A Model to Evaluate the Biomechanical Strain Modalities Delivered to Tissue by a Novel tNPWT+DL* Dressing

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Introduction

- Whilst traditional Negative Pressure Wound Therapy (tNPWT) systems have demonstrated efficacy in managing hard-to-heal wounds[1], they are limited by their localized pressure delivery. In contrast, advanced single-use NPWT‡ systems[2] have shown accelerated healing outcomes, attributed to their ability to distribute negative pressure across a wider therapeutic zone. To bridge this gap, a novel cuttable and repositionable tNPWT dressing, enhanced with a distribution layer (tNPWT+DL*) has been developed (**Figure 1**).

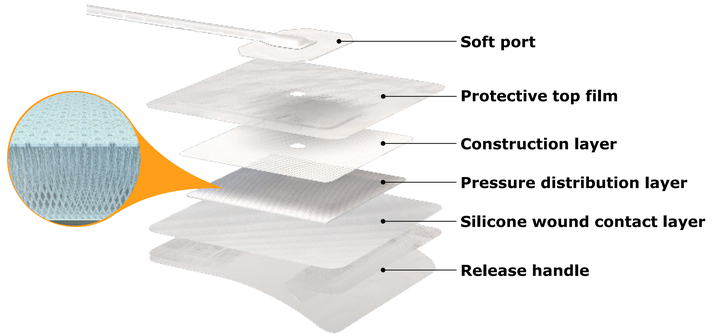

Figure 1. Novel tNPWT+DL* dressing and constituent layers.

Study Aim

- To investigate the biomechanical effects of NPWT *in vitro*. Peri-wound dermal strain range of interest (3.5-6.5%) was assessed for its influence on injured primary human dermal fibroblasts (HDFs) using a bespoke cell culture model.

Methodology

- **Tissue strain measurements:** *In vitro* studies were conducted with porcine tissue substrates (pork belly tissue, n=3).
- Wounds (~3 cm diameter, ~1.5 cm deep) were created prior to application of tNPWT† and tNPWT+DL*.
- Computational Finite Element Analysis (FEA) was used to determine tissue strains calculated from real-time displacement data (measured with metallic markers inserted into the porcine tissue in a grid pattern and imaging with micro-CT concurrently with the application of the NPWT) at depths of 1, 2, and 3 cm from the surface for both tNPWT† and tNPWT+DL*.
- **Effect of strain on injured primary human dermal fibroblasts (HDF):** Strain range of interest (3.5–6.5%) were identified within ~3.5 cm from the wound centre, consistent with the peri-wound region.[3]

Methodology cont.

- A bespoke cell stretching device (CSD) capable of delivering the target strains (3.5–4.5% and 5.5–6.5%) was developed, fabricated & validated. Crucially, this device also delivered cyclic§ strain to maintain strain experienced by the cells (i.e. as the cells migrate & proliferate) over the full duration of the experiment.
- The CSD employed specialized collagen I-coated elastic membranes (StageFlexer™, Flexcell™ International Corp.) to support cell adhesion and facilitate strain application. Strain was delivered biaxially via a vacuum-controlled system, allowing precise modulation of mechanical forces across the cell culture surface.
- Primary HDFs were isolated from five donors (ages 27–55) with full informed consent (NHS REC 17/SC/0220): one male (leg) and four females (abdomen ×3, breast ×1). All cells were used at <14 passages.
- HDFs were cultured on StageFlexer membranes housed within the CSD and maintained at 37 °C, 5% CO_2. At ~90% confluence, a vertical scratch was made, debris removed with PBS and fresh medium added. Mechanical strain cycles were then applied for 24 hours under identical incubation conditions. Post treatment, fixation and staining were performed using 1% crystal violet. Scratch images were captured via Optika™ IM-3LD4D microscope and quantified using ImageJ (v1.54g).

§The application of cyclic strain is intended to more accurately simulate the relative constant strain experienced by cells in the peri-wound tissue during application of NPWT. Importantly, this cyclic strain **does not** represent fluctuations in negative pressure itself.

Results

- The tNPWT+DL* dressing delivers 3.5-6.5% strain to a significantly greater extent in the peri-wound tissue region than tNPWT†, with a 6.3-fold (528%) increase in coverage at 2 cm sub-dermal tissue depth (**Figure 2**).

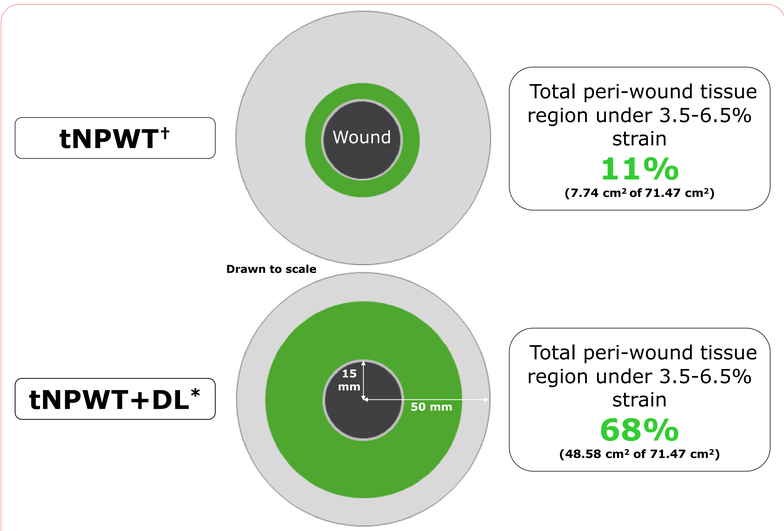

Figure 2. Map of peri-wound strain range of interest. Visual representation of tissue strain distribution in the peri-wound region, 2 cm sub-dermal tissue depth, highlighting: ■ Tissue under 3.5-6.5% strain, ■ Tissue strain outside range of interest.

Results cont.

- The effect of application of the peri-wound region dermal strain of interest (3.5-6.5%) on injured primary human dermal fibroblasts was positive (**Figure 3**).
- For the 3.5–4.5% cyclic strain group, mean scratch closure increased from 71.94% (no strain control) to 79.23%, representing a 7.29% improvement (approximately a 0.1-fold increase), which was statistically significant ($p < 0.0001$).
- Similarly, the 5.5–6.5% cyclic strain group showed an increase in mean scratch closure from 77.10% (no strain control) to 85.07%, a 7.97% improvement (approximately a 0.09-fold increase), also statistically significant ($p < 0.01$).

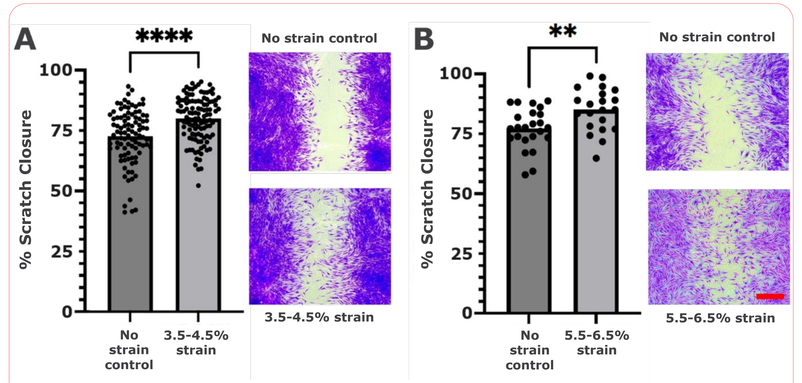

Figure 3. Scratch closure (%) and representative crystal violet-stained images of human dermal fibroblasts under cyclic mechanical strain. Quantification of scratch closure and representative images of human dermal fibroblasts subjected to biaxial cyclic strain between A) between 1.7% strain and 3.5-4.5% therapeutic strain in 30 min intervals for 24 hours compared to no strain controls (n=96-112 across Donors 4 and 5); and B) between 0% strain and 5.5-6.5% strain in 30 min intervals for 24 hours compared to no strain controls (n=21-24 across Donors 1, 2 and 3). Each image was treated as an independent data point, reflecting the heterogeneous microenvironmental strain distribution within the CSD. Statistical analysis was performed using Welch's t-test, **, $p<0.01$, ****, $p<0.0001$; Bar = 500 µm.

Conclusions

- **Strain distribution**: A greater proportion of peri-wound tissue is exposed to beneficial strain range (3.5–6.5%) with tNPWT+DL* (68%) compared to tNPWT† (11%). This represents a 6.3-fold (528%) increase for tNPWT+DL*, relative to tNPWT†.
- **Biological impact**: *In vitro* modelling of strains within this range demonstrated a positive effect on injured primary human dermal fibroblasts, with significantly enhanced cell migration observed in both strain groups compared to no-strain controls.
- These findings indicate that, by delivering beneficial mechanotransductive strains more broadly to the peri-wound region tissue than tNPWT†, the novel tNPWT+DL* dressing may further improve cellular responses and accelerate wound healing.

References: 1. Hurd T, *et al*. Adv Wound Care (New Rochelle). 2017 Jan 1;6(1):33-37. **2.** Kirsner R, *et al*. Wound Repair Regen. 2019 Sep;27(5):519-529. **3.** Dowsett C, *et al*. Wounds International. 2015 6(1):19-23.

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*RENASYS◊ WOUND+ Negative Pressure Wound Therapy dressing, Smith & Nephew. †RENASYS◊ Negative Pressure Wound Therapy with drape and foam filler, Smith & Nephew. ‡PICO◊ single-use Negative Pressure Wound Therapy, Smith & Nephew.

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