The Biological Impact of a Novel tNPWT+DL† Dressing Using *ex vivo* Human Skin

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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, single-use NPWT (sNPWT) 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 repositionable tNPWT dressing, enhanced with a distribution layer (tNPWT+DL†) has been developed (**Figure 1**).

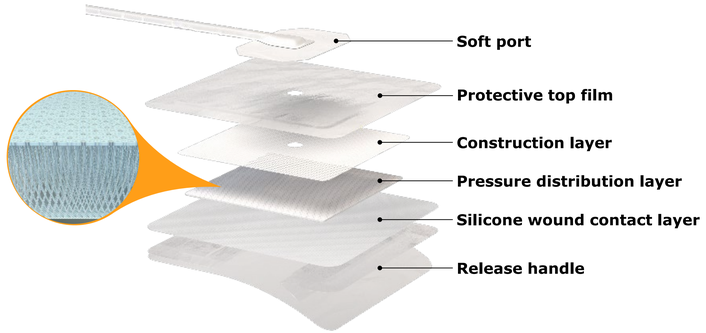

Figure 1. Novel tNPWT+DL† dressing and constituent layers.

Study Aim

- The work detailed here was carried out to investigate and compare the biological effect of tNPWT‡ and tNPWT+DL† on cells surrounding a wound, using an *ex vivo* human skin model.

Methodology

- A novel *ex vivo* culture system arranged human skin, donated from elective surgery with fully informed consent (NHS REC 19/NE/0150 and 17/SC/0220), onto a SILSKIN support, with access to culture media provided via channels. A 3 cm diameter, 1 cm deep wound was created in the *ex vivo* skin sample to trigger a host injury response (**Figure 2**). Treatments delivered therapy for 48 h.

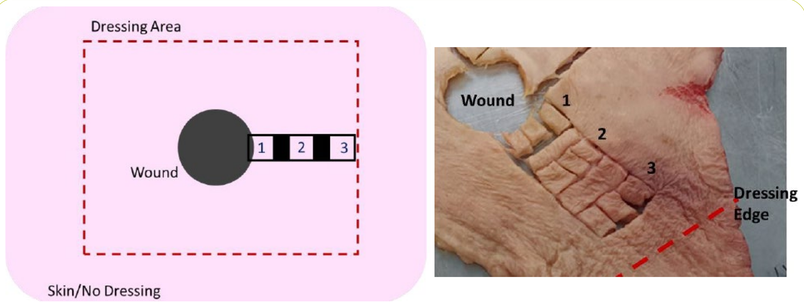

Figure 2. Collection of human skin. Skin was removed from the culture set up following 48hrs of culture at 37 °C and 5% CO_2. Samples were removed at defined locations; the wound edge (WE; 1; 1.5 cm from wound centre), the peri-wound (PW; 2; 2.75 cm from wound centre) and the extended zone (EZ; 3; 4 cm from wound centre).

Methodology cont.

- Observational measurements were taken from skin harvested at the wound edge (WE), peri-wound (PW) and extended zone (EX) locations for histological staining (**Figure 2**).

The following analyses were undertaken on tissue samples:

- Haematoxylin and eosin (H&E) staining was used to assess the degree of epidermal-dermal junction (EDJ) disruption. Images were assessed no a Nikon E400 microscope with SPOT camera (Image Solutions UK Ltd. , Preston).
- Immunohistochemical staining (rabbit anti-loricrin antibodies) was used to determine the percentage dysregulation of loricrin, an epidermal structural protein. Stain intensity was quantified using Image J (V. 1.8.0; NIH, US).
- Epidermal cell viability was measured using TUNEL staining. TUNEL positive (green) cells were manually counted and expressed as a percentage of total cells (DAPI, blue). Composite images for analysis were created using Image J (V. 1.8.0; NIH, US).

Results

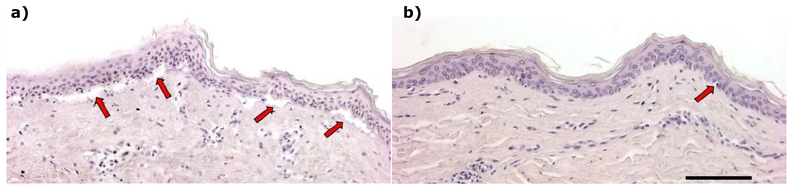

Figure 3. H&E stained wound edge sections treated with either a) tNWPT‡ or b) tNPWT+DL†. Higher-power images illustrate substantial epidermal-dermal junction damage following tNPWT‡, with little damage following tNPWT+DL† treatment. Epidermal-dermal separation (red arrows). Bar = 150 µm.

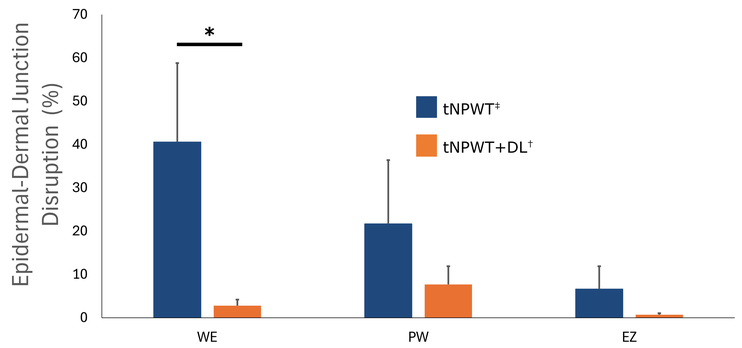

Figure 4. Mean (n=3 donor skin samples) percent EDJ disruption with respect to locations relative to the wound, WE=wound edge, PW=peri-wound, EZ=extended zone. NPWT delivered for 48 h. Graph show mean plus SEM, *p<0.01.

- The tNPWT+DL† dressing delivered significantly reduced disruption to the epidermal-dermal junction at the wound edge (WE) compared to tNPWT‡ (p<0.01) (**Figures 3** & **4**).

Results cont.

- The tNPWT+DL† dressing retained significantly greater epidermal cell viability at the wound edge (WE) compared to tNPWT‡ (p<0.02) (**Figure 5**).

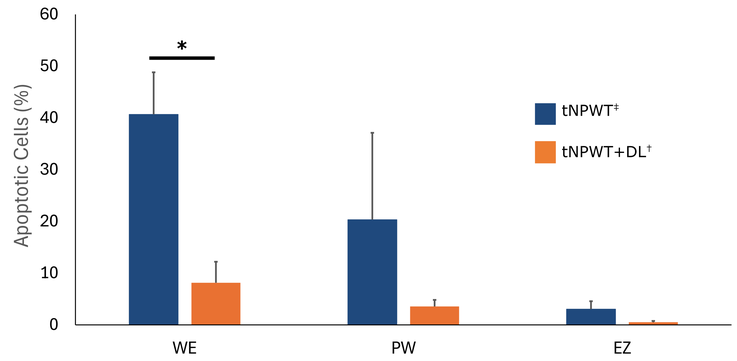

Figure 5. Mean (n=3 donor skin samples) percent apoptotic cells with respect to locations relative to the wound, WE=wound edge, PW=peri-wound, EZ=extended zone, NPWT delivered for 48 h, Graph show mean plus SEM, *p<0.02.

- The tNPWT+DL† dressing facilitated significantly greater retention of the epidermal structural protein loricrin at the wound edge (WE) and the peri-wound (PW), relative to tNPWT‡ (*p<0.05, **p<0.01) (**Figure 6**).

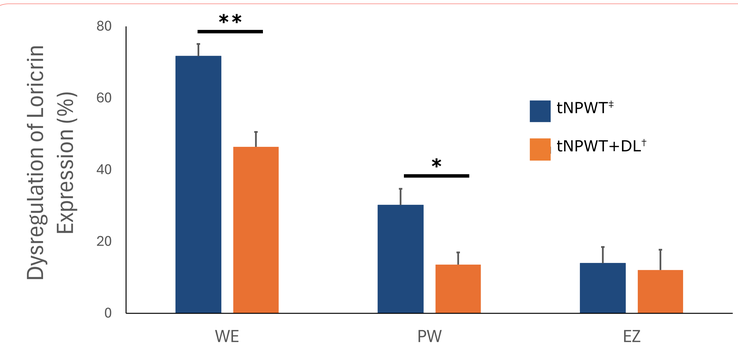

Figure 6. Mean (n=3 donor skin samples) percent dysregulation of loricrin expression at locations relative to the wound, WE=wound edge, PW=peri-wound, EZ=extended zone. NPWT delivered for 48 h. Graph show mean plus SEM, *p<0.05, **p<0.01.

Conclusions

- **Biological impact:** Treatment with the tNPWT+DL† dressing resulted in significantly reduced epidermal-dermal junction disruption and dysregulation of the epidermal structural protein loricrin. Additionally, a significantly greater epidermal cell viability at the wound edge was observed.
- These findings suggest that NPWT systems, such as the tNPWT+DL† dressing, which deliver consistent NWPT across the dressing pad may promote improved cellular responses and potentially 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.

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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.

Wound Management, Smith & Nephew Medical Ltd, 101 Hessle Road, Hull, HU3 2BN, UK.