Mechanically Tunable Hydrogel-Based Clearance Assay for Ovarian Cancer Modeling

Madilynn Clark, Trinity Devanney, Meghan Herlihy, Dr. Linqing Li

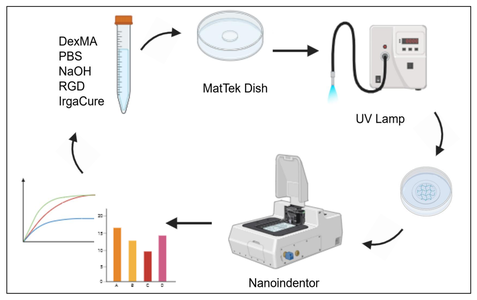
Department of Chemical Engineering and Bioengineering, University of New Hampshire

Introduction

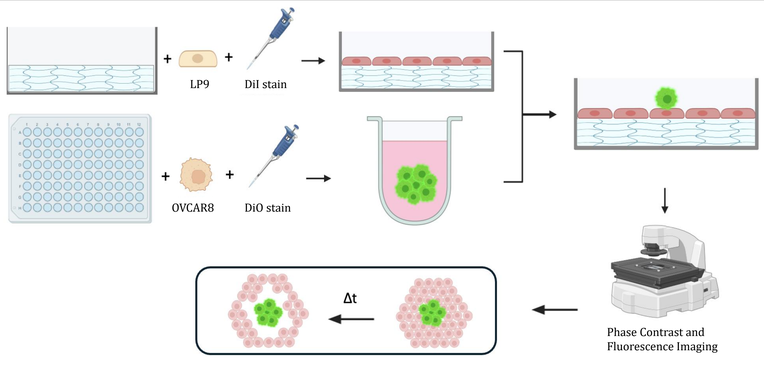
- Metastasis is the leading cause of cancer-related deaths and treatment failures. Many ovarian cancer patients present with metastasis, which complicates treatment and reduces the likelihood of successful outcomes (1).
- Drug discovery and development remain inefficient due to a lack of biologically relevant disease models that accurately predict clinical outcomes. Many drugs show promising results in lab testing but fail in animal or clinical trial phases (2).
- To bridge the gap between *in vitro* and *in vivo* testing, we modified traditional clearance assays by integrating a tunable Dextran Methacrylate (DexMA) hydrogel model that replicates the mechanical properties of native tissues.

Hydrogel Synthesis and Optimization

- 54% DexMA was synthesized from Dextran and Glycidyl Methacrylate.
- Hydrogels were optimized by UV crosslinking 30 mg/ml DexMA with PBS, RGD, NaOH, and Irgacure.
- Gel mechanical stiffnesses were determined with nanoindentation.



Clearance Assay Setup



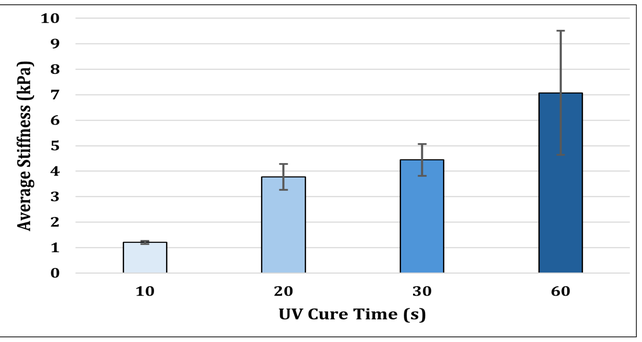
Stained LP9 cells were seeded onto the hydrogel, while stained OVCAR8 cells were plated onto a poly-HEMA 3-D culture plate. Once a full monolayer formed, spheroids were placed on top and clearance was evaluated.

Biologically relevant stiffness of 30 mg/ml DexMA Hydrogels

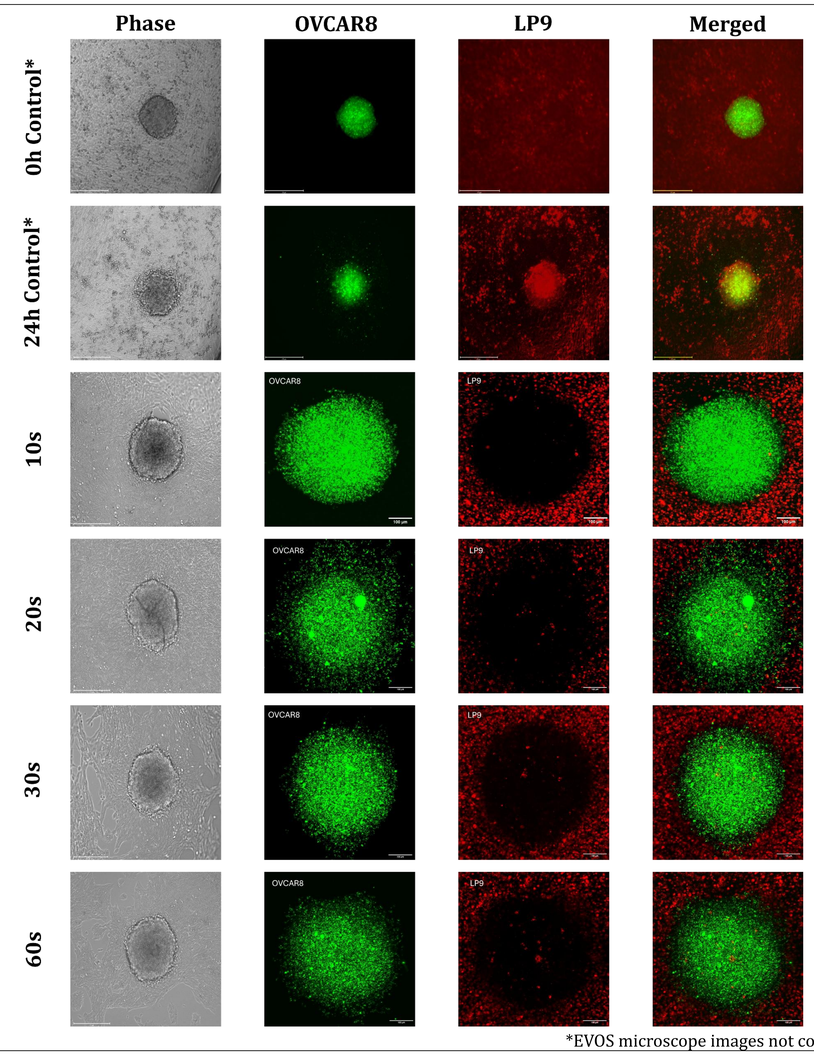
In vivo stiffnesses (3)***:***

- Ovaries: 0.8-5.8 kPa
- Uterus: 4.73-5.61 kPa
- Liver: 4-6.5 kPa
- Lungs: 1.96 kPa
- Intestines: 1-3 kPa

Goal hydrogel stiffness range 1-10 kPa.

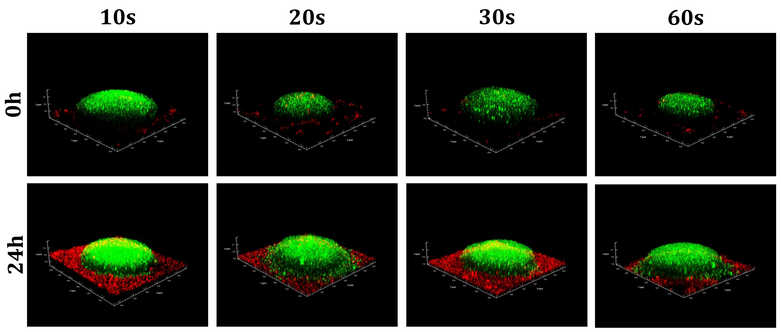


Mesothelial clearance by cancer spheroid



*EVOS microscope images not confocal

Spheroid flattening shows invasion potential



- 3D Volumetric view of clearance assay on hydrogels.
- Spheroid flattening and spreading by 24hr indicates focal adhesions to RGD in DexMA hydrogels.

Conclusions and Future Directions

Conclusions

- Created hydrogels with biologically relevant stiffnesses using DexMA.
- Successfully created an LP9 monolayer on hydrogel model to mimic the *in vivo* human peritoneal cavity lining.
- Successfully created OVCAR8 spheroids to mimic *in vivo* metastatic tumors.
- Combined OVCAR8 spheroids with the LP9 monolayer to create a mesothelial clearance assay.
- Control model showed clearance underneath and around the spheroid.
- Gel models showed clearance in the area underneath the spheroid, but minimal clearance in the surrounding area.
- The softest (10s) gel showed the least amount of clearance.

Future Directions

- Use cell-degradable gel instead of DexMA to evaluate invasion and clearance.
- Application of the hydrogel model for cancer drug testing.

Acknowledgements

Thank you to Dr. Linqing Li, Jaxson Libby and Dr. Sarah Walker for their assistance and guidance throughout this project. Thank you to the CHBE department for funding our senior design project.

References

1. Qian, C.-N., Mei, Y., & Zhang, J. Cancer metastasis: issues and challenges. *Chinese Journal of Cancer*, 2017.
2. Loewa, A., Feng, J. J., & Hedtrich, S. Human disease models in drug development. *Nature reviews bioengineering*, 2023.
3. Guimares, C. F., Gasperini, L., Marques, A.P., Reis, R. L. The stiffness of living tissues and its implications for their engineering., 2020.
4. Kloeckner, A. A., & Walker, S. R. Endometriosis Cell Spheroids Undergo Mesothelial Clearance in a Similar Manner to Ovarian Cancer Cell Spheroids. *Cells*, 2025.