

Supplementary Information

Spatially Heterogeneous Epidermal Growth Factor Release from Microporous Annealed Particle (MAP) Hydrogel for Improved Wound Closure

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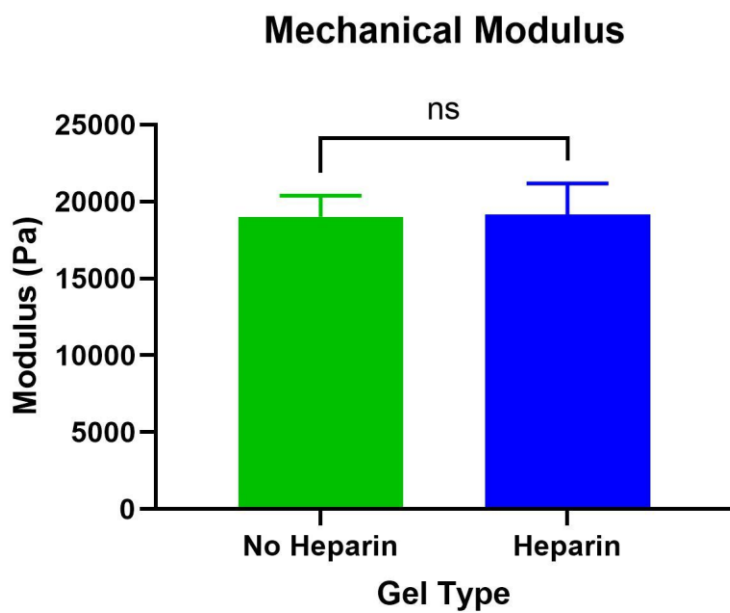


Figure 1. Mechanical Matching. Heparin and no heparin microgels were matched with a Young's modulus of approximately 19kPa as determined via Instron mechanical testing of chemically identical nanoporous scaffolds. N=3. Statistics: Student's t-test.

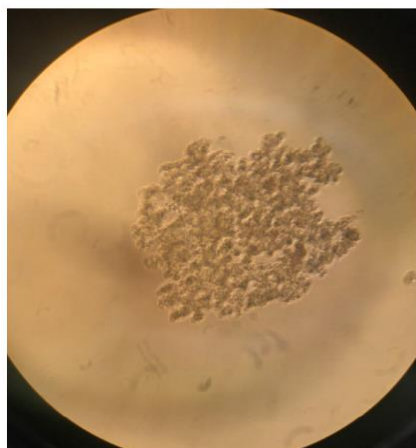
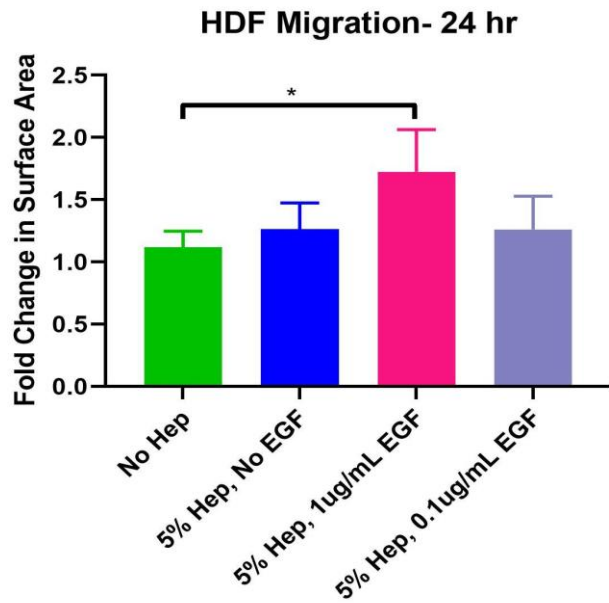
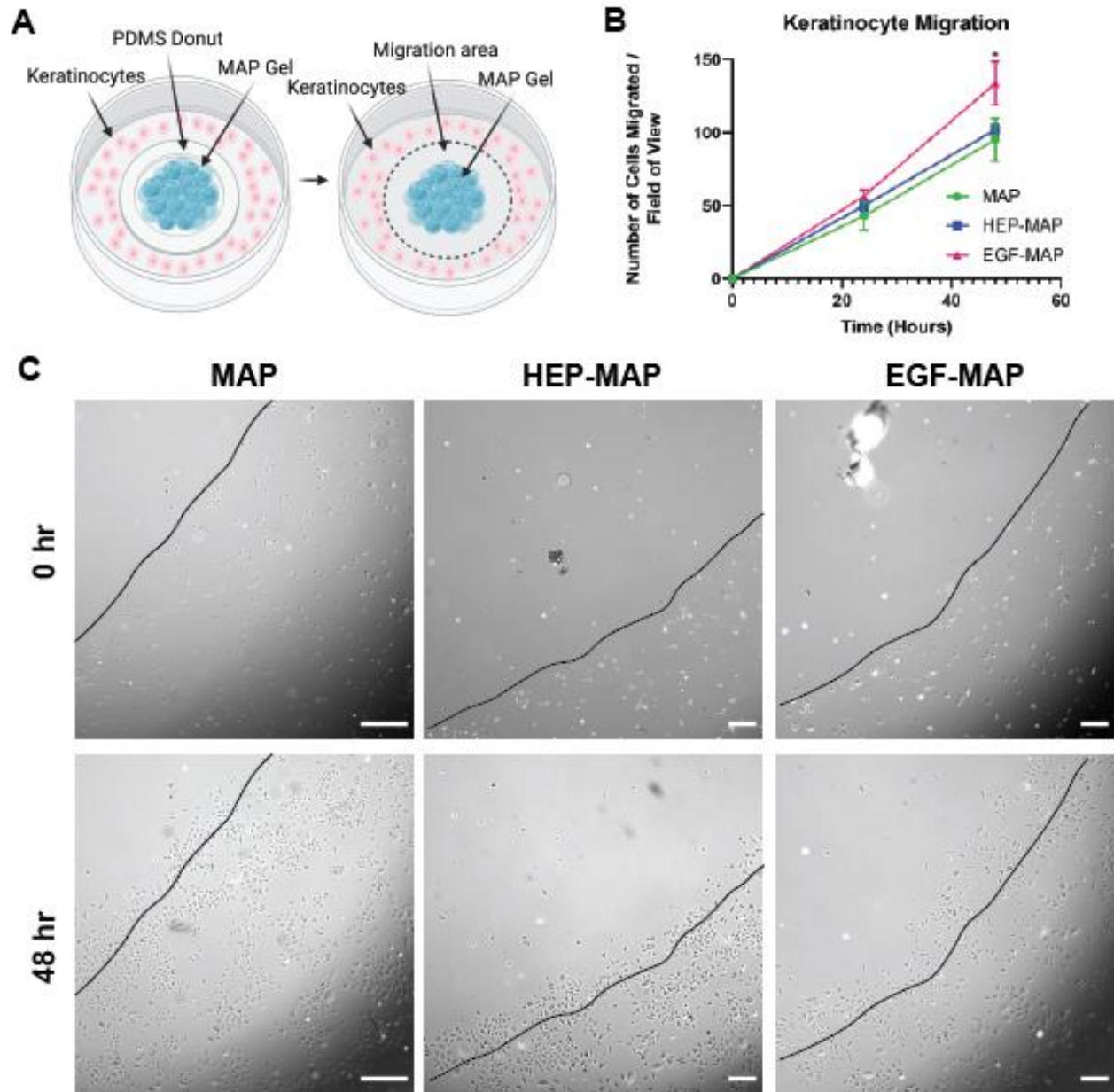


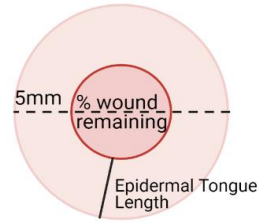
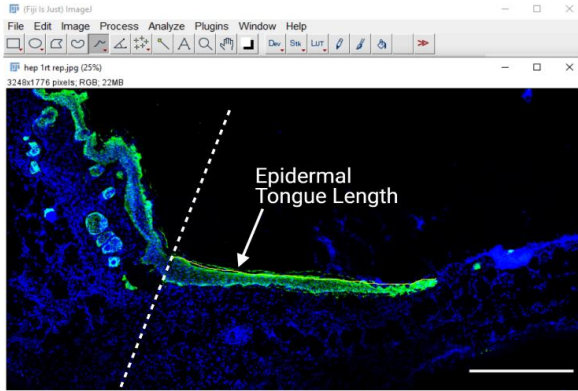
Figure 2. Keratinocytes were unable to form a spheroid after 7 days even with the addition of methylcellulose, therefore we were only able to perform the 3D migration assay for Human Dermal Fibroblasts.



Supplemental Figure 3. In a pilot HDF migration study (N=4) we observed that 5% hep, 1 μ g/mL significantly increased HDF migration whereas 5% heparin with 0.1 μ g/mL did not have a significant effect. Therefore, we chose to use 1 μ g/mL as our EGF concentration for the migration and wound healing studies. N=4. Statistics: ANOVA followed by multiple comparisons post-hoc test (Tukey HSD). *p<0.05.



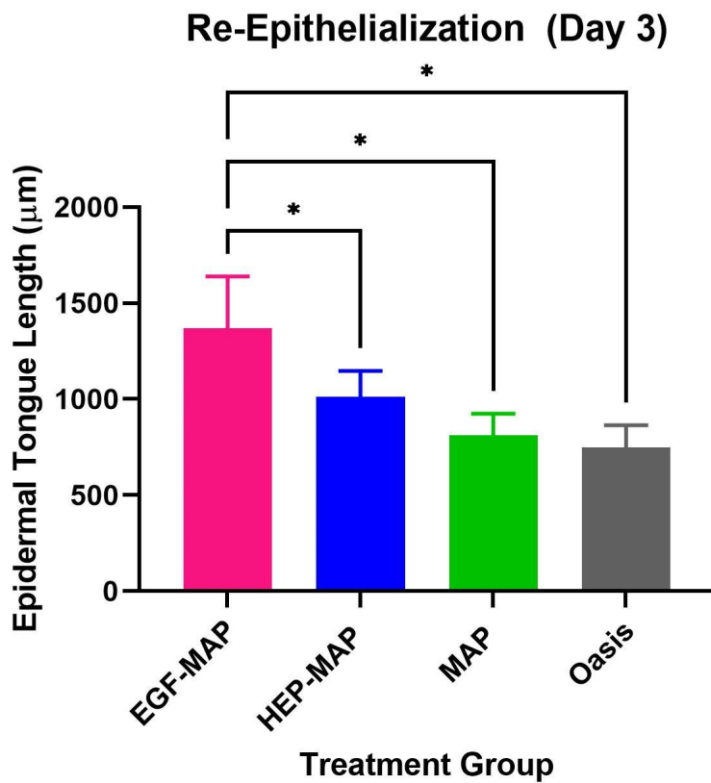
Supplemental Figure 4. A) The 2D keratinocyte migration assay consisted of first seeding keratinocytes around a PDMS donut with annealed MAP scaffolds in the center. After the keratinocytes had adhered, the PDMS donut was removed to allow the cells to migrate in the blank area towards the center. B) The EGF-MAP direct formulation promoted significantly more migrated cells per field of view compared to HEP-MAP ($p=0.0425$) and MAP alone ($p=0.0193$). C) Representative images of keratinocyte migration images at 0 hours and 48 hours. The line represents the keratinocyte boundary at 0 hours which was used to determine the number of migrated cells at each time point. Statistics: $N=3$, ANOVA followed by post-hoc tests (Tukey HSD) at each time point. $*p<0.05$.



$$\% \text{ wound remaining} = \frac{(2.5\text{mm} - \text{epidermal tongue length})^2 \cdot \text{PI}}{2.5^2 \cdot \text{PI}}$$

$$\% \text{ wound closed} = 1 - \% \text{ wound remaining}$$

Supplemental Figure 5. Epidermal tongue length was measured by tracing the length on ImageJ. Epidermal tongue length was used to calculate percentage wound closure.



Supplemental Figure 6. Epidermal tongue length measurements at Day 3 corresponding to the percentage wound closure in Figure 4. Statistics: ANOVA followed by multiple comparisons post-hoc test (Tukey HSD). N=6. *p<0.05.