicity when in contact with oxygen plasma treated PDMS right after peeling off from zein film.

Noble metal coating of zein film surface

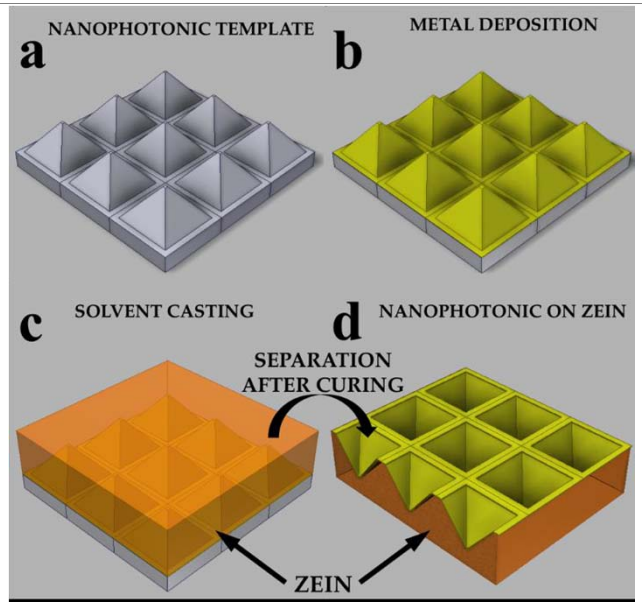
Challenge

Zein films melt in

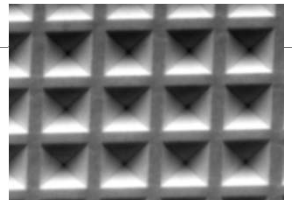
E-beam evaporator



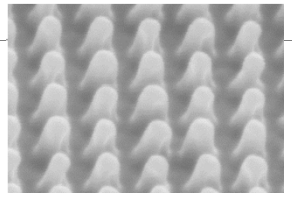
Direct metal transfer onto zein films



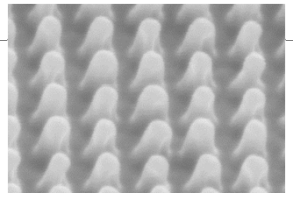
Nanophotonic patterns transfer successfully onto zein films



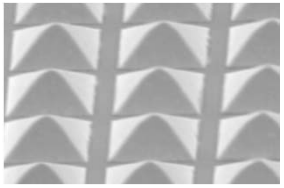
Master mold with inv. pyramids



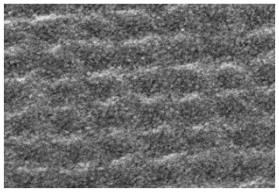
Master mold with nanopillars



Master mold with nanopillars

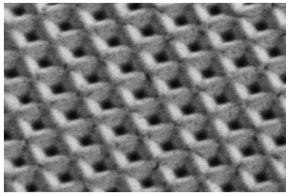


PDMS with pyramids

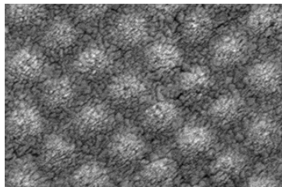


PDMS with pores

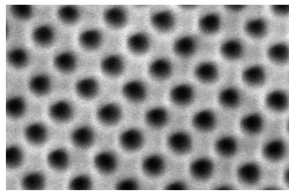
Master mold with nanopillars



Zein with inv pyramids



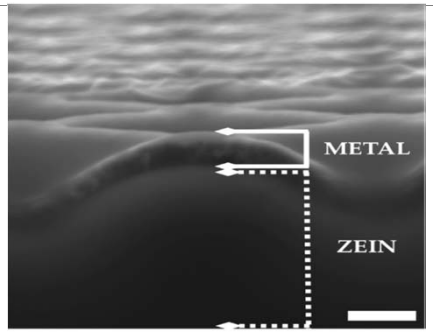
Zein with nanodomes



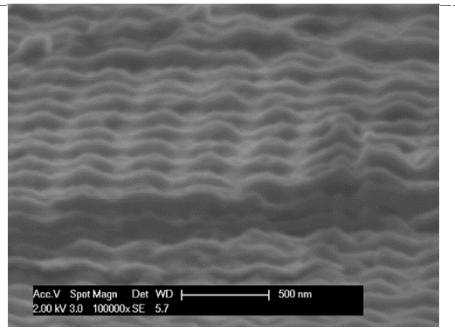
Zein with nanopores

Zein shows perfect adhesion with metal

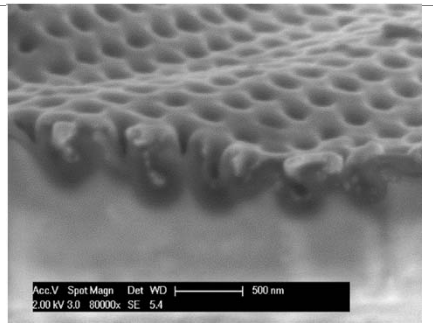
Inverted pyramids on zein



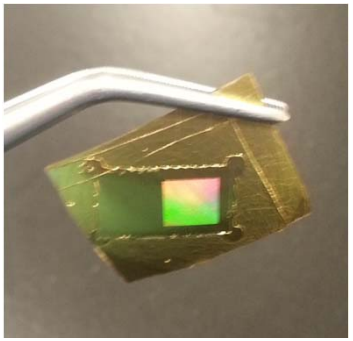
Nanodomes on zein

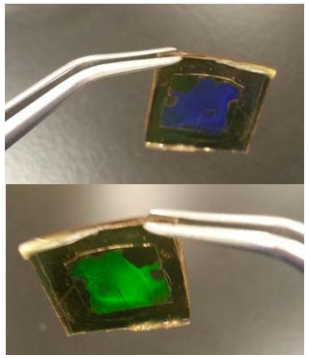


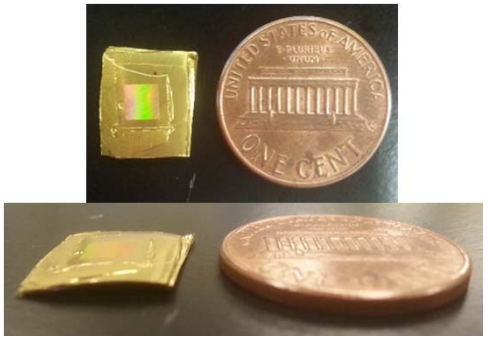
Nanopores on zein



Macro images

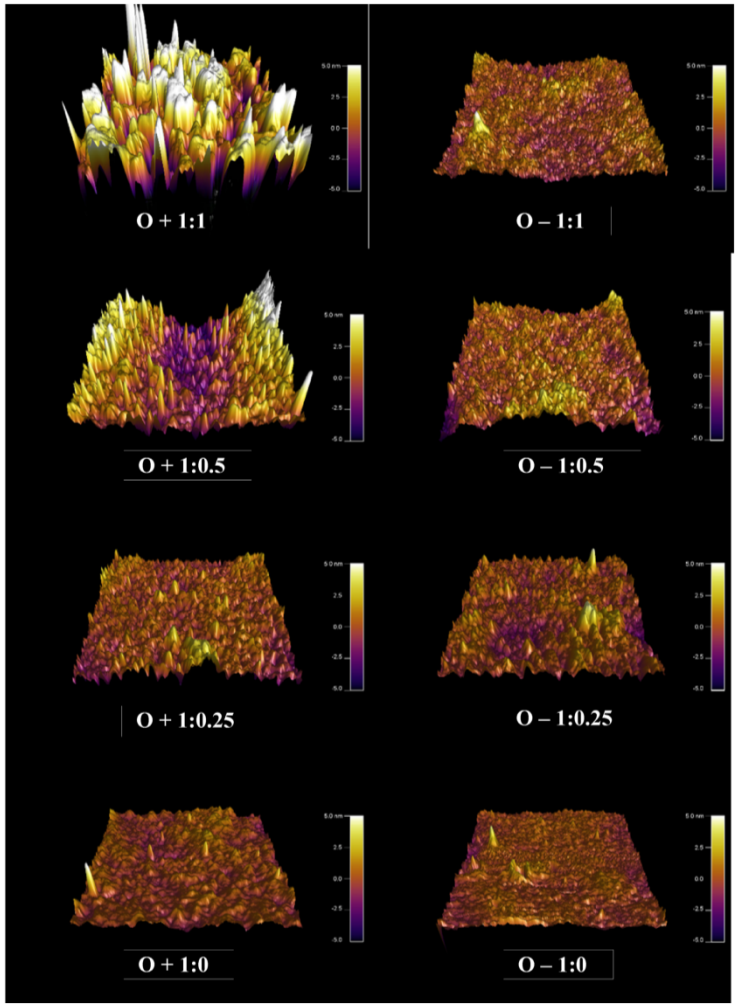






AFM to understand th effect of plasma

- AFM analysis might help to understand more about surface properties of the cast zein films. AFM results show that in all oleic acid contents, the zein film surfaces were rougher after oxygen plasma exposure treatment. For oleic acid-zein ratio of 1:1, the RMS (root mean square) of the zein film is 4.5 and 0.6 nm with and without oxygen plasma treatment, respectively.
- AFM results also show that the difference between RMS values between zein films cast on untreated and treated PDMS, decrease when oleic acid content decreases. These differences for zein to oleic acid ratios of 1:1, 1:0.5, and 1:0.25 were 3.7, 0.45, and 0.19 nm, respectively. Both oleic acid content and chemistry of the pattern can change orientation of oleic acid-zein film.



AFM images of zein films.

O+: zein was cast on PDMS exposed to oxygen plasma,

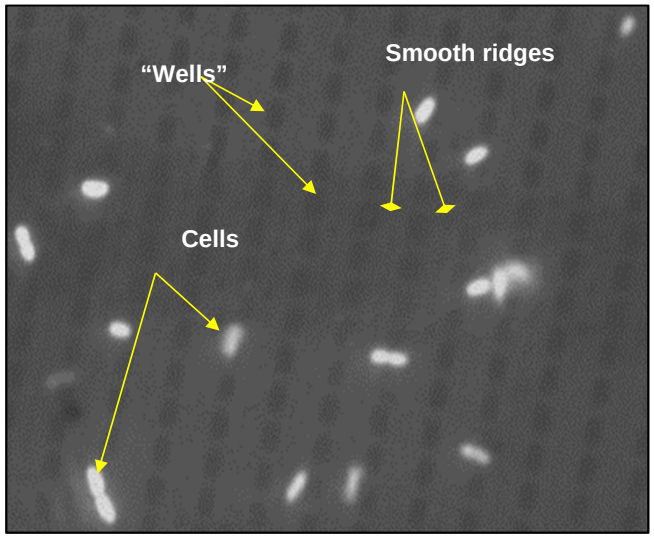
O−: zein was cast on PDMS.

Numbers represent the zein: oleic acid ratio.

Scale bars range from 5 nm (yellow) and −5 nm (blue) (Gezer et al. 2015).

Microbial repellant surfaces based on nanoscale topography

Hsu et al (2013) used silica wafers with surface details of various shapes and sizes in the micrometric range to study bacterial attachment, and found out that orientation of the attached cells occurred preferentially such that cells maximized their contact area with the surface

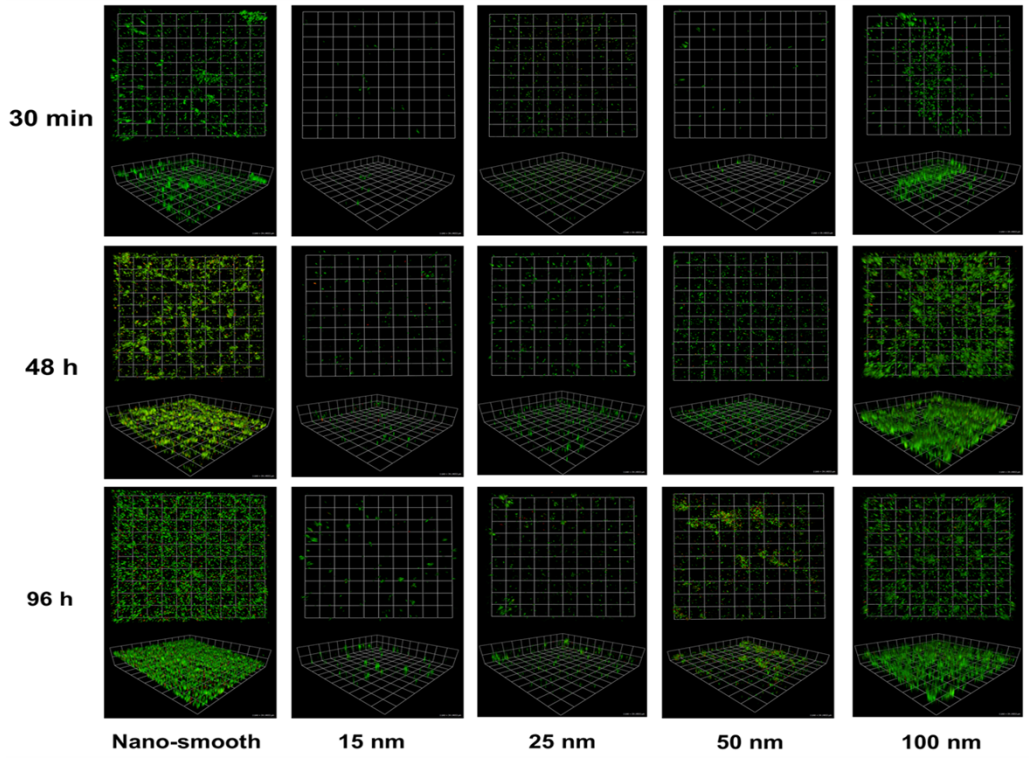


P. fluorescens cells on silica wafers, showing preferential positioning relative to surface topography (Hsu et al., 2013).

Effect of surface topography on bacterial attachment

Nanoporous surfaces with pore sizes of 15 nm and 25 nm, significantly reduced attachment of bacteria compared to a nanosmooth control including

Surfaces with pore diameters of 50 nm or larger increased attachment compared to both the small scale pore surfaces and the control



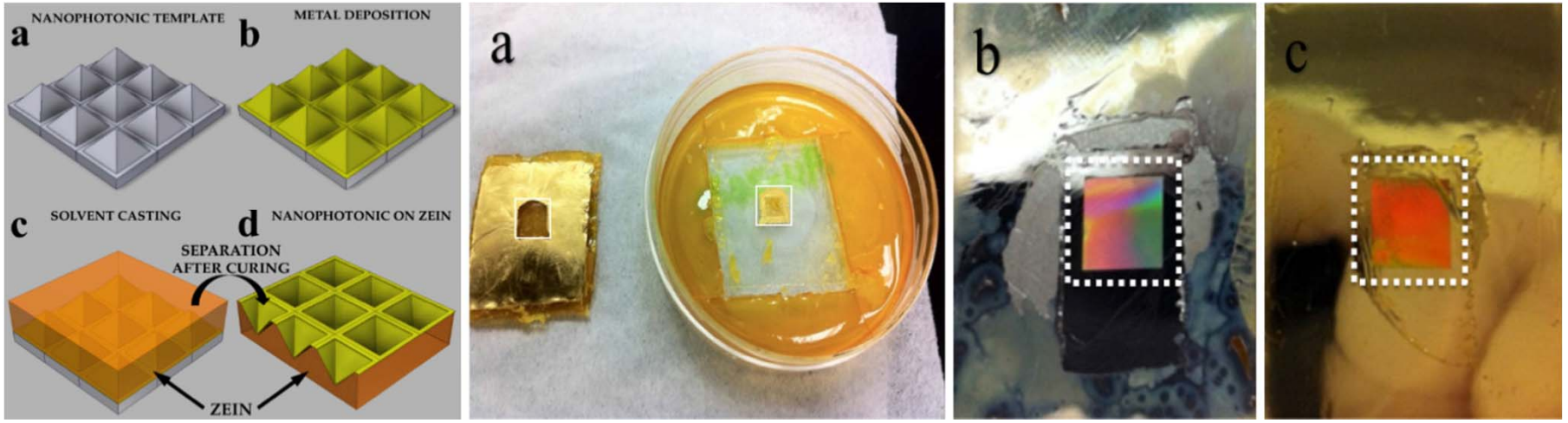
Attachment and biofilm formation by *E. coli* ATCC 25922 at 30 min, 48 h and 96 h on nano-smooth alumina (control) and anodized surfaces of 15 nm, 25 nm, 50 nm and 100 nm pore diameter. (Feng et al., 2014).

Application of nanotechnology in fabrication of *biosensors*

Fabrication of zein nanophotonic platform

- Surface-enhanced Raman spectroscopy (**SERS**) is a technique for molecular detection and characterization that relies on the enhanced Raman scattering of molecules that are adsorbed on, or near, **SERS**-active surfaces, such as nanostructured gold or silver.
- Combination of Raman spectroscopy and Signal enhancement by nanophotonic structures can help fabricate nanobiosensors.

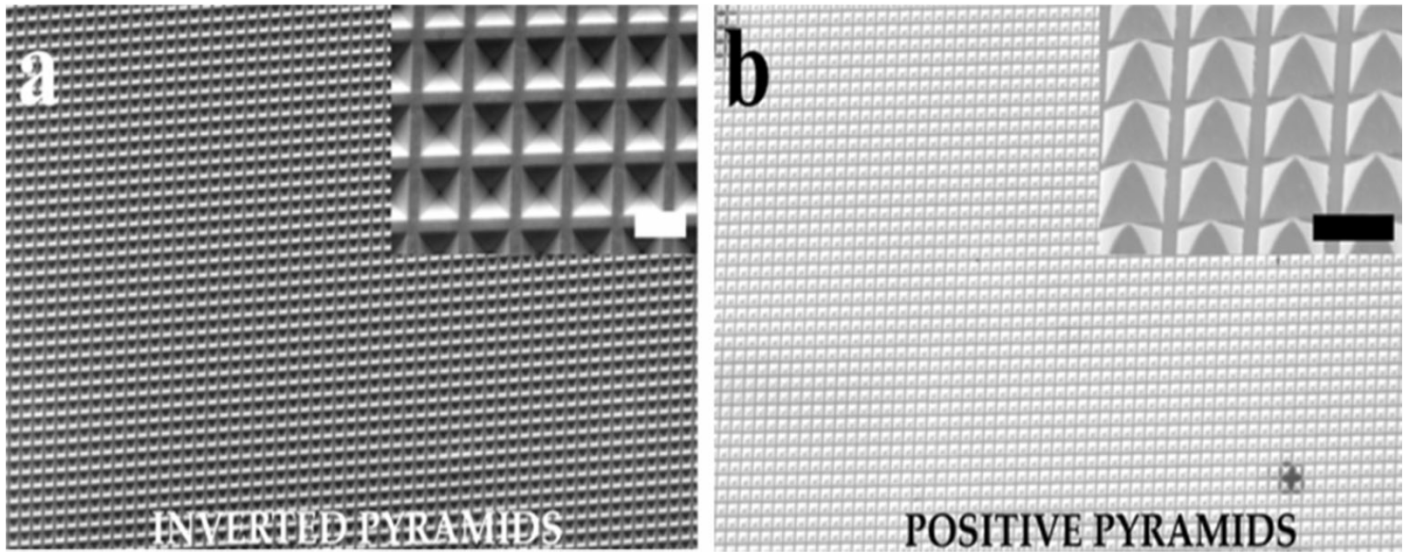
Zein-based SERS biosensor production



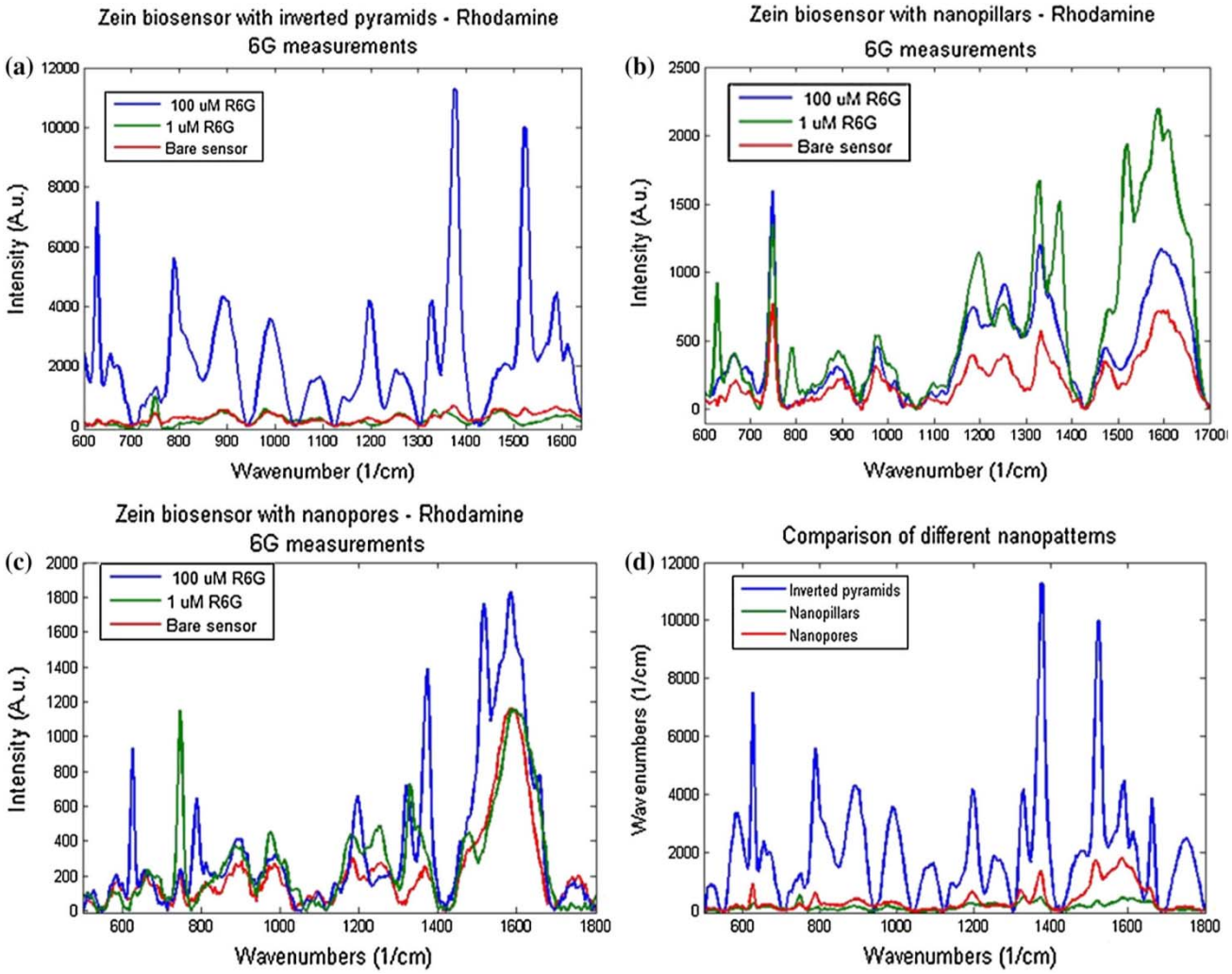
Left: direct transfer of three dimensional metallic nanophotonic structures onto zein (a) is deposited with 200 nanometers of noble metal using E-Beam Evaporation (b). Zein solution is solvent-casted over the metal-coated template (c), and after fully solidifying; the zein film with three-dimensional metallic nanophotonic structures is separated from the template (d).

Right: The transfer of noble metal onto zein film. Unsuccessful transfer evident by the squared area having the patterns (a) did not transfer onto zein film (on the right), successful transfer of silver (b) and gold (c) (Gezer et al., 2016a).

Nanophotonic patterns for Biosensor development



Scanning electron microscopy images of a) top-down view of the inverted pyramid nanophotonic structures and b) positive pyramid nanophotonic structures transferred onto zein (scale bars 2 μm) (Gezer et al. 2016a).



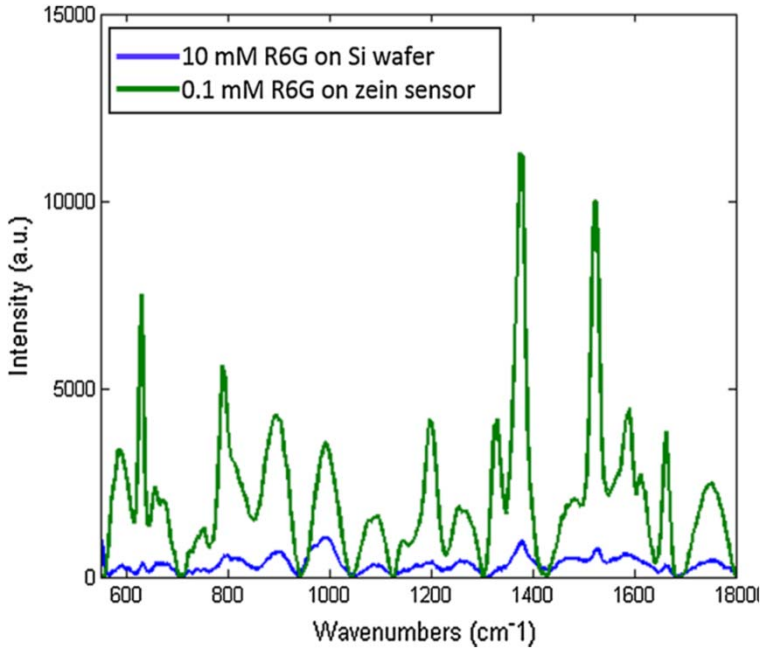
Different concentrations of Rhodamine 6G on:

a) 200 nm gold-coated inverted pyramid sensor on zein

b) 80 nm gold-coated nanopore sensor on zein,

c) 80 nm gold-coated nanopillar sensor on zein

d) comparison of 100 µM concentration of these sensors (Gezer et al. 2016a).

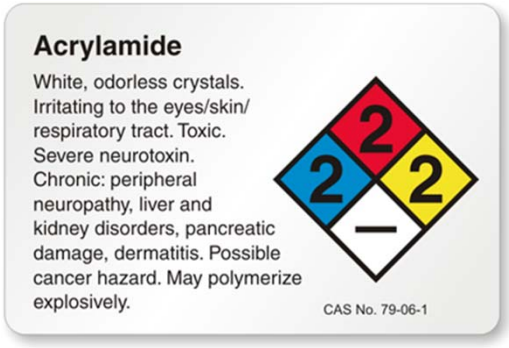


Comparison of SERS and normal Raman spectra of Rhodamine 6G (Gezer et al. 2016a).

The effect of gold-coated inverted pyramid zein platform on the intensity of the Raman peaks. Enhancement factor (EF) for this platform is 1.3×104, and this factor is similar to the original substrate which was used to prepare the platform.

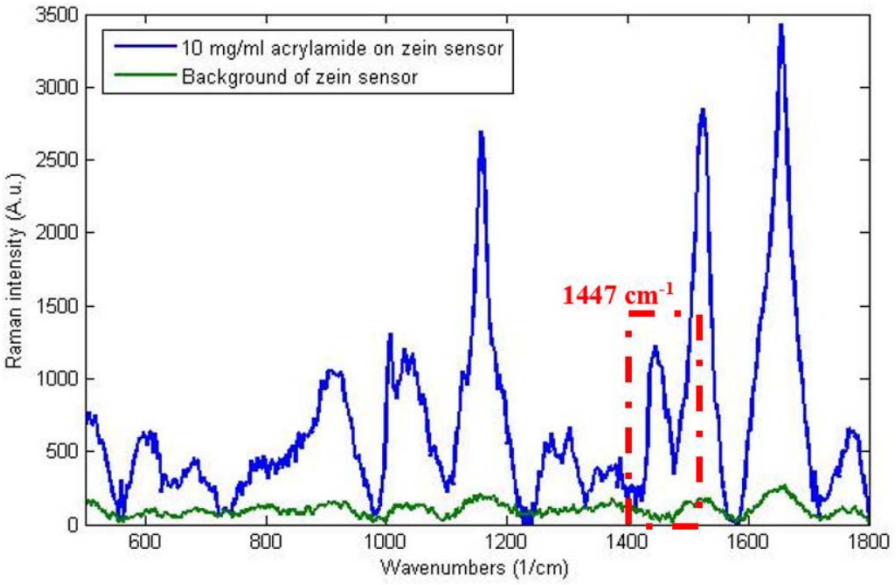
Toxicology applications of SERS biosensors

- Acrylamide is a chemical used primarily as a building block in making polyacrylamide and acrylamide copolymers.
- Acrylamide is formed during food processing operations such as frying and baking
- Acrylamide is considered a potential occupational carcinogen by U.S. government agencies and classified as a Group 2A carcinogen.



Zein-based acrylamide biosensor

Gold-coated zein-SERS platform is utilized to detect acrylamide.

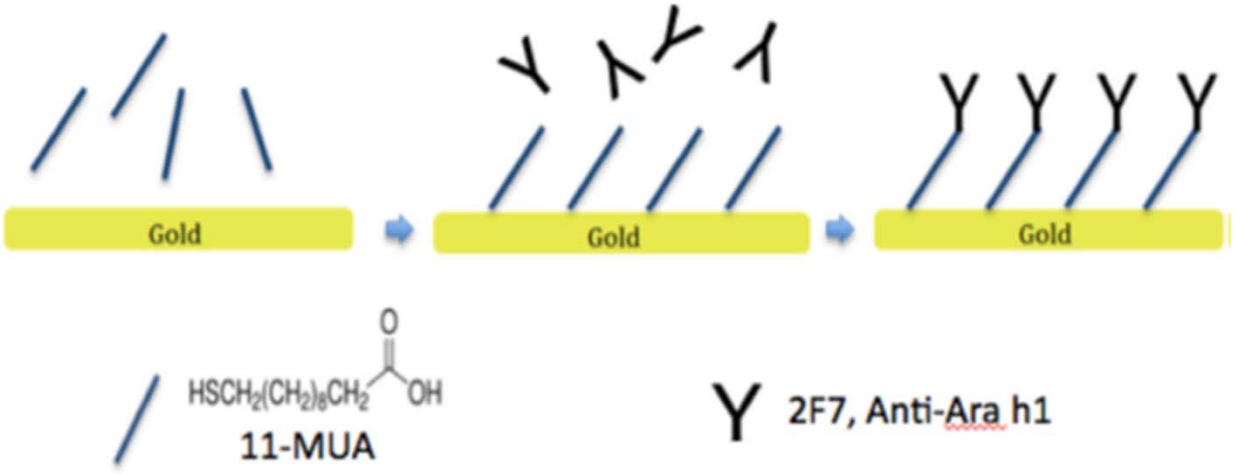


Comparison of the background signature of zein-SERS sensor (green) with acrylamide on top of the sensor (blue). Red dotted square indicates the peak at the wavenumber of 1447 cm^{-1}, which does not exists in the background, but exists in acrylamide signature. (A.u.: arbitrary units) (Gizer et al., 2016c).

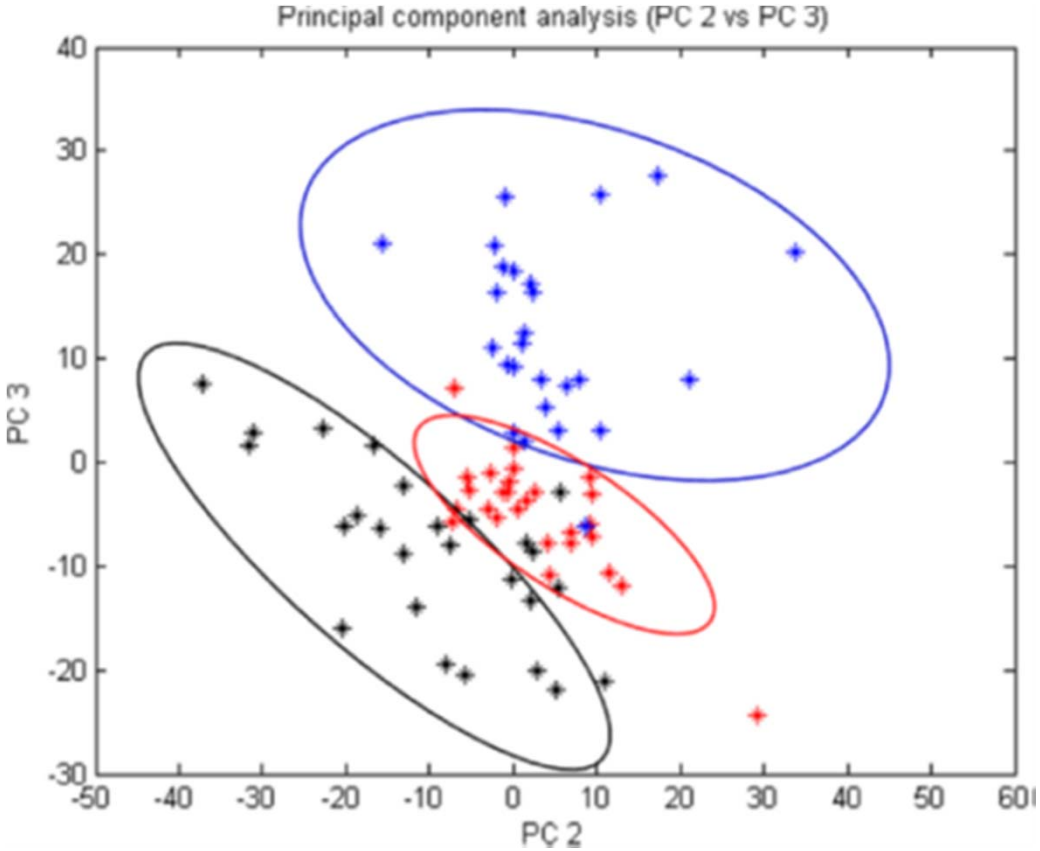
Toxicology applications

- The detection of peanut protein presence in food products is important for safety of consumers who are allergic to them.
- Ara h1 is the most abundant protein among these allergens.
- Enzyme-linked immunosorbent assays (ELISA) along with monoclonal or polyclonal antibodies is the current most commonly used technique for detection of the allergenic proteins of peanuts, including Ara h1-h8.

Fabrication of biodegradable platform coated with gold nanopattern along with SERS can be developed to detect the Ara h1 protein. Principal component analysis is employed for both detection and quantification purposes. The first step to prepare zein gold coated platforms for this application is functionalization of the surface of the platform with the monoclonal antibodies of Ara h1, 2F7.



Schematic illustration of the functionalization of the gold surface (Gezer, Liu, Kokini, 2016).



Principal component analysis (PC2 vs PC3) of baseline-corrected Raman spectra for background zein-SERS sensor (black), antibody-functionalized zein-SERS sensor (blue) and Ara h1 protein captured by antibody-functionalized zein-SERS sensor (red) (Gizer, Liu, Kokini, 2016).

Future trends

While methods for nanoparticle characterization have been developed and used in determining the physical-chemical characteristics of the nanostructures in their freshly synthesized form, little attention has been placed on tracking properties change under conditions of use.

Functionality of the entrapped bioactive and safety of the nanostructure is dictated by the fate of the nanostructure and therefore

1. Development of detection methods for organic, biodegradable nanoparticles in a complex food matrix,
2. Understanding nanoparticle-food component interaction, and
3. Tracking the nanoparticle in vivo, under conditions of use are important.

The question of safety can only be addressed by multidisciplinary collaborative groups with complementary expertise in food systems, nanostructure synthesis and characterization, and toxicology.

Thank you for having me as your dinner speaker