USC Viterbi School of Engineering

Characterizing GelMA Stiffness for 3D Tumor-Microenvironment Studies

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Introduction

Engineered Tumor Models

- In Dr. Shen's lab, researchers look to study the interaction between tumors and their microenvironment using engineered models.

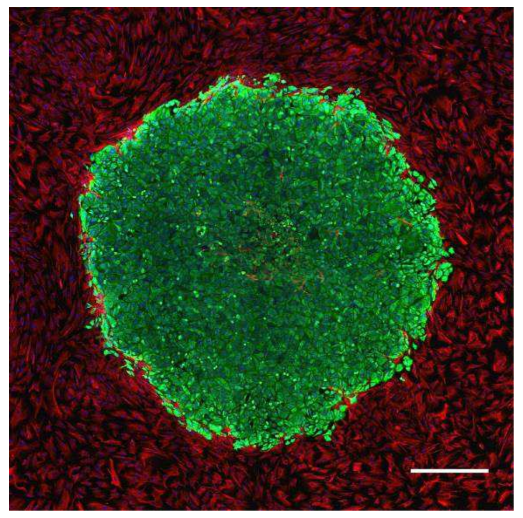

Figure 1. 2D patterning of cancer (green) and stromal cells (red) P.C.: Peter Ta

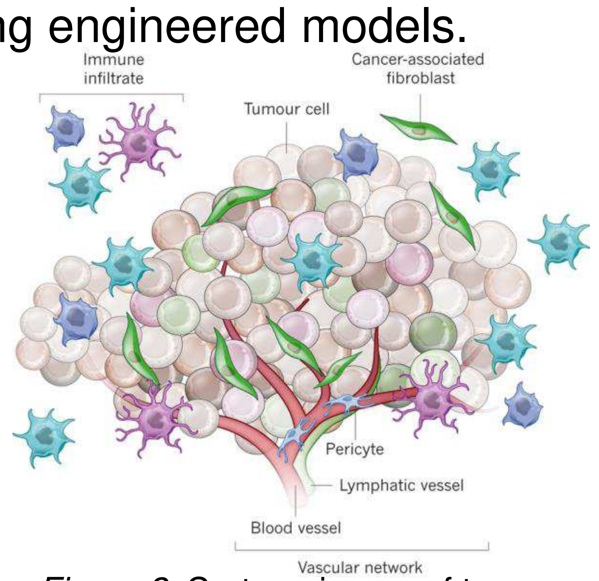

Figure 2. Cartoon image of tumor microenvironment. P.C.: Nature

- Our lab mainly utilizes 2D models, but 3D models are more representative of the *in vivo* setting.

Hydrogel Use in Culturing

- Hydrogels (highly cross-linked polymer networks) serve as the scaffolding material for these 3D models.
- Gelatin methacrylate (GelMA) is a highly customizable hydrogel that interacts well with cells and is biodegradable, making it ideal for culturing.

Our goal is to characterize GelMA hydrogels to create 3D cultures that effectively replicate the *in vivo* environment.

Methods

GelMA Synthesis:

Dissolve gelatin 10% (w/v) at 60° C. → Add methacrylic anhydride → Dilute 2x with DPBS at 40° C.

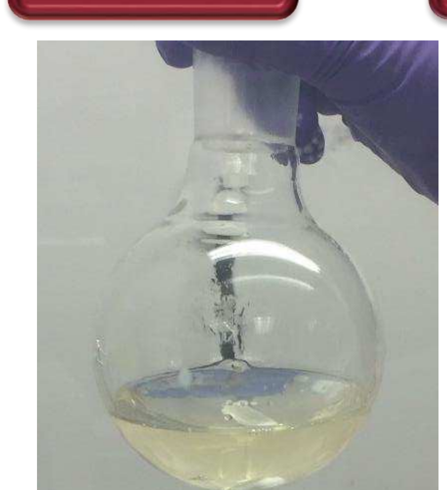

Figure 3. An image of gelatin and PBS solution after mixing.

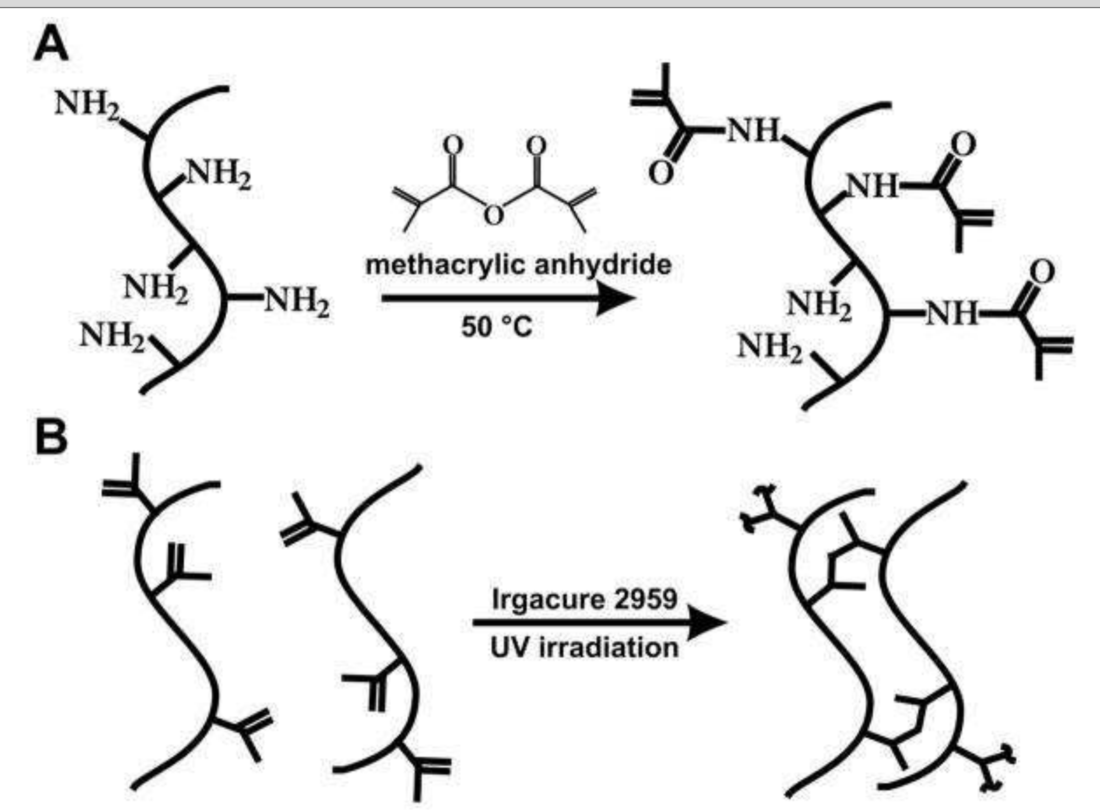

Figure 4. Binding of methacrylic anhydride to the gelatin and subsequent crosslinking by UV light creating final version of gel. P.C.: Nichol et al., 2010.

Dialyze GelMA for 1 week at 40°C. → Lyophilize for 1 week at -80°C. → Create different GelMA w/v ratios → Crosslink using UV light.

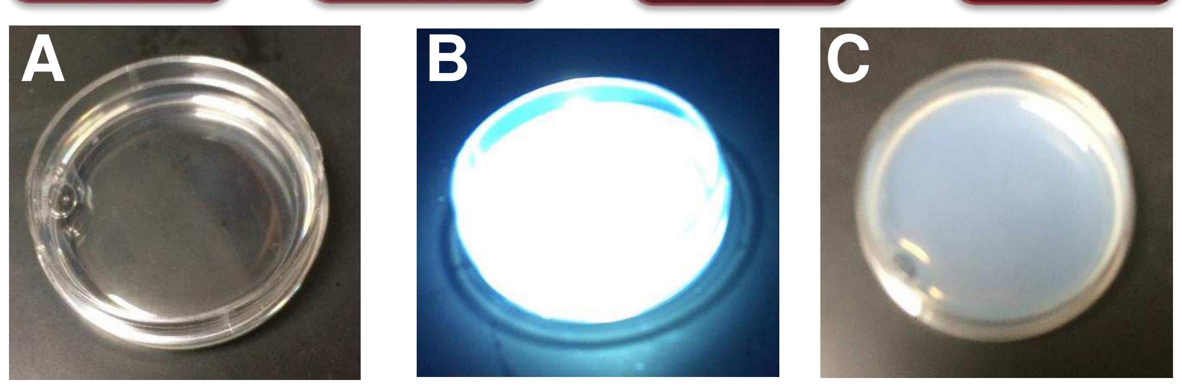

Figure 5. Process of crosslinking GelMA. (A) GelMA in pre-polymer solution. (B) Exposure to UV radiation. (C) Crosslinked GelMA hydrogel.

Mechanical Testing

Biopsy punch disks of GelMA for each formulation. → Measure height and diameter of each of the disks. → Mechanically test disks using Instron machine.

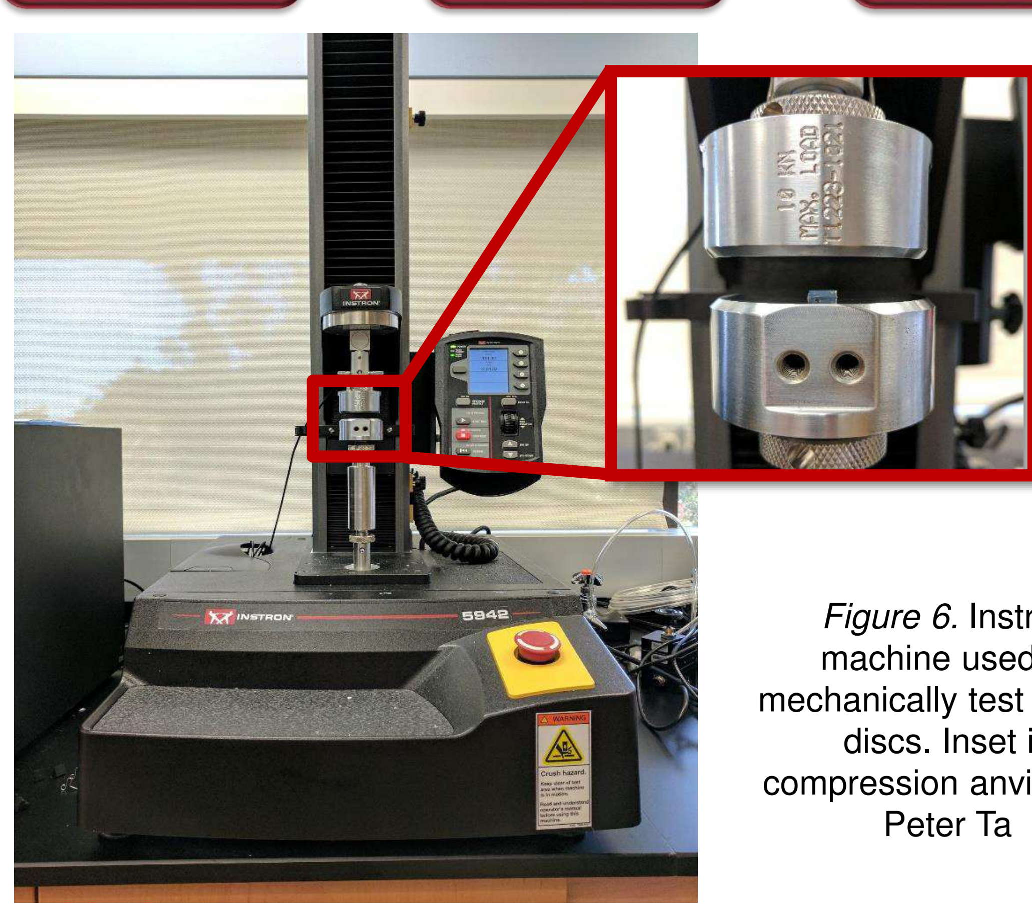

Figure 6. Instron machine used to mechanically test GelMA discs. Inset is compression anvil. P.C.: Peter Ta

Results and Discussion

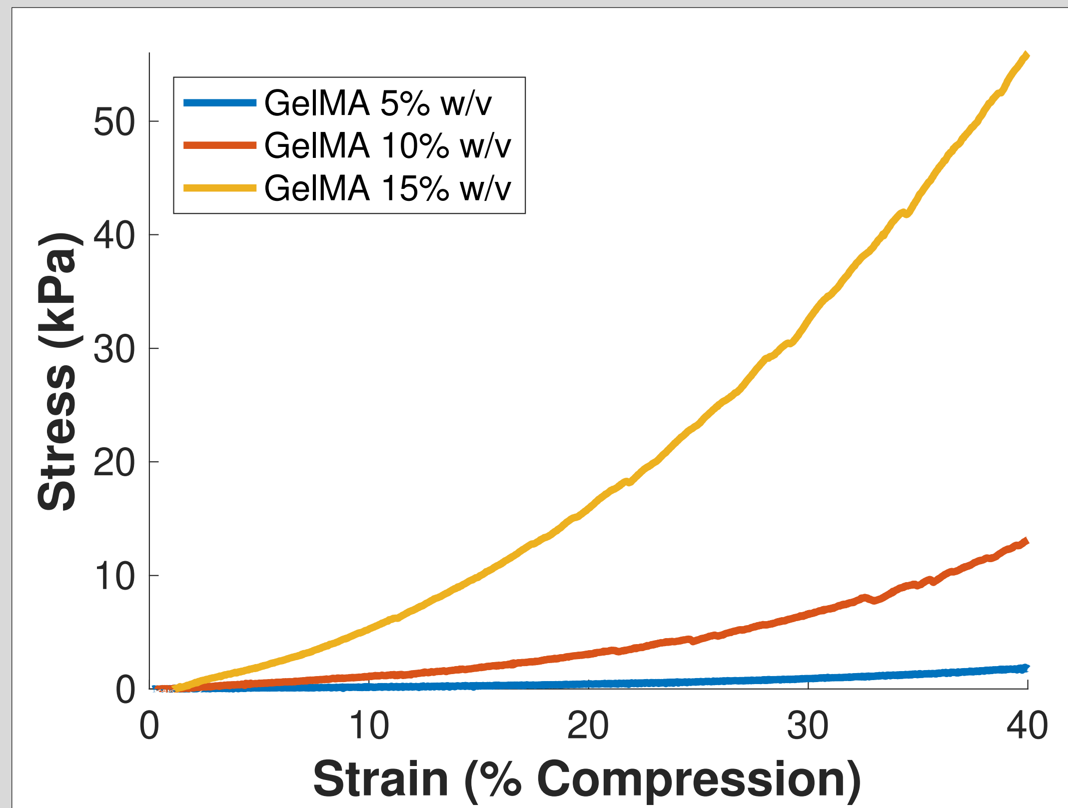

Figure 7. Stress vs Strain curve for different w/v ratios of the 8% degree of methacrylation batch.

- As strain increases, need much more force to continue compression of disks.
- Higher w/v ratios have steeper curvature.
- Linear range was from 0 to 10% strain.

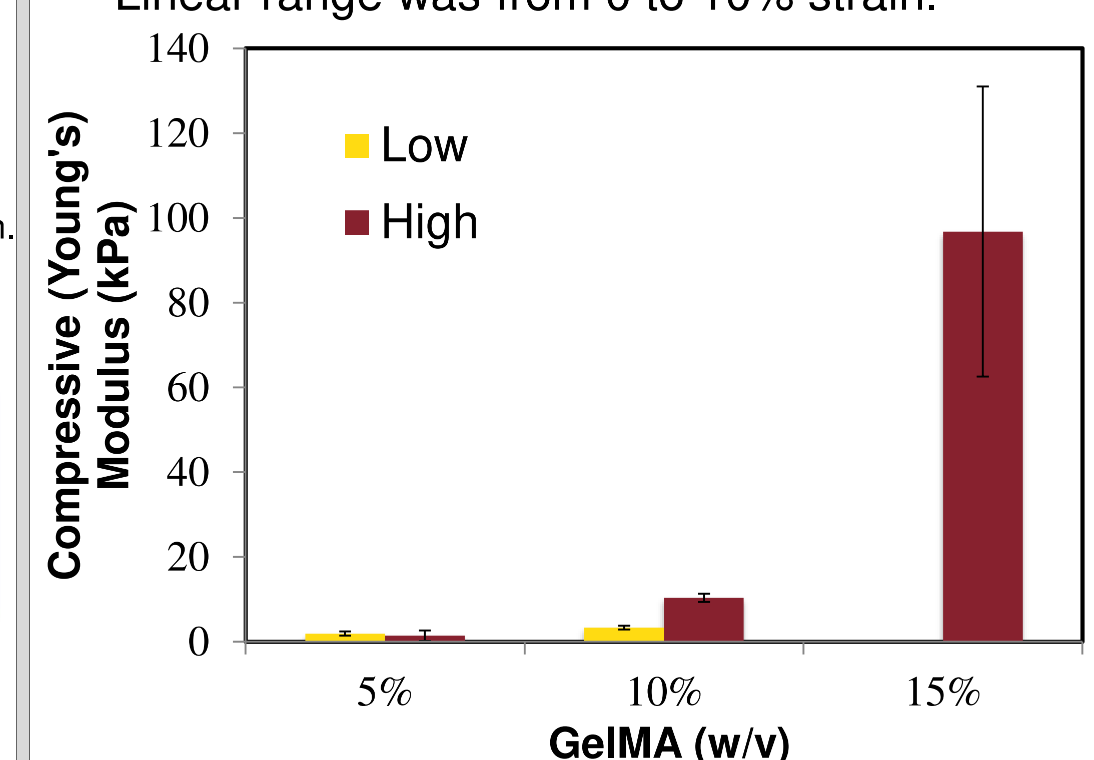

Figure 8. Compressive modulus for the different GelMA formulations. Note: we were unable to test 15% w/v of 0.25% degree of methacrylation because it did not properly crosslink.

- For each degree of methacrylation, average value of compressive modulus increases as w/v ratio increases.
- High methacrylation groups have higher average values of compressive modulus in comparison to low degree counterparts.
- Exception: 5% w/v group.
- Here, the average value for the high degree of methacrylation is actually lower than the value for the low degree group.

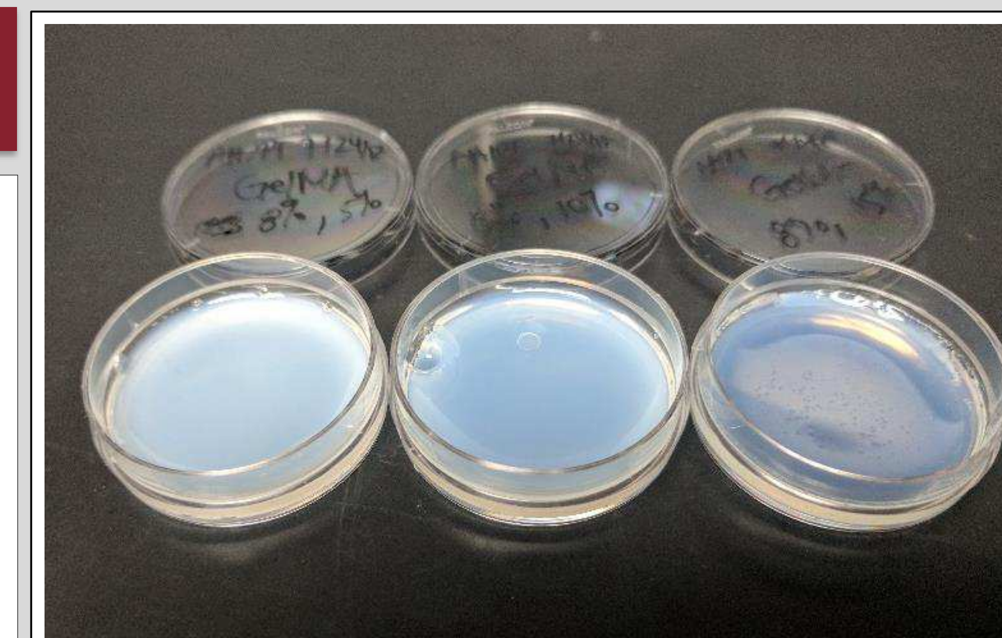

Figure 9. Different w/v samples of 8% methacrylation degree.

Summary

- Greater w/v → greater stiffness.
- Greater methacrylation → greater stiffness.
- Broad possible ranges of stiffness

Future Steps

- Culture different cell types onto differing GelMA hydrogels to optimize stiffness for the cell types.
- Also, could calculate true degree of methacrylation in the crosslinked samples.

References

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