Supplementary Information

Investigating the interplay between substrate stiffness and ligand chemistry in directing mesenchymal stem cell differentiation within 3D macro-porous substrates

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Computational model material parameters and validation

For the purpose of our model, the substrate was assumed to behave as an isotropic, almost incompressible ($\nu = 0.49$), linear elastic material ($E_M = 1$ MPa), with no time-dependent properties. The properties of the embedding medium were assigned based on the uniaxial response of the porous substrate. This was simulated computationally, and it was determined that the modulus of the porous substrate (E_P) was 132.1 kPa. This is comparable to a modulus of 160 kPa predicted by Gibson-Ashby model of cellular solids.[1] As a validation step, we compared the compressive modulus of the porous substrates fabricated in this study to the values predicted by the computational model. Mechanical testing of porous substrates (10 wt.% ELP, 0.5:1 Reactive Groups:Amine Groups) revealed that the modulus of the porous hydrogels was 1.93 kPa which compares well with the Modulus 2.11 kPa predicted by our model, indicating that our model accurately captures the mechanical behavior of the porous substrates utilized in this study (Figure

S1). The resulting local stiffness values (k_L, N/m) output by the model are based on a substrate with a material modulus (E_M) of 1 MPa. For the 1 MPa substrate, the local stiffness ranged from 1033-1447 pN/nm in shear and 2218-2685 pN/nm in a normal orientation (Table S1). However, as k_L can be assumed to scale linearly with E_M, the local stiffness of substrates with different material modulus values can be easily extrapolated for the range of stiffness' used in this study.

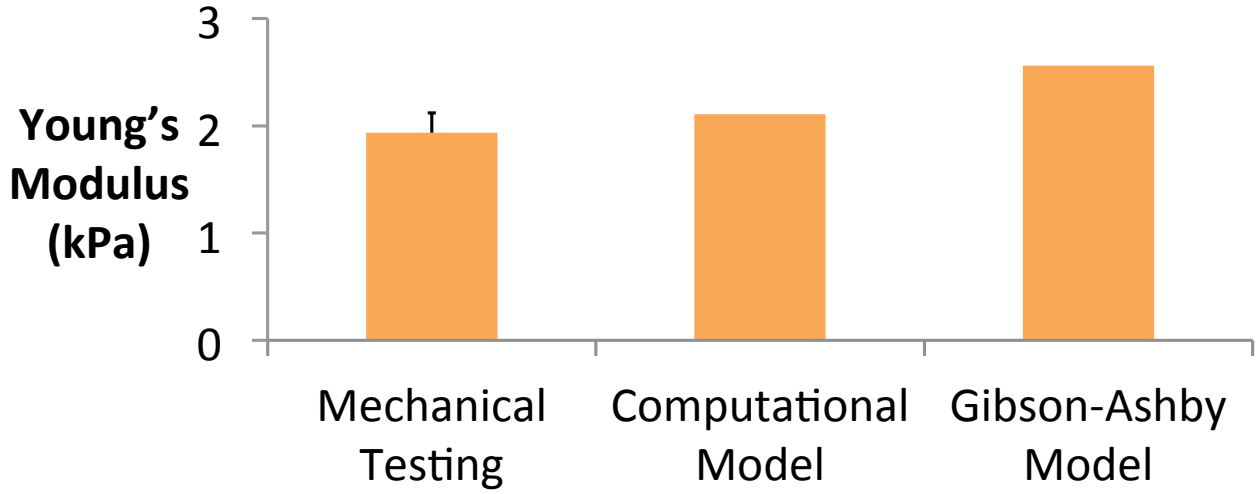

Figure S1. A comparison of the Young's modulus of the porous substrates fabricated in this study (10 wt.% ELP, 0.5:1 Reactive Groups:Amine Groups) to the values predicted by our computational model and the Gibson-Ashby model of cellular solids.

Material Modulus (E_M), MPa	Porous Substrate Modulus (E_P), kPa	Median Local Equivalent Modulus (E_{eq}), kPa		Median Local Stiffness (k_L), pN/nm	
		Shear	Normal	Shear	Normal
1	132.1	925.5	1785.5	1292.2	2493.0

Table S1. The results of the computational model of a porous substrate with a material modulus of 1 MPa.

Supplementary References

1. Gibson, L. J. & Ashby, M. F. *Cellular solids: structures & properties*. (Pergamon Press, 1997).