In situ crosslinked oxidized hyaluronic acid-based hydrogels for soft tissue engineering

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Introduction

External forces, particularly those associated with traumatic brain injury, can result in tissue-level damage [1]. Meanwhile, mechanical cues on a smaller scale are pivotal in shaping the development, behavior, and function of individual neural cells [2]. Understanding injury and disease mechanisms within the central nervous system (CNS) requires grasping the influence of cellular forces and tissue mechanics [3]. This study examines a soft tissue engineering method using hydrogels made from oxidized hyaluronic acid (OHA) and gelatin, crosslinked with microbial transglutaminase (mTG). This approach mimics the neuronal extracellular matrix and creates a three-dimensional (3D) platform for studying how cells respond to mechanical changes. We will explore how different hydrogel compositions influence neuron growth and behavior, particularly in relation to matrix stiffness.

Material and Methods

- OHA was synthesized by oxidizing hyaluronic acid with sodium meta periodate
- Crosslinking stability is assisted by enzymatic crosslinking of GEL
- Hydrogel compositions are investigated regarding mechanical properties/porosity/stability/ degradation
- Neuronal cell seeding with primary E18 cortical neurons for testing cell compatibility and suitability for neuronal TE

Swelling/Degradation

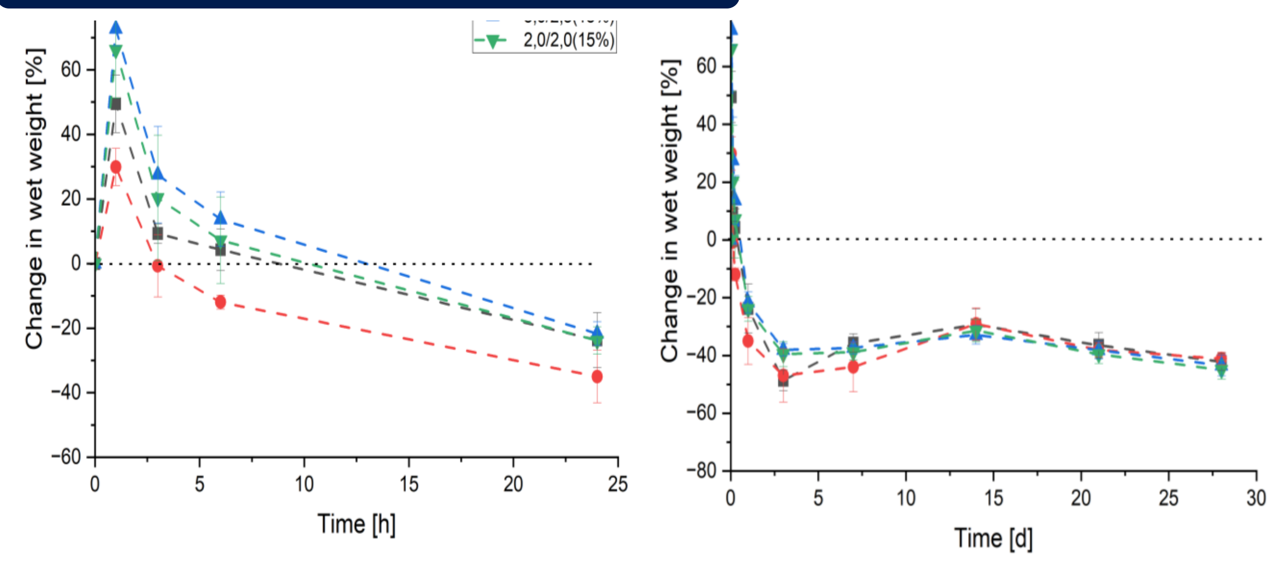

Figure 1: Swelling/ degradation over 28 days under cell culture conditions. Left:24 h. Right: 28 d

- Strong swelling behavior in the first hours for all hydrogels
- Weight loss within the first 3 days, stable behavior afterwards

Mechanical testing

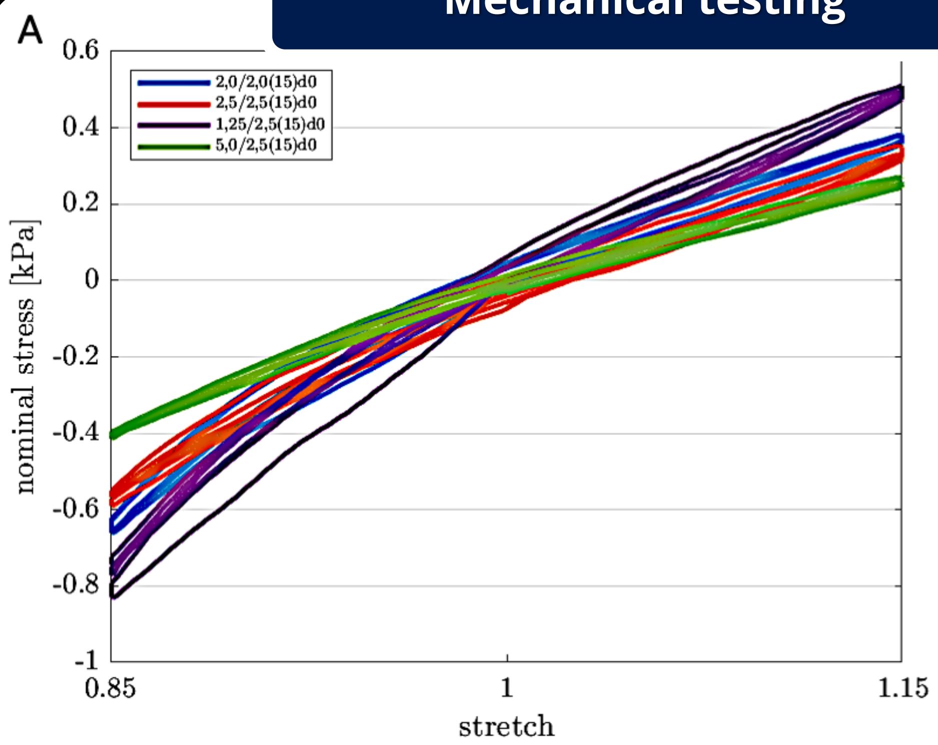

Figure 2: cyclic compression tension up to 15 % stretch / compression

- All tested hydrogel compositions showed mechanical properties in the range of physiological conditions
- comp. tens. asymmetry like brain tissue

Cell seeding

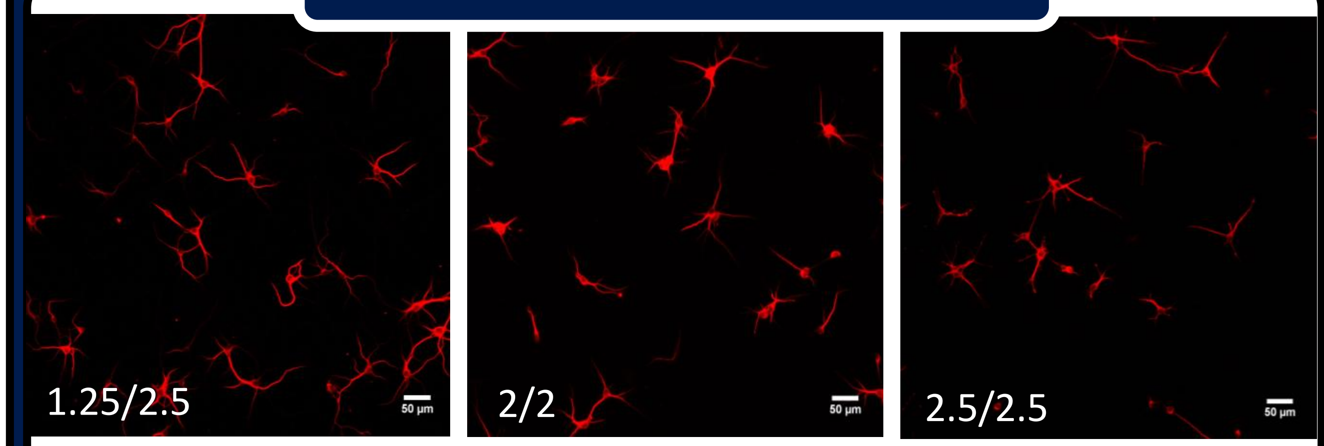

Figure 4: Primary neurons after 7 days *in vitro*, stained with β-tubulin. Scale bar is 50 µm.

- Primary E18 cortical neurons seeded on top of the hydrogels
- Neurons survived and developed over 7 days *in vitro*
- Best cell development and survival rate on the hydrogel with lowest OHA content

Conclusions

Four OHA/GEL hydrogels were developed to explore variations in mechanical properties. Among them, three compositions (1.25/2.5, 2/2, and 2.5/2.5 OHA/GEL with 15% mTG) were identified as promising for neuronal tissue due to their stiffness and stability. Multimodal analysis confirmed the hydrogels' stability and high cell viability. 1.25/2.5 OHA/GEL demonstrated the best neuron development with an ideal balance of stiffness and stability.

Acknowledgements

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References

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