Engineering thymus viscoelasticity and microstructure integrating chaotropic salts in gels

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Motivation

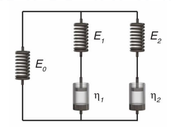
The *thymus extracellular matrix* is crucial for the development and selection of T-cells [1], [2]. However, *3D scaffolds* fabricated with natural/synthetic polymers are currently of limited relevance due to the absence of **thymus-specific micro-structural and mechanical cues** [3]. Indeed, to date, the mechanical properties of this organ have been poorly investigated and its viscoelastic behavior is still unknown [4]. This work aims to tackle these issues: from a previous thorough structural and viscoelastic characterization of the bovine thymus, **thymus-mimetic materials were designed and fabricated** and represent the first successful attempt to reproduce thymic mechanostructure in vitro**.**

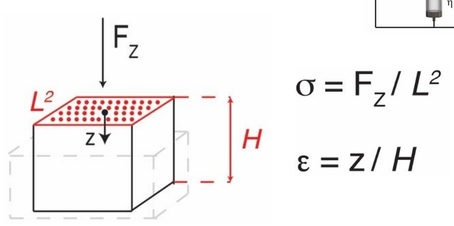
Mechanical characterization[5]

Mach-1 V500CSS (Biomomentum Inc., Montreal, Canada)

Bulk stress-relaxation tests

Generalized Maxwell model

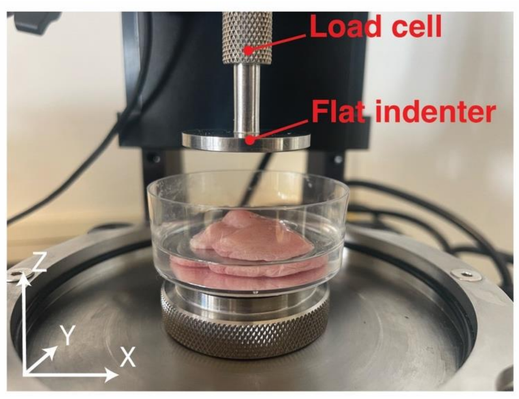




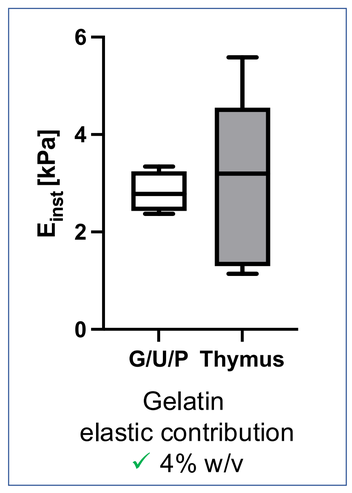
$$\sigma = F_Z / L^2$$

$$\varepsilon = z / H$$

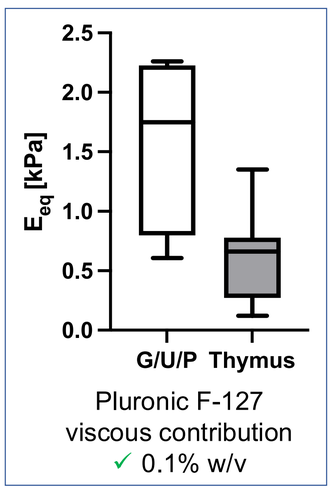
Thymus
from Bovine



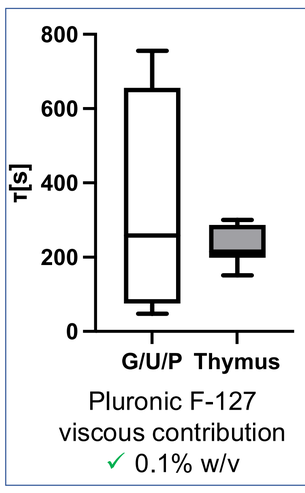
Input stimulus: strain step;
Indenter strain rate = 1 Hz;
Strain amplitude = 20%;
Duration = 900 s.



Gelatin
elastic contribution
✓ 4% w/v



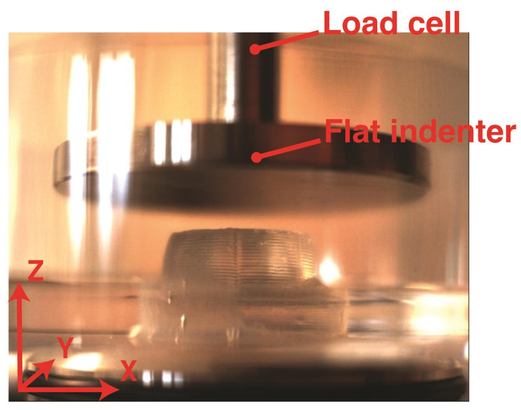
Pluronic F-127
viscous contribution
✓ 0.1% w/v



Pluronic F-127
viscous contribution
✓ 0.1% w/v

Mann-Whitney (GraphPad Prism 10), $N = 4$

In vitro model
Gelatin/Urea/Pluronic F-127 (G/U/P)

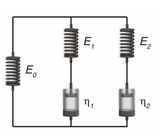


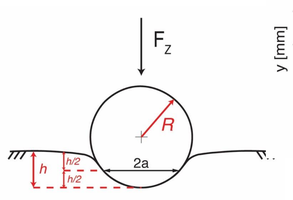
Input stimulus: strain step;
Indenter strain rate = 1 Hz;
Strain amplitude = 20%;
Duration = 900 s.

Still to replicate *in vitro*

Indentation stress-relaxation tests [6]

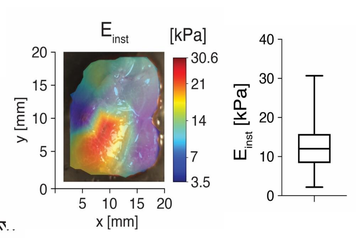
Generalized Maxwell model

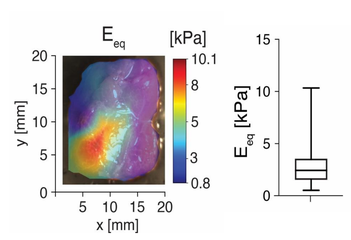


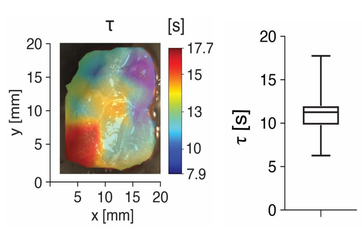


$$\sigma = \frac{F_Z}{R\sqrt{hR}}$$

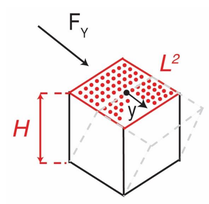
$$\varepsilon = \frac{4}{3(1-\nu^2)}\frac{h}{R}$$





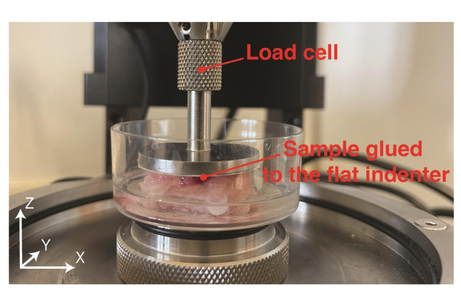


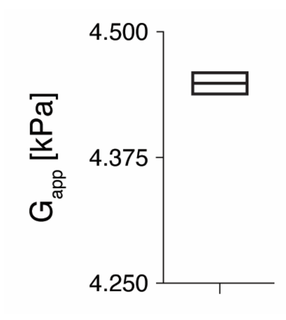
Shear tests



$$\sigma = F_Y / L^2$$

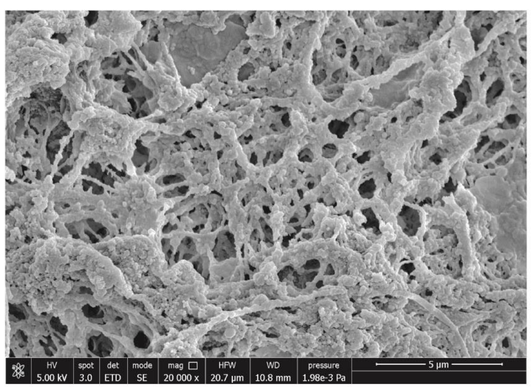
$$\varepsilon = y / H$$



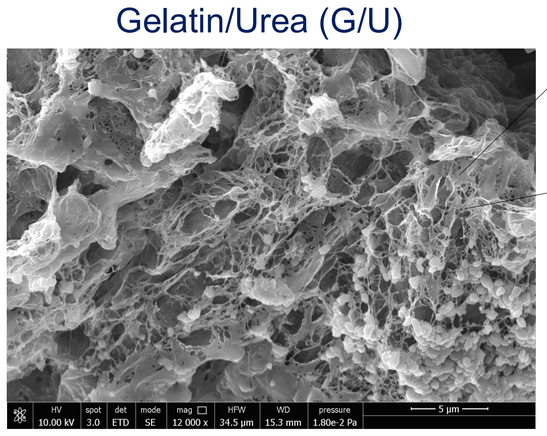


Structural characterization

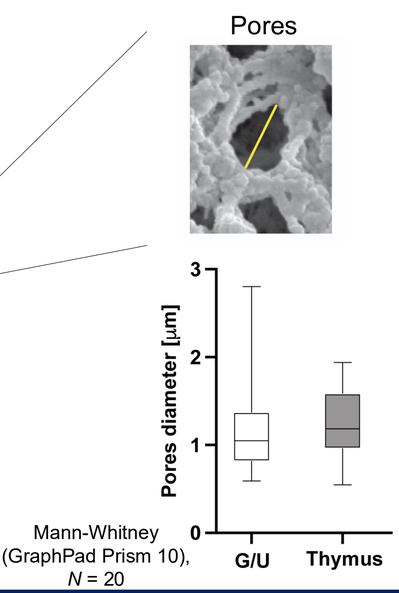
Thymus
from Bovine



In vitro model
Gelatin/Urea (G/U)



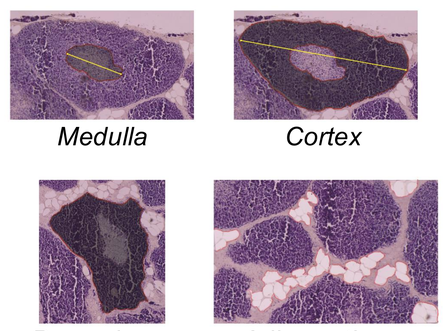
Urea contribution (Hofmeister effect)
✓ 4% w/v



Mann-Whitney (GraphPad Prism 10), $N = 20$

Next steps

- *Merging mechanical and structural tuning for complete hydrogel optimization*
- *Biological testing of thymic relevant cell lines seeded on the hydrogel*
- *Rigorous thymic niche replication from histological images parameters quantification*



References

[1] M. Duah et al, Front. Immunol., 12:1-17, 2021.
[2] V. P. Shichkin et al, Front. Immunol., 13:1-18, 2022.
[3] H. Sharma et al, Adv. Sci., 8:1-22, 2021.
[4] C. S. Silva et al, Adv. Healthc. Mater., 10:20, 2021.
[5] L. Cacopardo et al, Tissue Eng. Part B Rev., 28:912-925, 2022.
[6] G. Mattei et al, J. Mech. Behav. Biomed. Mater., 50:150-159, 2015.

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