Problem 3: Mathematical Model of Mechanical Properties of Constrained Cell-Adhered Collagen Gels

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Background

- *in vivo*, cells respond differently when unconstrained or constrained by natural boundaries

- Cells respond differently to differences in extracellular matrix stiffness
  - Cells pull on the matrix and modulate their pulling force depending on stiffness

- Cells can sense substrate depth differently on different types of substrates

Background

- Using collagen gels to represent the *in vivo* extracellular matrix, we are interested in understanding changes in the mechanical properties of the gels in response to the presence of an adhered cell, varying lateral and longitudinal boundary conditions, and varying collagen concentrations

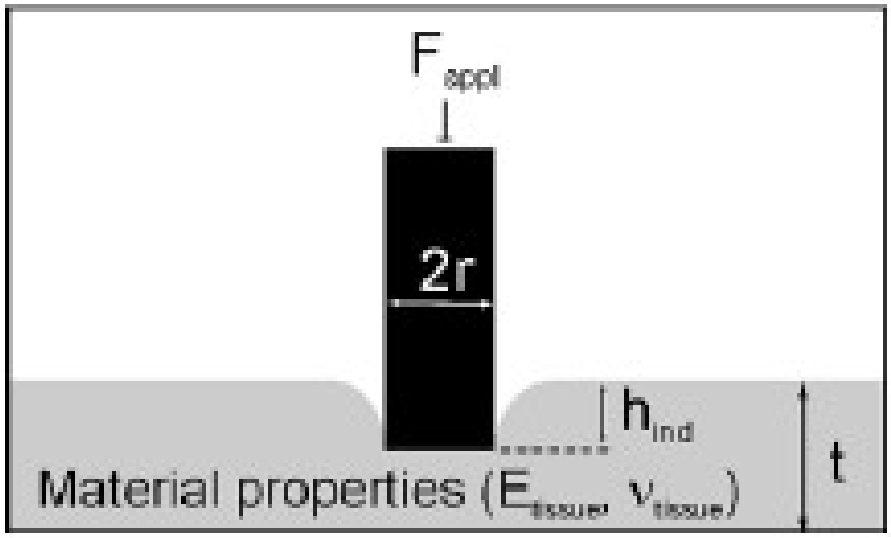
- Collagen fibres that comprise these gels are initially randomly orientated and homogeneously distributed
  - Cells pull on fibres resulting in non-homogenous distribution and local changes in orientation

Objectives

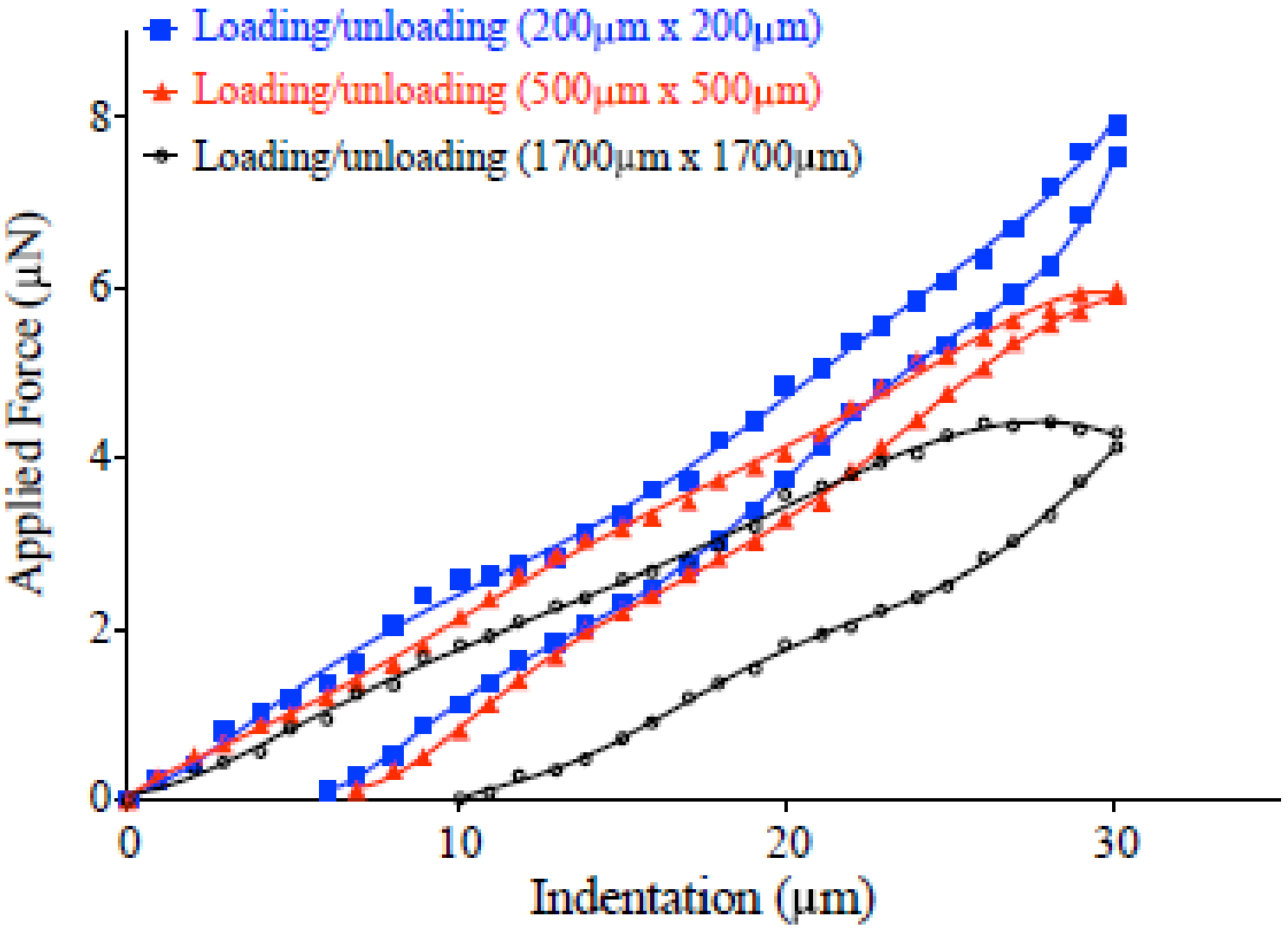
- Develop mathematical models of the mechanical properties of collagen gels subject to
  - different collagen concentrations
  - the presence of an adhered cell
  - the presence of different sized boundaries
- Utilize available experimental data along with results from numerical simulations to develop these models
- In the longer term, distinguish between various important features of the gel, such as nonlinearity, plasticity, and reorientation of the collagen fibres

Gel material property characterization

- Material properties of gels measured using indentation
  - No cell present
  - Homogenous distribution and random orientation of collagen fibres
  - Three different collagen concentrations



Experimental indentation force vs. depth



Calculation of gel material properties

- Assume gels are linear elastic and isotropic
  - First approximation – can be modified at a later time

- Use Hertz contact model to calculate compressive modulus of gels from experimental indentation data
  - For cylindrical indenter

$$F = 2aE'd$$

where F is indentation force, a is the tip radius, d is indentation depth, and $E' = E/(1-\nu^2)$.

  - Therefore

$$E = \frac{2(1-\nu^2)}{a}\frac{F}{d} = \frac{8}{3a}\frac{F}{d} \quad \text{if incompressible}$$

Calculated compressive moduli

- 4 cases
  - Small grid (w = 200 μm), 1 mg/mL concentration: E = 8.4 kPa
  - Med. grid (w = 500 μm), 1 mg/mL concentration: E = 7.0 kPa
  - Large grid (w = 1700 μm), 1 mg/mL concentration: E = 4.9 kPa
  - Large grid (w = 1700 μm), 3 mg/mL concentration: E = 5.6 kPa

- E decreases with increasing grid size and increases with increasing concentration

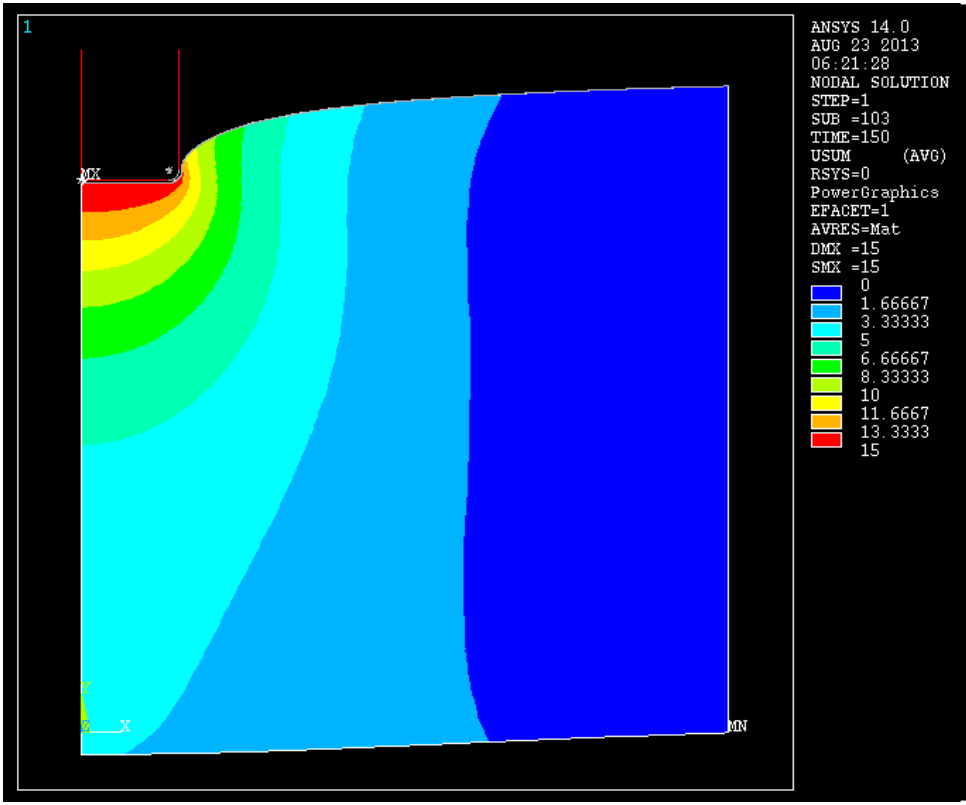
Numerical simulations of indentation

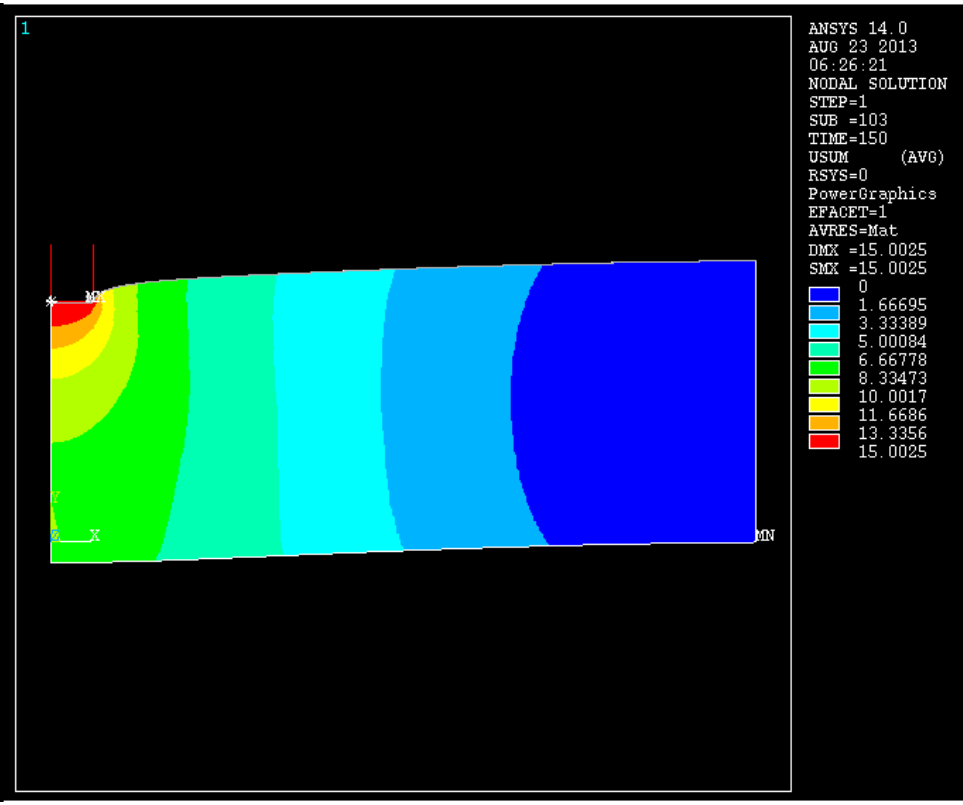
- Simulations performed using commercial finite element software ANSYS

- Indenter is cylindrical and rigid with diameter = 25 μm
  - 2-D axisymmetric simulations

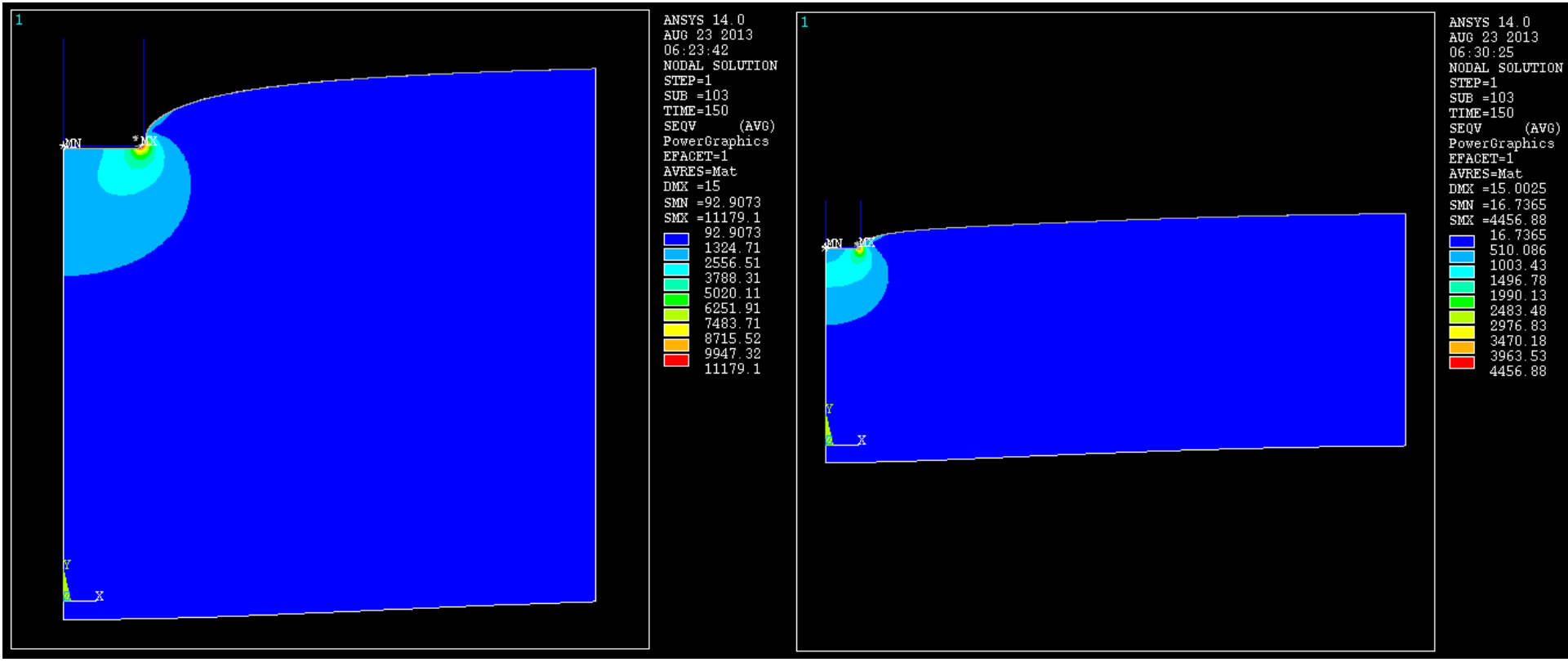
- Gel is 100 μm high
  - Three grid sizes: 200, 500, 1750 μm
  - One concentration: 1 mg/mL

Simulation results - Displacement



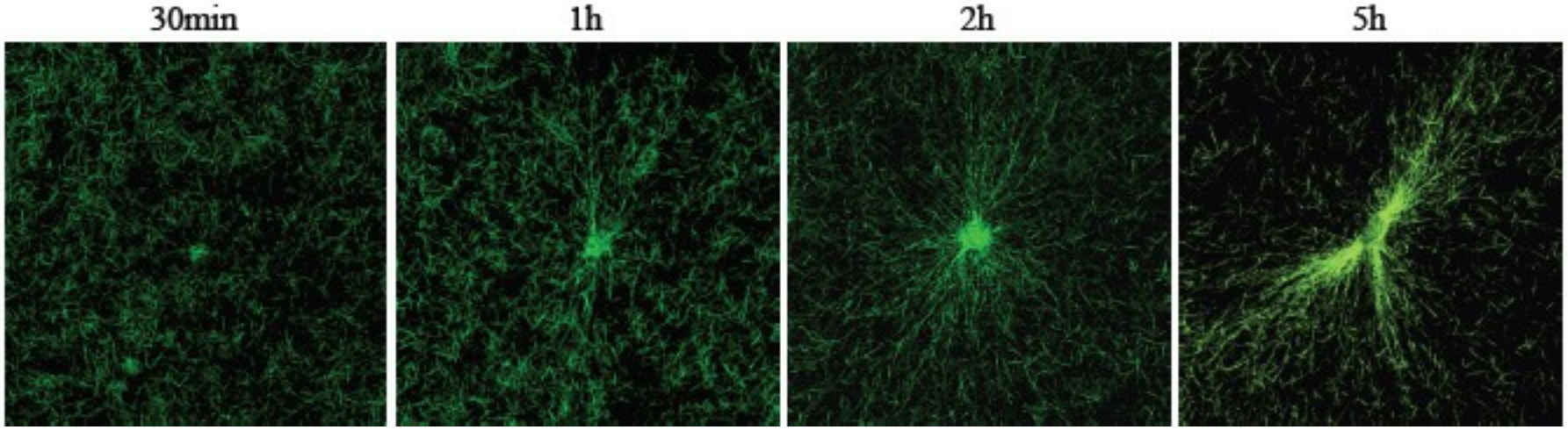


Simulation results - Stress

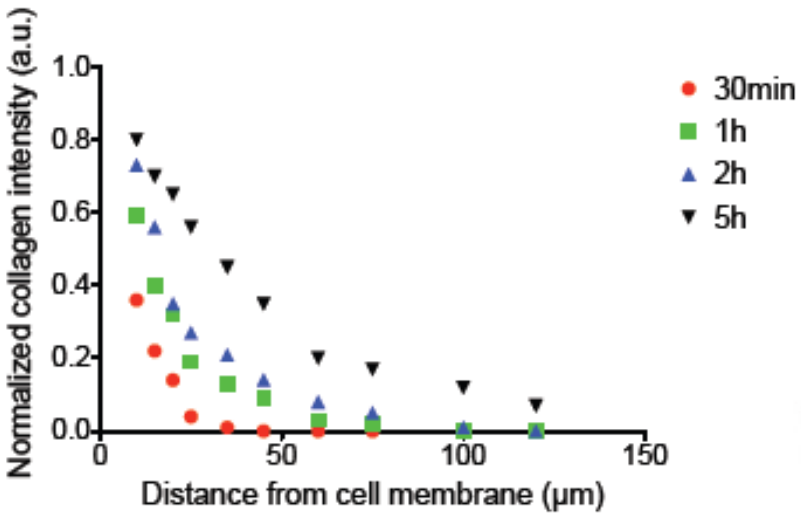


Cell experiments – Confocal microscopy

- A single cell seeded onto a collagen gel
- Confocal microscopy images of collagen intensity taken at several time points during experiment



Collagen intensity vs. concentration



- Want to correlate collagen intensity with collagen concentration
- Using relationship between concentration and compressive modulus, we can develop a piecewise spatially varying linear elastic model for collagen gel after cell presence

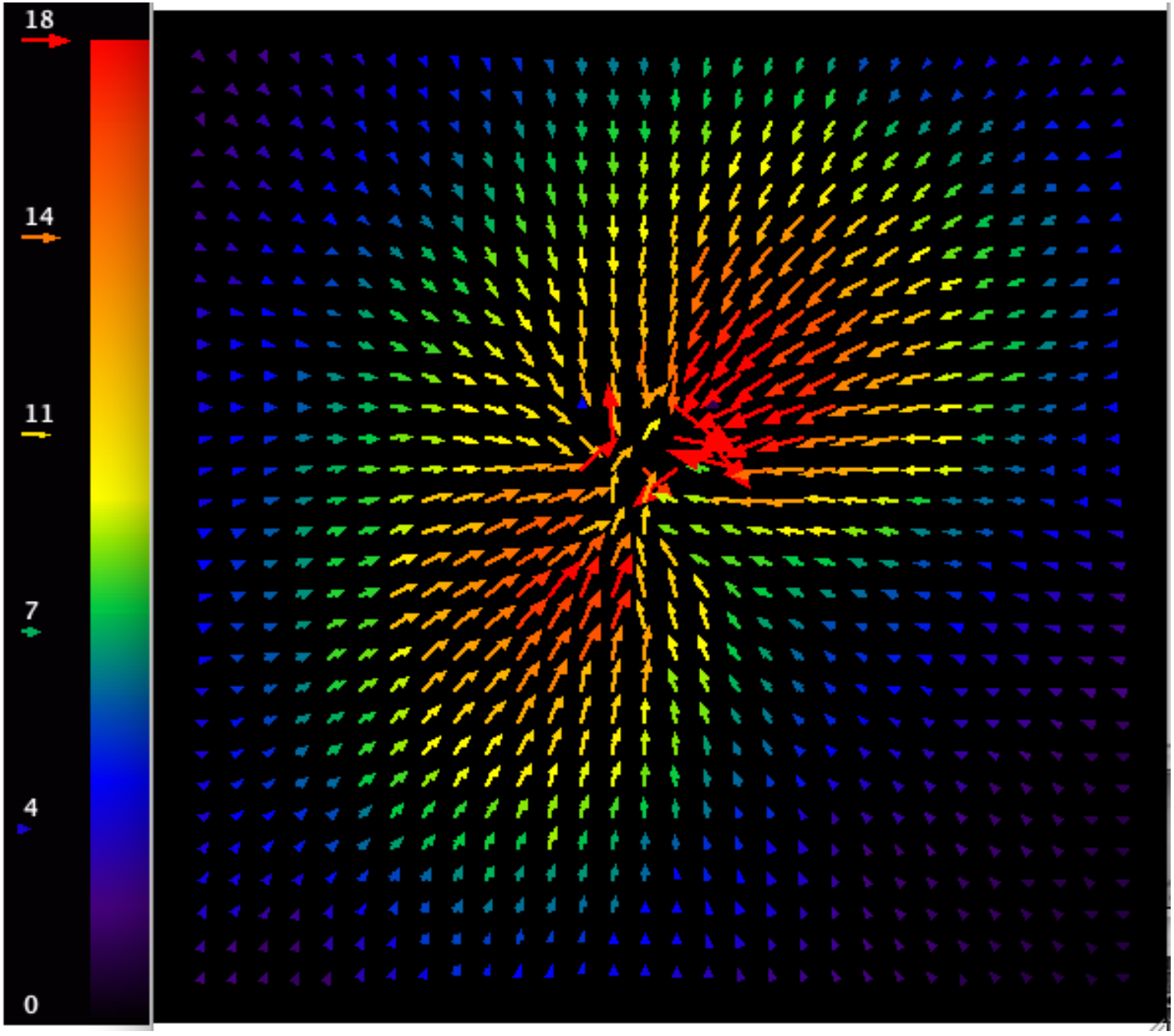
Traction force microscopy

- Gels seeded with beads and imaged at various time points during experiments

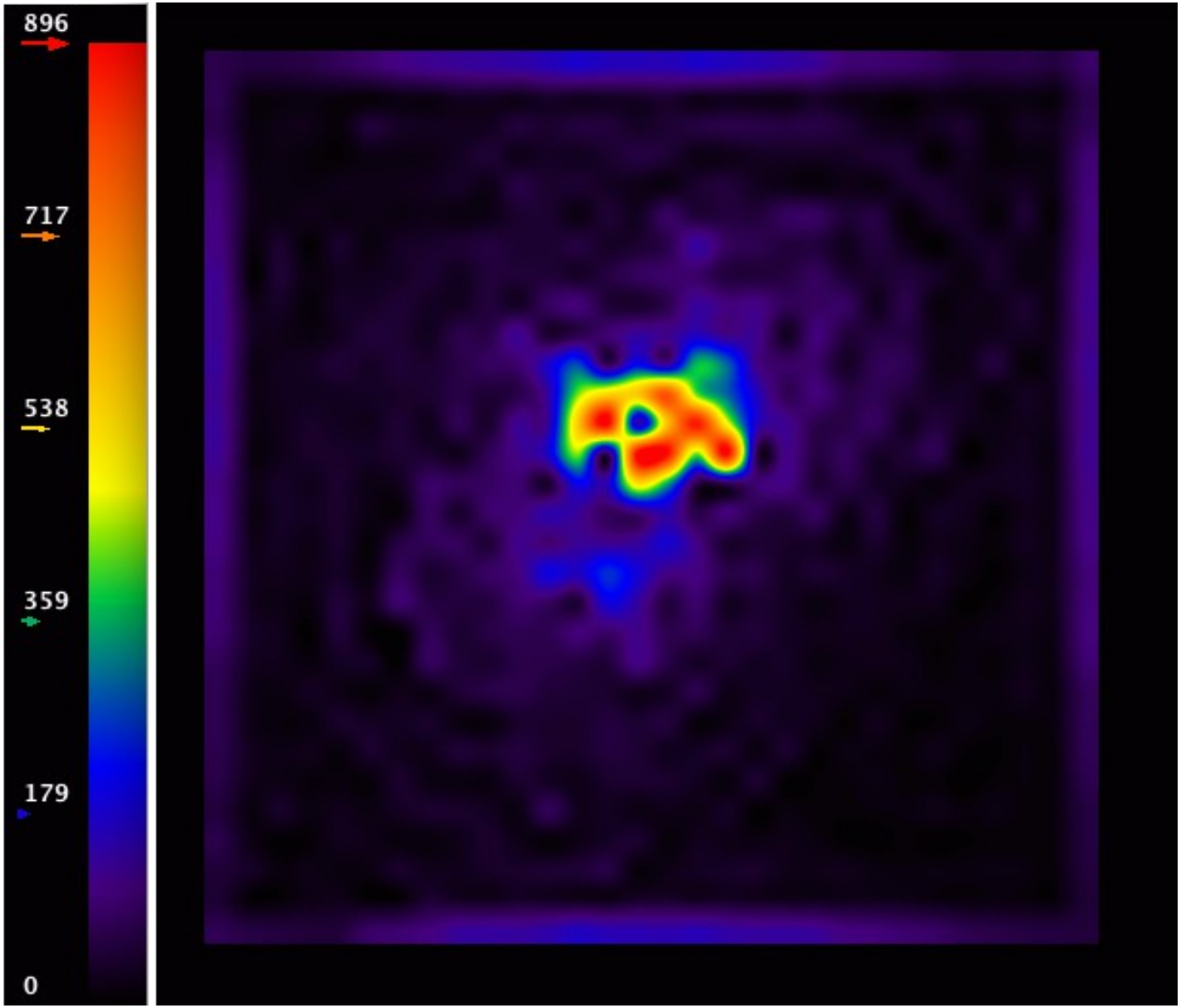
- Using a technique developed in fluid mechanics to process particle image velocimetry (PIV) data, we can track the bead motion in time and determine the strain field from the images

- Assuming a stress-strain relationship, we can then compute the stress field

Displacement field



Stress field



Comparison to simulations

- Rudnicki *et al*. (2013) recently developed a finite element model of cell traction on fibrous gels

- Using this technique, we can compare our simulated stress and strain fields with the measured ones to solve the inverse problem and deduce the mechanical properties of the gels

Summary

- We proposed three approaches to study the mechanical properties of collagen gels subject to boundaries and the presence of an adhered cell
  - Indentation --> Mechanical properties of gel with no cell
    --> E changes with boundary size and collagen conc.
  - Confocal --> Collagen intensity as function of radial position
    --> Need to relate intensity with concentration
  - TFM --> Strain and stress fields as function of time
    --> Simulations of cell traction could deduce props.

Conclusions

- We have used a Hertz contact model to calculate compressive moduli for collagen gels (under the assumption of linear elasticity) from indentation measurements

- We have created a finite element model of gel indentation, which can be used to study the impact of changing model inputs on the deformation of the gels subject to indentation
  - This model can be extended to simulate traction forces exerted on the gels by the cells

Conclusions

- We have identified a need to correlate collagen intensity, measured from confocal microscopy images, with collagen concentration
  - By identifying this correlation, we will be able to calculate spatial variation of collagen contraction within the gel, which we can correlate with spatial changes in the effective compressive modulus

- Using available plug-in for ImageJ, we calculated the in-plane gel displacement field during deformation by the cells, and by assuming linear elastic material properties, we calculated the corresponding stress field
  - This approach could be extended to a spatially-varying linear elastic material model

Future work

- If linear elasticity insufficient to explain mechanical behaviour, extend to non-linear elasticity, visco-elasticity, elasto-plasticity, etc.
  - Need some deviation from pure elasticity to account for compaction of gel by cell

- Radial symmetry assumed
  - Look at changes in fibre alignment and deviations from radial symmetry