A Novel Pro-Angiogenic Fibrin-Alginate Technology for Repair and Regeneration of Multiple Tissues

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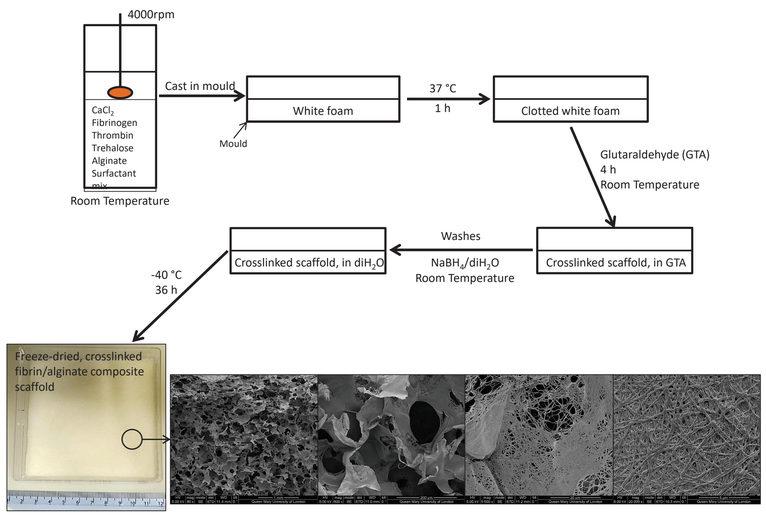
Introduction: We describe a novel patented technology developed in our laboratory based on a fibrin-alginate mesh that is pro-angiogenic and shows excellent cell attachment and infiltration properties, making it an ideal platform technology for repair and regeneration of multiple tissues.

The first product developed using this technology is a dermal replacement scaffold called Smart Matrix®. Extensive *in vitro* and *in vivo* analysis has shown that Smart Matrix® allows a rapid initial infiltration of cells and blood vessels. Advantageously, this fibrin-alginate technology can be combined with synthetic polymers, either inert (i.e. silicones) or bioactive (i.e. polycaprolactone, PCL) in various shapes (sheets, 3D structures), or osteogenic components for repair and regeneration of various tissues.

Aim: to introduce a novel fibrin-alginate technology for repair and regeneration of multiple tissues

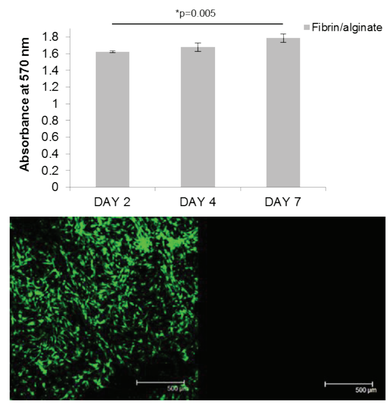
Method & Results



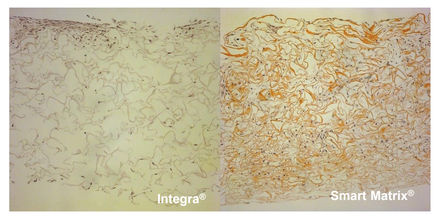
Graphical summary of the manufacturing process of fibrin/alginate scaffolds. Bottom, macroscopic and scanning electron microscopy photos of the scaffolds showing the micro- and nano-features (nano-pores and nano-fibres) of the matrix (García-Gareta *et al*. 2013; Sharma *et al*. 2016; Sharma *et al*. 2017).

Parameter	Smart Matrix®
Average porosity (% Vol)	83.22
Pore interconnectivity (%)	100
Average pore size (µm)	132.26
Average roughness Sa (nm)	114.776
Average G' (kPa)	8.26

Physical and rheological parameters of Smart Matrix® (Sharma *et al*. 2016; Levin *et al*. 2018).

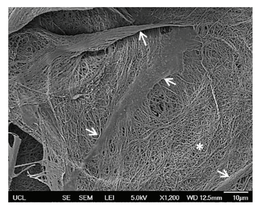


Cytotoxicity (live/dead assay, bottom) and proliferation (Alamar Blue metabolic assay, top) of primary human dermal fibroblasts on the fibrin alginate matrix showing significant growth over 7 days of culture under standard conditions. (García-Gareta *et al*. 2013; Sharma *et al*. 2016, 2017)

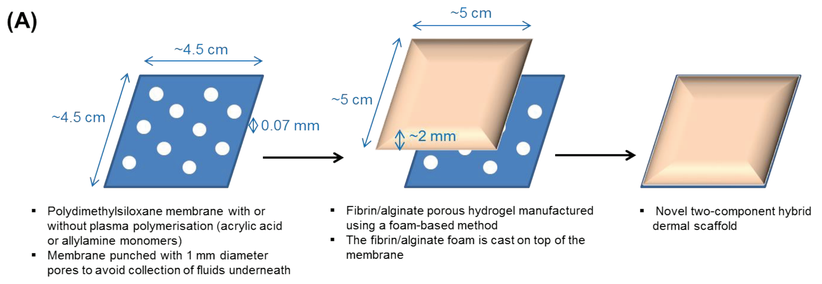


Haematoxylin & Eosin (H&E) stained cross-sections of Smart Matrix® seeded with primary human dermal fibroblasts at day 7 compared to the collagen-based and clinically well-established Integra®. Cells are stained purple and the scaffold structure appears pink/red. All photographs are at 10X magnification.

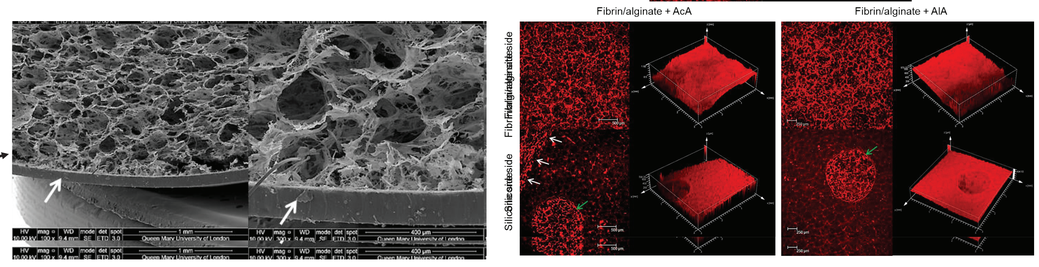
Results show an even distribution of cells throughout the Smart Matrix® scaffold while cells in Integra® were mostly observed on the surface. (Sharma *et al*. 2016)



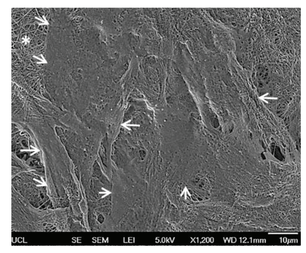
FESEM image of fibrin/alginate scaffold seeded with primary human dermal fibroblasts at day 7 of culture. White arrows point at cells which are seen embedded in the matrix. (Sharma *et al*. 2017).



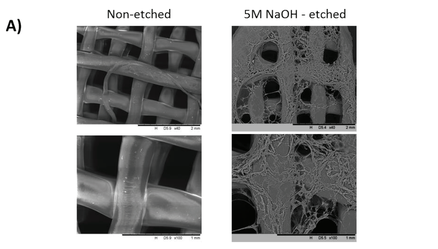
A novel two-component dermal scaffold for the treatment of pressure sores was designed using a perforated polydimethylsiloxane backing membrane to make the fibrin-alginate dermal scaffold more robust: A) scheme of the novel two-component hybrid dermal scaffold, B) macroscopic photos of the fibrin/alginate + silicone composite (Sharma *et al*. 2015, 2017).



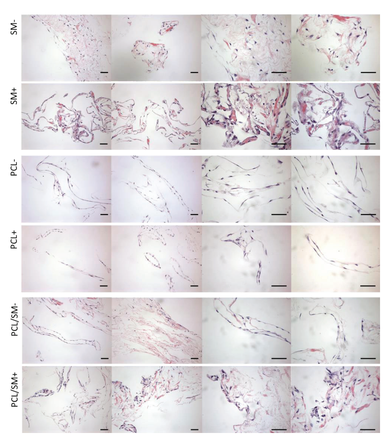
Representative SEM (left) and confocal microscopy (right) images of fibrin/alginate + silicone composites. White arrows in SEM point at the silicone membrane while in confocal images point at the edge between the silicone and the fibrin/alginate matrix. Green arrows point at the pores present in the silicone membrane. Results showed that the characteristic micro- and nano-structure of the fibrin/alginate matrix was preserved (Sharma *et al*. 2017).



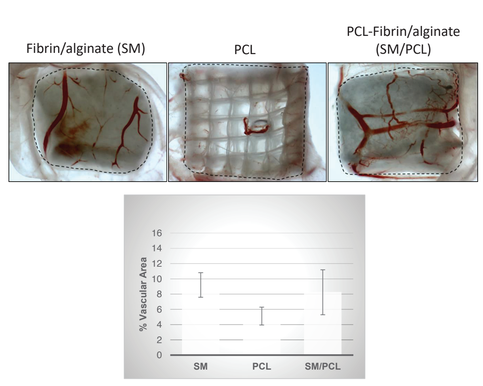
SEM image of fibrin/alginate + silicone composite seeded with primary human dermal fibroblasts at day 7 of culture. *shows the nano-fibres and nano-pores present in the fibrin/alginate matrix. White arrows point at cells which are seen embedded in the matrix. (Sharma *et al*. 2017)



SEM photos of PCL-fibrin/alginate composites fabricated using melt electrospun PCL structures. A) Comparison between native PCL and etched PCL (5M NaOH for 5 h) surfaces. B) Composites used for tissue culture. (Manuscript in preparation)

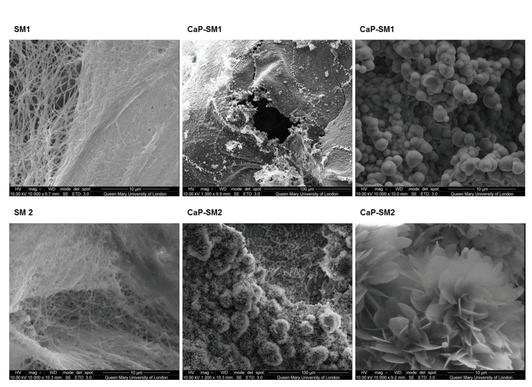


H&E staining of cellularised fibrin/alginate (SM), PCL and PCL-fibrin/alginate (PCL/SM) composites cultured in either standard (-) or osteogenic medium (+) for 28 days. The scaffold is bright pink while the cells appear purple/black. Scale bar = 50 µm. (Manuscript in preparation)



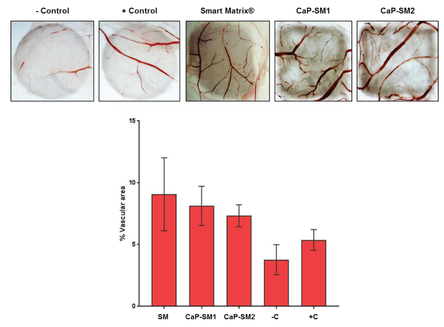
Ex ovo CAM assay results (5 mm x 5mm scaffold size) showing higher % vascular area on the composite and SM compared to PCL alone. (Manuscript in preparation).

- For the *Ex ovo* CAM assay details, visit poster a92254.



SEM representative images of fibrin/alginate-calcium phosphate (CaP-SM) scaffolds showing different CaP morphology depending on preparation method. (Material patented; Manuscript in preparation).

- For details on this material, please visit poster a92058.



Ex ovo CAM assay results (5 mm x 5mm scaffold size) showing similar % vascular area on the composites and SM, which have higher % vascular area that the positive control (filter disc dipped in VGEF solution), showing the pro-angiogenic potential of the fibrin/alginate technology. (Manuscript in preparation).

- For the *Ex ovo* CAM assay details, visit poster a92254.
- For details on this material, please visit poster a92058.

Conclusions

- We present a fibrin-alginate platform technology which is pro-angiogenic and promotes a rapid initial cellular infiltration.
- The first product out of this platform technology is a dermal replacement scaffold (Smart Matrix®) which is under clinical development at the spin-out Smart Matrix Ltd.
- Future development of this platform technology will see its combination with 3D printing for development of custom-made implants.

References:

1. García-Gareta et al. Bioresearch Open Access 2(6), 412, 2013.
2. Sharma et al. Biomedical Materials 11, 055001, 2016.
3. Sharma et al. Biochimie Open 1, 40, 2015.
4. Sharma et al. Macromolecular Bioscience 1700185, 2017.
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Acknowledgements: work was supported by the Restoration of Appearance and Function Trust (UK, charity No. 299811) and Smart Matrix Limited.

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