AGAROSE-BASED HYDROGELS AS MODEL FOR MECHANOBIOLOGY

UNIVERSITÀ DEGLI STUDI DI TRIESTE

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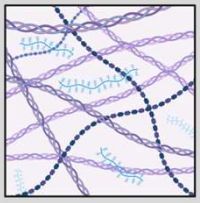
INTRODUCTION

The extracellular matrix (ECM) is a highly complex and dynamic material, essential for maintaining tissue structure and functions.

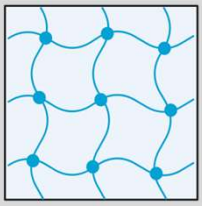
Cells continuously sense and respond to the mechanical cues of the ECM and are exposed to forces such as shear stress (blood vessels) and compression (cartilage and bone).

This cellular sensitivity is harnessed in mechanobiology, where biomaterials are designed to have specific mechanical properties to mimic the native ECM and steer cells behaviour.

Extracellular matrix



Polymer network



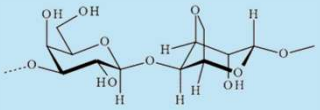
✓ Biocompatible

✓ Similar mechanical properties

✗ Lack of binding sites for cells

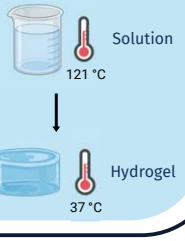
AIM OF THE PROJECT

The main aim of this project is to present agarose as optimal candidate for an easy and rapid fabrication of dynamic natural hydrogels with tunable mechanical properties to use as substrates for mechanobiological investigations.



Agarobiose
(D-galactose and 3,6-anhydro-Lgalactose)

Thermoresponsive

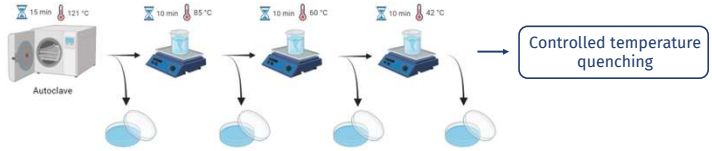


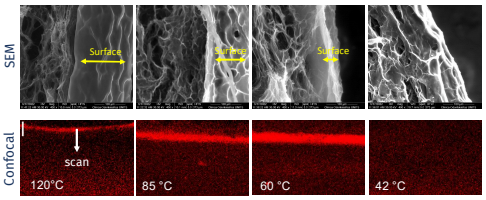
RESULTS



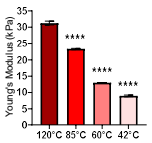
Temperature

The major impact of quenching is observed on the hydrogel surface: a clear agarose film was identified on the surface of the gel at the higher temperature (121 °C), which gradually disappeared as the curing temperature decreased. Consistently, the lower the curing temperature, the lower the stiffness of surface.



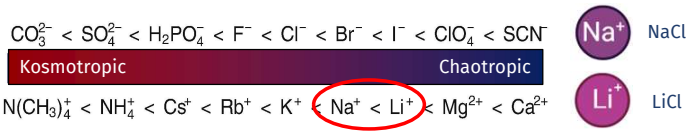


Surface stiffness



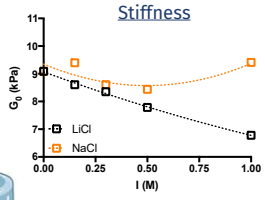
The ionic strength of the solvent can influence the mechanical and structural properties of agarose hydrogels. Using kosmotropic (NaCl) and chaotropic (LiCl) salts, we observed a decrease in shear modulus with the increase of the ionic strength. The linear elasticity is also affected and increases with increasing salt concentration.

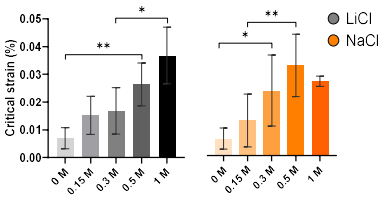
Ionic strength



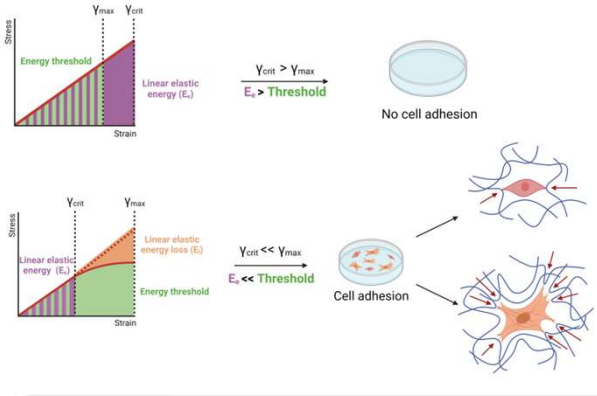
Viscoplasticity

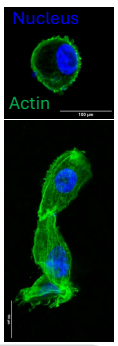
Stiffness



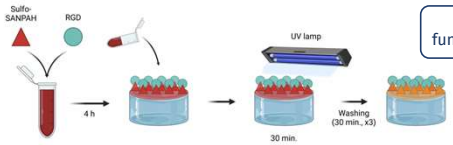


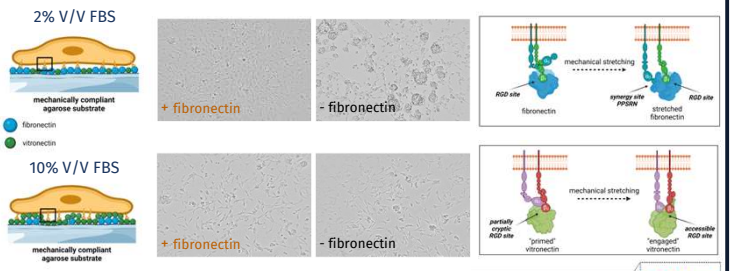
Agarose hydrogel

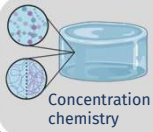




Surface functionalization



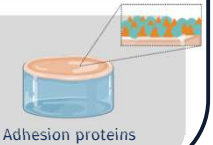




Concentration and chemistry

Using agaroses with different chemical structures at different concentrations it is possible to fine tune the linear elastic energy of the hydrogels. With these substrates, we were able to determine a threshold energy value above which cell adhesion is impaired. Below this threshold, cell growth is promoted. The plasticity of the material was also considered by the energy loss in the nonlinear region.

Hydrogel surface was covalently bound to an RGD-containing protein. RGD was able to improve cell adhesion at a low FBS concentration (2% V/V), but its effect appeared to be concealed when using higher FBS concentration in the medium (10% V/V). This happen because FBS proteins (vitronectin), coat upon the functionalized RGD, hiding it from the cells.



Adhesion proteins

CONCLUSIONS

Agarose displays several key advantages including its ease of gel preparation and the possibility to fine-tune its mechanical properties by different parameters such as temperature, ionic strength, polymer structure and concentration.

Overall, the results obtained show that both viscoelastic and plastic properties of hydrogels play a crucial role in regulating cellular activities, including adhesion, spreading and remodeling.

ACKNOWLEDGEMENTS

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