

ELECTRONIC SUPPLEMENTARY INFORMATION

Stem cell secretome-rich nanoclay hydrogel: a dual action therapy for cardiovascular regeneration

Renaë Waters,^a Settimio Pacelli,^a Ryan Maloney,^a Indrani Medhi,^b Rafeeq P.H. Ahmed,^c and Arghya Paul^{*a}

^a Biointel Research Laboratory, Department of Chemical and Petroleum Engineering, Bioengineering Graduate Program, School of Engineering, University of Kansas, Lawrence, KS, USA.

^b SRM University, Kattankulathur 603203, Tamilnadu, India

^c Department of Pathology, University of Cincinnati, 231-Albert Sabin Way, Cincinnati 45267, OH, USA.

Reprint requests and correspondence to: arghyapaul:arghyapaul@ku.edu

Experimental procedures:

Scanning Electron Microscopy (SEM). Hydrogels in fully swollen state were frozen in nitrogen liquid and lyophilized afterwards. Subsequently, the lyophilized samples were cut to expose their cross-sections and coated with gold using a sputter coater. The detailed structure of the sample cross-sections was imaged using an SEM (Hitachi Model S4700, Japan).

Swelling studies: Swelling studies were performed on freeze-dried nanocomposite hydrogels in phosphate buffer (PBS, pH 7.4, ionic strength $I = 0.1$). Aliquots (20 mg) of the gels were placed in the buffer and allowed to swell at $37.0 \pm 0.1^\circ\text{C}$. After defined intervals of time the hydrogels were weighed and placed back in the media. The results were expressed as swelling ratio over time according to this formula:

$$\text{Swelling ratio (\%)} = ((W_s - W_d) / W_d) \times 100 \quad [\text{Eq 1}]$$

where W_s and W_d are the weights of the swollen and dry hydrogels, respectively. Each experiment was carried out using 4 sample for each group.

Supplementary Data.

In order to investigate the influence of Laponite on the network structure, SEM images were obtained for the different groups.

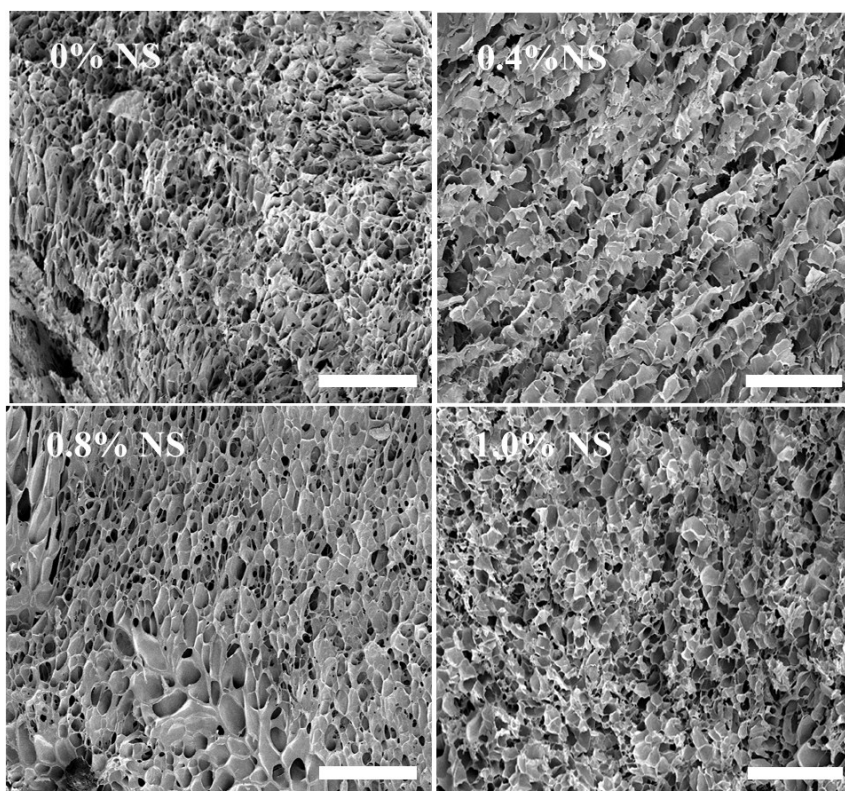


Fig S1. SEM images of Gel-MA nanocomposite hydrogels having different concentration of Laponite Scale bar: 100 μm .

No significant difference in the microstructure was observed in each of the gels. The porosity was mainly dictated by the degree of crosslinking of the polymeric network, which was not influenced by the concentration of Laponite introduced during their preparation.

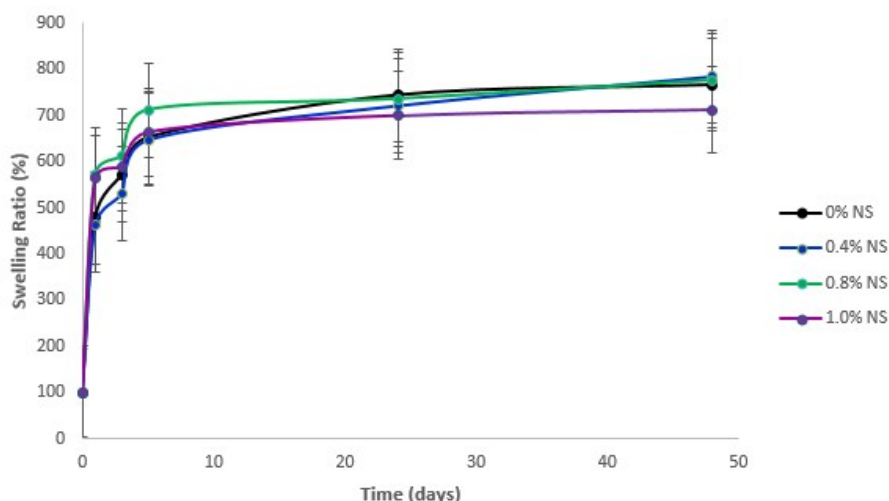


Fig S2. Swelling equilibrium profiles of the different nanocomposite hydrogels over time. The study was carried out in PBS (pH 7.4) n=4.

No significant difference in the swelling equilibrium was observed among the different groups, therefore, Laponite was unable to influence the hydrogel's ability to uptake PBS.

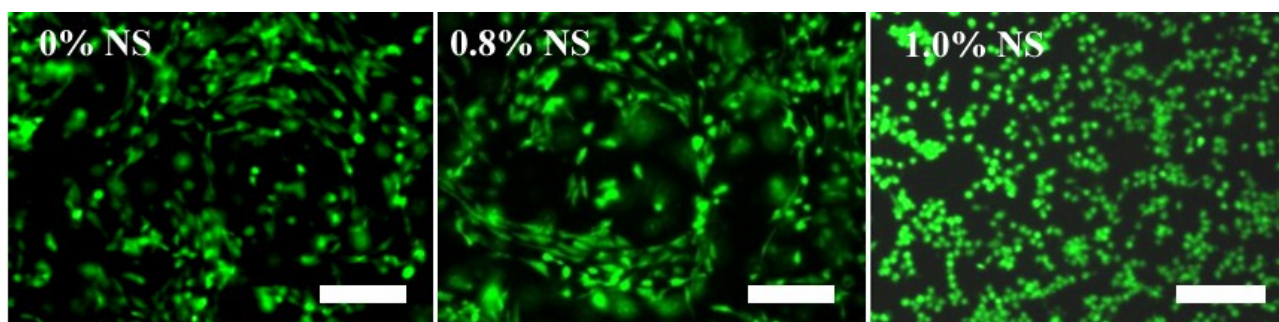


Fig S3. Calcein staining of HUVECs in 3D GelMA nanocomposite hydrogels having different concentration of Laponite. Scale bar: 100 μ m

HUVECs were able to spread in GelMA nanocomposite hydrogels having 0.8% w/v of Laponite. On the contrary, HUVECs showed a round morphology in the hydrogel containing 1.0% w/v Laponite mainly due to the increase stiffness observed for this hydrogel in the mechanical studies.