ADVANCED+ BIOMEDICAL MATERIALS
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Developing a 3D skin model to analyse the cellular and epigenetic effects of IL-17 inhibitors in psoriasis

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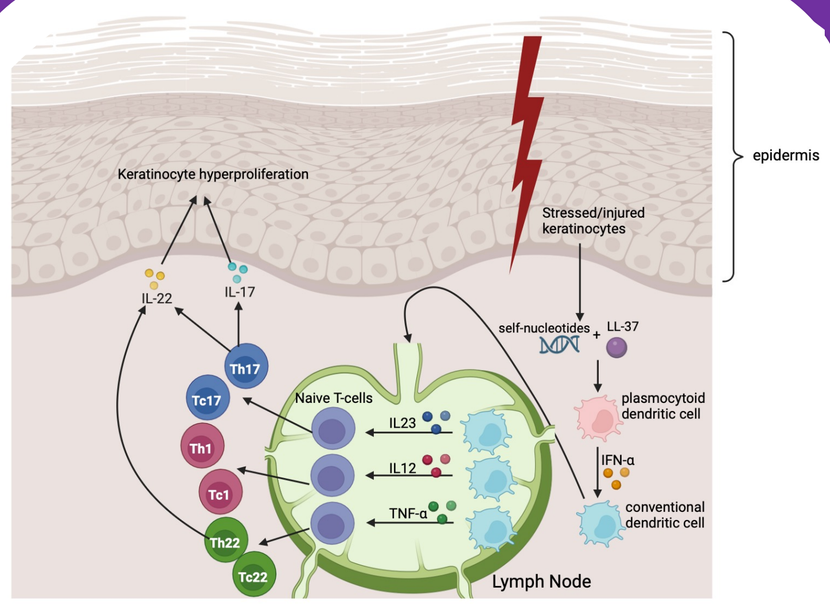
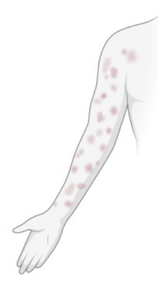


Figure 1. A simplified diagram of the main psoriasis pathogenesis axis, including the crucial IL-23/Th17 pathway. Created with BioRender.com. Adapted from (Orsmond *et al*., 2021) [3].

Background:



- Psoriasis is a chronic, immune-mediated, inflammatory skin disease [1].
- There is currently no cure [1].
- IL-17 inhibitors are a successful therapeutic treatment for Psoriasis [2].
  - Though, many patients fail to respond, and many patients relapse.

Aims:

- Develop a full-thickness 3D skin model with integrated immune cells.
- Use CRISPR gene-editing to to model psoriasis for biologic drug testing.

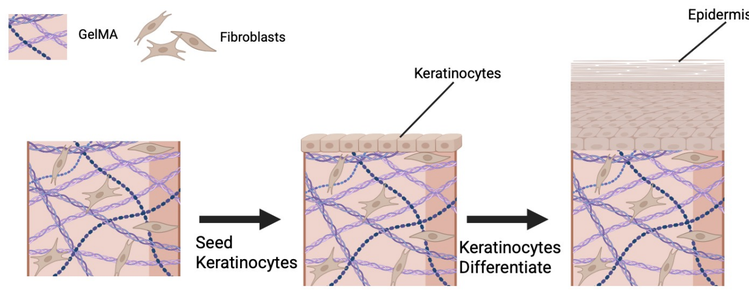


Figure 2. A simplistic diagram of or 3D skin model. Created with BioRender.com.

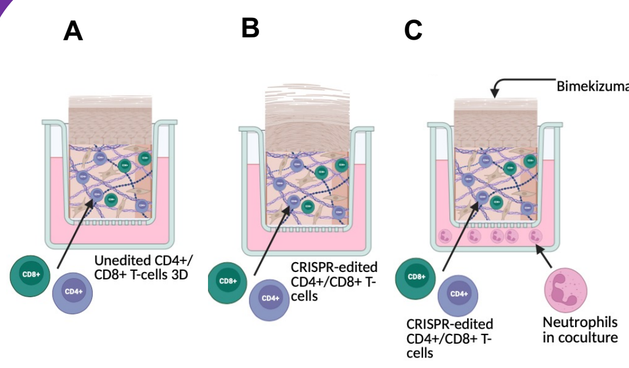


Figure 3. Our 3D skin model: (A) a 'healthy' skin model with unedited CD4+/CD8+ T-cells embedded in the; (B) a 'psoriatic' skin model with CRISPR-edited CD4+/CD8+ T-cells embedded in the; (C) the 'psoriatic' skin model with CRISPR-edited CD4+/CD8+ T-cells embedded in the matrix, treated with Bimekizumab. Created with BioRender.com.

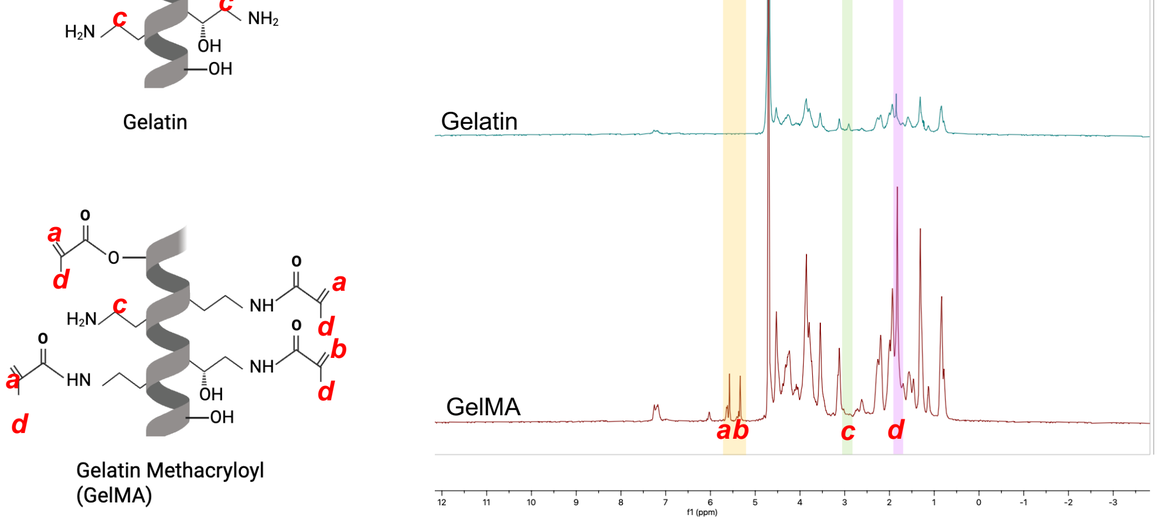


Figure 4. ^{1}H nuclear magnetic resonance (NMR) spectra of gelatin (porcine) and GelMA, with labelled proton groups (a to d) as referenced in the molecular structures of gelatin and GelMA.

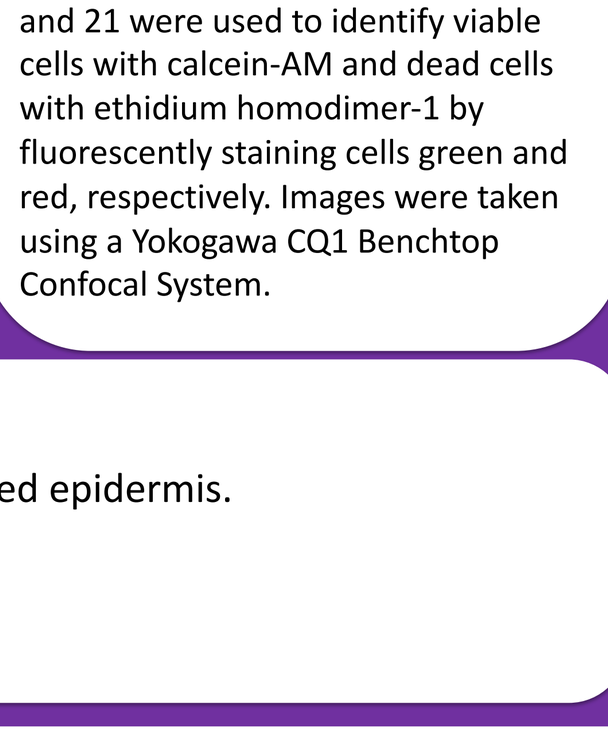


Figure 6. Cell viability of NHDFs encapsulated at ~2x10^{6} cells/mL in 10% (*w/v*) GelMA hydrogels. LIVE/DEAD assays at days 1, 7, 14 and 21 were used to identify viable cells with calcein-AM and dead cells with ethidium homodimer-1 by fluorescently staining cells green and red, respectively. Images were taken using a Yokogawa CQ1 Benchtop Confocal System.

SUCCESSFULLY ANALYZED SAMPLES

SAMPLE	GUIDE TARGET	NUCLEASE	INDEL %	MODEL FIT (R^2)	KO-SCORE
D3_ZFP36_kO-Premixed	GCTACAAGACTGAGCTATGT CAACCCTAGCGAAGACCTGG	SPCAS9	100	0.94	100
D3_ZFP36L_KO-Premixed	CTGCCACTTCATCCACAACG CGCCGTTCTCGCTAAACGAG	SPCAS9	99	0.9	99

Figure 7. EditCo analysis of knockout efficiency of *ZFP36 and ZFP36L2* in T Cells.

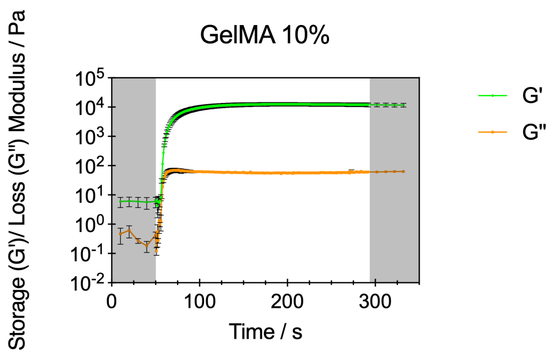


Figure 5. In-situ photorheology performed on 10% (w/v) GelMA, using a 400-500nm light source and 0.2% (w/v) LAP. Shaded areas represent periods where the light is off. Green and orange data points represent the storage (G') and loss (G'') moduli, respectively. Data presented as mean with standard error of the mean.

Future Work:

- Seeding keratinocytes on top of GelMA hydrogels and lifting to air/liquid interface to produce a fully-stratified epidermis.
- Histological analysis of skin model.
- Integration of immune cells into the dermal matrix.
- Treatment with Bimekizumab.

References:

1. World Health Organization. Global report on psoriasis. World Health Organization, Geneva. 2016.
2. Gordon KB, Foley P, Krueger JG, Pinter A, Reich K, Vender R, Vanvoorden V, Madden C, White K, Cioffi C, Blauvelt A. Bimekizumab efficacy and safety in moderate to severe plaque psoriasis (BE READY): a multicentre, double-blind, placebo-controlled, randomised withdrawal phase 3 trial. The Lancet. 2021 Feb 6;397(10273):475-86.
3. Orsmond A, Bereza-Malcolm L, Lynch T, March L, Xue M. Skin barrier dysregulation in psoriasis. International journal of molecular sciences. 2021 Oct 7;22(19):10841.