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Influence of advanced wound matrices on observed vacuum pressure during simulated negative pressure wound therapy

Robert W.F. Veale, Tarek Kollmetz, Navid Taghavi, Claudia G. Duston-Fursman, Matthew T. Beeson, Dorrin Asefi, Henry D. Chittock, Ananth S. Vikranth, Shane G. Dowling, Sandi G. Dempsey, Hamish J. Rose, Isaac T.T. Mason, Barnaby C.H. May *

Aroa Biosurgery Limited, Airport Oaks, Auckland, 2022, New Zealand

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ABSTRACT

Biomaterials and negative pressure wound therapy (NPWT) are treatment modalities regularly used together to accelerate soft-tissue regeneration. This study evaluated the impact of the design and composition of commercially available collagen-based matrices on the observed vacuum pressure delivered under NPWT using a custom test apparatus. Specifically, testing compared the effect of the commercial products; ovine forestomach matrix (OFM), collagen/oxidized regenerated cellulose (collagen/ORC) and a collagen-based dressing (CWD) on the observed vacuum pressure. OFM resulted in an ~50% reduction in the observed target vacuum pressure at 75 mmHg and 125 mmHg, however, this effect was mitigated to a ~0% reduction when fenestrations were introduced into the matrix. Both collagen/ORC and CWD reduced the observed vacuum pressure at 125 mmHg (~15% and ~50%, respectively), and this was more dramatic when a lower vacuum pressure of 75 mmHg was delivered (~20% and ~75%, respectively). The reduced performance of the reconstituted collagen products is thought to result from the gelling properties of these products that may cause occlusion of the delivered vacuum to the wound bed. These findings highlight the importance of *in vitro* testing to establish the impact of adjunctive therapies on NPWT, where effective delivery of vacuum pressure is paramount to the efficacy of this therapy.

1. Introduction

Complex wounds present a significant economic burden on healthcare systems and are associated with significant levels of morbidity, and reduced quality of life (Lo et al., 2020). It is projected that the number of patients with chronic non-healing wounds will continue to increase due to an ageing population and the prevalence of related conditions such as obesity and diabetes (Gottrup et al., 2013; Hjort and Gottrup, 2010). As such, strategies to accelerate healing are critically important to address this unmet need. Negative pressure wound therapy (NPWT) has become widely adopted in modern wound care and has been shown to improve healing outcomes across a range of different wound types, including diabetic foot ulcers (Nain et al., 2011; Wynn and Freeman, 2019; Ilonzo et al., 2018; Blume et al., 2008), venous leg ulcers (Vuerstaek et al., 2006), and complex surgical wounds (Dowsett et al., 2013). NPWT is an adjunctive system that creates a sub-atmospheric ('vacuum') pressure in the wound bed to accelerate healing via a biomechanical effect (Ilizarov, 1989). Studies have shown that NPWT at 125 mmHg leads to a four-fold increase in blood perfusion at the site (Morykwas et al., 1997), significantly increases granulation tissue formation, decreases tissue bacterial counts and stimulates angiogenesis (Kilarski et al., 2009; Jacobs et al., 2009). NPWT exerts its effect by inducing a physical response ('macrostrain') and a biological response ('microstrain') (McNulty et al., 2010). Macrostrain draws wound edges together and removes exudate and infectious material, thus reducing edema and promoting perfusion (Vellingiri et al., 2020). Microstrain creates tissue microdeformation, causing cells to stretch. Cell stretch leads to the migration of cells and proliferation that result in granulation tissue formation (Stafford et al., 2002). Various studies have shown that effective delivery of the vacuum pressure to the wound bed is critical to the clinical performance of NPWT. For example, Borgquist et al. (2011) have shown that the optimal pressure for changes in wound dimension is achieved at >75 mmHg, while the optimal pressure for wound drainage is 125 mmHg.

Wound healing practioners have embraced advanced dermal matrix

Abbreviations: NPWT, Negative Pressure Wound Therapy; OFM, Ovine Forestomach Matrix; CWD, Collagen Wound Dressing.

* Corresponding author.
E-mail address: Barnaby.May@aroabio.com (B.C.H. May).

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technologies to accelerate wound closure, and reconstituted collagen and decellularized extracellular matrix (dECM) products are now commonplace in modern wound care (Urciuolo et al., 2019; Mendibil et al., 2020). Reconstituted collagen products, first developed in the 1980s, are produced from solubilized animal collagens that are reformed to a matrix in order to mimic the collagen structure found in tissues (Meyer, 2019). In contrast, dECM-based dermal matrices are prepared from intact animal or human tissues using processes to remove the donor cells, leaving an intact ECM with the same structure and composition as tissue ECM (Badylak et al., 2009). Both reconstituted collagen and dECM matrices are designed to aid wound healing by scaffolding the patient's own cells during the repair process.

Ovine forestomach matrix (OFM) (Endoform Natural™, Aroa Biosurgery, Auckland, New Zealand) is a dECM bioscaffold with a composition and structure that closely mimics human soft tissue ECM (Lun et al., 2010). OFM contains a large number of matrisome proteins (Dempsey et al., 2019), stimulates cellular differentiation, migration, and angiogenesis (Lun et al., 2010; Irvine et al., 2011). Once placed in contact with the tissue deficit, patient cells infiltrate the scaffold, and over time OFM is fully bio-absorbed into the regenerating soft tissues and remodelled as the tissue matures (Irvine et al., 2011). OFM has found clinical applications across a wide range of acute and chronic wounds (Liden and May 2013; Ferreras et al., 2017; Bohn and Gass, 2014; Lullove, 2017; Raizman et al., 2020). A recent real world evidence study of over 2200 diabetic foot ulcers demonstrated that wounds closed up to ~5.5 weeks faster when treated with OFM versus a reconstituted collagen matrix (Bosque et al., 2022).

The combined use of NPWT and collagen-based matrix technologies have been widely adopted in clinical practice and clinical outcomes using the combination have been described for a variety of wound types (Jeschke et al., 2004; Lehrman, 2020; Bohn and Chaffin, 2020; Dillingham and Jorizzo, 2019; Fleming et al., 2014; Loh et al., 2020; Gabriel and Gollin, 2006; Sreelesh and Laxminarayan Bhandari, 2017). Despite this, studies investigating the effects of these collagen-based products on the observed vacuum pressure delivered by NPWT systems are limited. Most *in vitro* testing of pressure transduction and fluid removal has focused on characterizing the impact of foam interface dressings on observed vacuum pressure. For example, McNulty et al. (2010) showed the use of gauze led to a significant pressure drop across multiple set vacuum pressures when compared to reticulated open-cell foam. Delgado and Sammons (2016) mapped vacuum pressures and wound fluid extraction efficiency of three different NPWT dressing systems and found that pressure distribution varied between the systems. In this study, we investigated changes in the observed vacuum pressure during simultaneous use of NPWT with reconstituted collagen and dECM dermal matrices. This study demonstrates that the composition and design of the dermal matrices can significantly impact the vacuum pressure delivered by NPWT systems.

2. Materials and methods

2.1. General

Test samples of OFM (Endoform Natural™, Aroa Biosurgery Limited, New Zealand), OFM-High Flow (OFMHF) (Endoform Natural™ – High Flow, Aroa Biosurgery Limited), collagen/oxidized regenerated cellulose (collagen/ORC) (Promogran™, 3M/KCI, Saint Paul, MN, USA), and collagen wound dressing (CWD) (Puracol® Plus, Medline, Northfield, Il, USA) were obtained from commercial suppliers and terminally sterile at the time of testing. The previously reported thicknesses of collagen/ORC, CWD, and OFM (including OFMHF) are 3 mm (Karr et al., 2011), 2 mm (Product Information Sheet, 2011), and 0.25 mm (Floden et al., 2011), respectively. OFMHF devices are prepared from sheets of OFM using a proprietary cutting process to introduce the fenestration pattern presented in Fig. 1.

Simulated wound fluid (SWF) was prepared according to the method of Rusanu et al. (2017), and comprised collagenase (35 ng/mL, *Clostridium histolyticum*, Sigma-Aldrich, St. Louis, MO, USA), bovine serum albumin (2%, w/v) (Sigma-Aldrich, St. Louis, MO, USA), NaCl (0.4 M), and $CaCl_2$ (0.02 M). Statistical analysis, including descriptive statistics (e.g., mean, standard error of the mean (SEM)) was conducted using GraphPad Prism (Prism 9 for Windows 64-bit, Version 9.3.0, 463). Significance between test articles was determined using ordinary one-way ANOVAs (Dunnett's multiple comparisons test), where a p-value less than 0.05 was considered statistically significant.

2.2. Negative Pressure Wound Therapy (NPWT) test apparatus

The NPWT test rig was custom fabricated by Aroa Biosurgery Limited. The base of the test rig (20 cm × 20 cm) was machined from acetyl plastic via a computer numerical control (CNC) and supported on high acetyl plastic legs (5 cm). Within the base of the test rig a recessed 'wound bed' was milled measuring 10 cm × 10 cm, and 5 cm deep. At the base of the wound bed were mounted two pressure sensors ('transducers') (MPS-3117-006GC, Metrodyne Microsystem Corporation, Hsinchu, Taiwan), one central (P1) and one peripheral (P2). The P1 and P2 sensors measured in real-time the observed vacuum pressures (Fig. 2).

Vacuum pressure was delivered to the test rig by a vacuum pump comprised of two parts; a variable speed motor (JRF/K–370CH, Guangdong Kingly Gear Co. Ltd., Huizhou, China), and a proprietary diaphragm vacuum pump head (Fig. 3). Vacuum is delivered to the test rig to the center of the 'wound bed' via the NPWT drape.

The pump was connected to the test rig via a polyvinyl chloride (PVC) tube (ID∅ 2 mm; OD∅ 4 mm) (Qosina, Ronkonkoma, NY, USA) to a polyurethane drape creating a closed system. The re-useable polyurethane drape (thickness of 130 μm), measured 25 cm × 25 cm and was

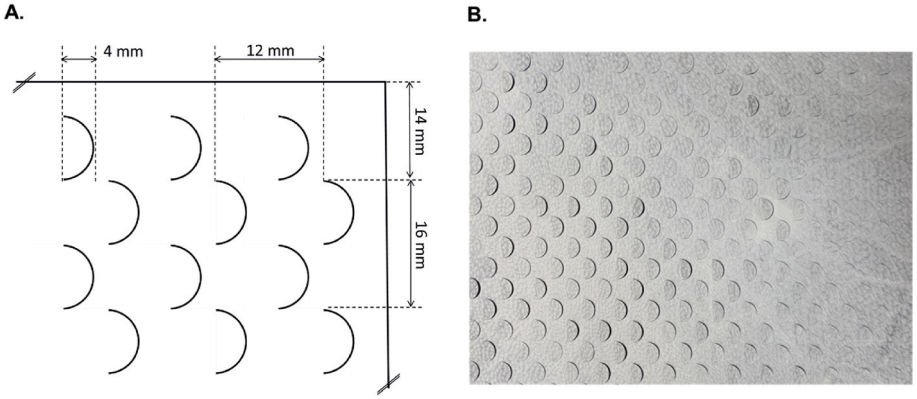

Fig. 1. Diagram (**A.**) and representative image (**B.**) of the ovine forestomach matrix-High Flow (OFMHF) test samples.

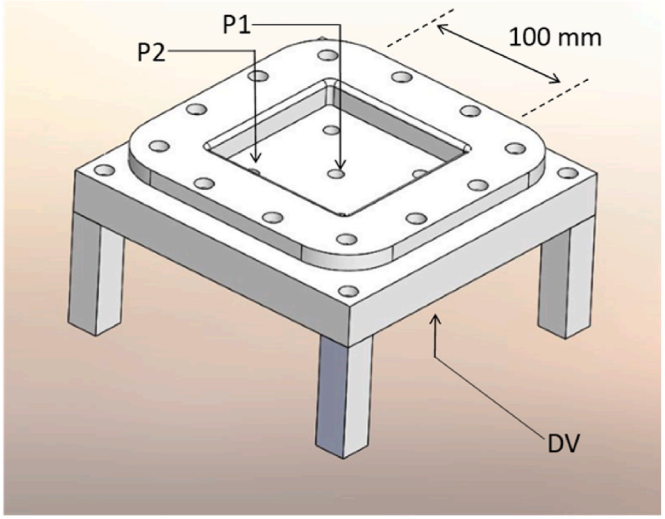

Fig. 2. Simplified isometric representation of the negative pressure wound therapy (NPWT) testing rig. DV = delivery port, positioned at the base of the test rig, for the delivery of SWF; P1 = central pressure sensor; P2 = peripheral pressure sensor – both sensors are placed in the base of the test rig.

fitted with a PVC port (IDØ, 9 mm) by radio-frequency welding in the center of the drape (de France Productions Limited, Auckland, New Zealand). The drape was fitted to cover the wound bed with 2 cm overhang during testing and sealed using an isobutylene copolymer sealant (General Sealants Incorporated, City of Industry, CA, USA). An inline waste collection canister was included to collect SWF from the test rig. A third pressure sensor (P′) was positioned on the system controller (SC) to record the actual vacuum pressure delivered to the test rig. SWF was administered to the rig via a multi-channel syringe pump (SyringeSix, NE1600, Lab Supply, Dunedin, New Zealand) equipped with 60 mL disposable syringes (Livingstone International Pty Ltd, Sydney, Australia).

The pump and vacuum pressure sensor (P1, P2 and P′) were connected to the SC comprised of two development boards that were developed in-house, linked to a personal computer (Fig. 3). The SC was additionally linked to an air bleed valve (ABV) (KSV2WM-5A, Koge Europe GmbH, Ellingen, Germany) and the ABV pre-programmed to maintain the target vacuum pressure. Vacuum pressure data was recorded using an analogue data acquisition system incorporated into the SC to record real-time vacuum pressure measurements from the pressure sensor at the base of the wound bed (P1 and P2) and delivered vacuum pressure (P′). Data was captured as a CSV file and exported to Excel (Microsoft Corporation, Redmond, WA, USA) for analysis.

2.3. Test method

Test articles were cut to 10 cm × 10 cm squares, rehydrated in 0.9% saline until fully hydrated, then placed in the base of the test rig. A 20 cm × 30 cm silicon-based non-adherent dressing (Silflex®, Advancis Medical, Nottinghamshire, UK) was cut to 10 cm × 10 cm and placed on top of the test article. Reticulated polyurethane NPWT interface foam (V.A.C. Granufoam™, KCI Corporation, San Antonio, TX, USA) was cut

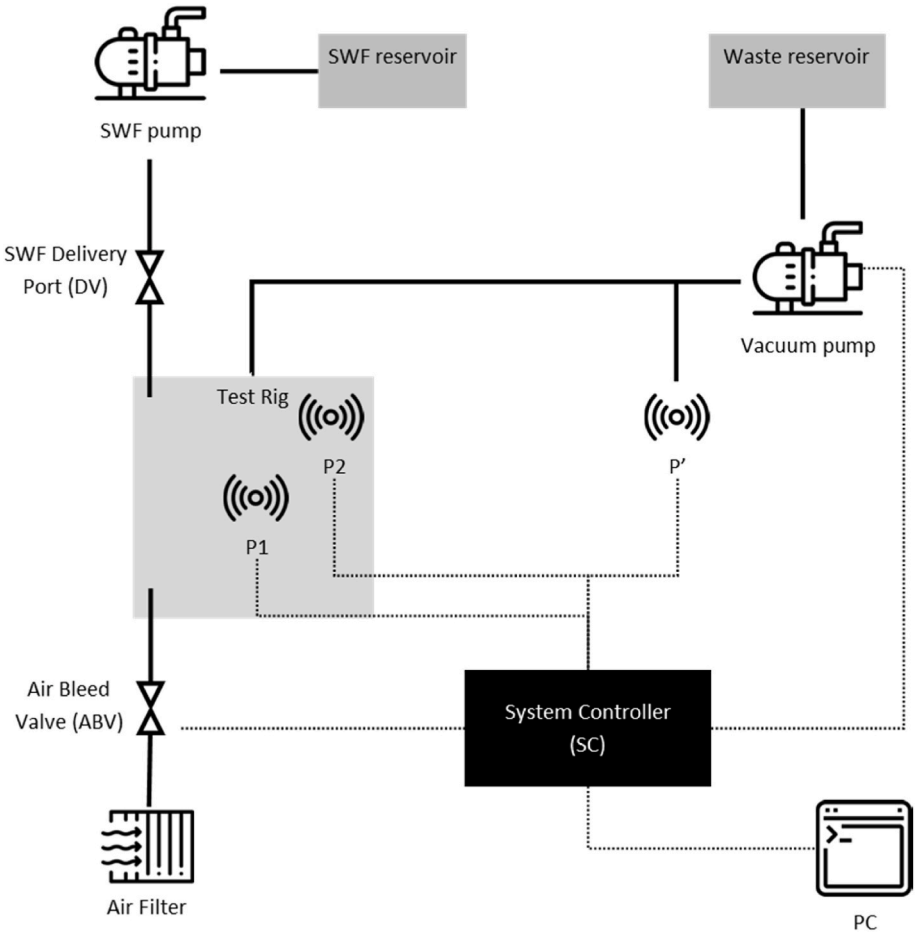

Fig. 3. Schematic of the test rig system. SWF = simulated wound fluid; PC = personal computer; SC = system controller; ABV = air bleed valve; P1 = central pressure sensor; P2 = peripheral pressure sensor; P' = inline pressure sensor. Solid lines = tubing connections; dashed lines = electronic connections.

to 10 cm × 10 cm and placed in contact with the non-adherent dressing, then the polyurethane drape applied. SWF was delivered at a rate of 1.3 mL/cm^2/24 h, 0.09 mL/min (Dealey et al., 2006). The vacuum pressure at P1, P2 and P′ were recorded every 5 min for 48 h. Test articles were tested at vacuum pressures −125 mmHg and −75 mmHg with three repeats per test condition. Each test was done with and without SWF. The observed vacuum pressure drop (% Reduction) was calculated from the formula:

$$\%\ Reduction = (P' - P1)/P'$$

3. Results

A consistent level of vacuum pressure was supplied to the test rig (P′) during all testing with the samples receiving the target 125 mmHg and 75 mmHg set pressure (Table 1).

The observed vacuum pressure (P1 and P2) of OFM was greatly reduced relative to the target vacuum pressure over the course of 48 h (Fig. 4A).

OFM gave observed vacuum pressures of 58.8 ± 0.27 mmHg (53.1% ± 0.21%) and 35.6 ± 0.23 mmHg (53.0% ± 0.30%) at 125 mmHg and 75 mmHg, respectively (Fig. 5.).

Additionally, there was a difference between the P1 and P2 sensors at both 125 mmHg and 75 mmHg, suggesting the delivered vacuum pressure was heterogenous across the wound surface due to interference of the OFM sample (Fig. 4A). OFMHF represents a second-generation dermal matrix, that includes additional novel fenestrations to optimize wound exudate egress and allow compatibility with NPWT (Fig. 1). OFMHF showed little variation between the supplied pressure at P′ and P1+P2 the observed pressure at the simulated wound bed (the mean of the P1 and P2 vacuum pressure sensors) at 125 mmHg and 75 mmHg, with a percentage vacuum pressure decrease of 0.2% ± 0.01% and 0.3% ± 0.01%, respectively (Table 1). A representative plot of observed vacuum pressure versus time (Fig. 4B) showed consistent delivery of the vacuum pressure to both P1 and P2 across the 48 h of testing OFMHF. Collagen/ORC reduced the observed vacuum pressure, with a greater effect at lower set vacuum pressures. At 75 mmHg, the observed vacuum pressure was reduced by 32.4 ± 0.34, while at 125 mmHg, a 16.1% ± 0.04% reduction was observed (Table 1). CWD also reduced the observed vacuum pressure at 75 mmHg and 125 mmHg by 57.0 ± 0.24 and 51.5% ± 0.31%, respectively (Table 1). At 125 mmHg, the observed vacuum pressure at P1 (40.4 ± 0.29 mmHg) was substantially less than the P2 vacuum pressure (81.3 ± 0.15 mmHg) (Table 1 and Fig. 4D), and these differences were less apparent at a vacuum setpoint of 75 mmHg.

Administration of SWF during testing generally changed the observed vacuum pressure reductions for the four test articles (Table 2). However, the same overall trend in device performance was observed (Fig. 5). OFM showed a substantial decrease in P1 (61.7 ± 0.38 mmHg) and P2 (98.3 ± 0.18 mmHg) pressures (Table 2), similar to the experiments conducted without SWF. OFMHF showed a slight reduction between the supplied vacuum pressure P' (126.2 ± 0.01 mmHg) and P1 (118.6 ± 0.12 mmHg) and P2 (119.1 ± 0.11 mmHg) (Fig. 5C) and largely consistent P1 and P2 over 48 h (Fig. 4F). Collagen/ORC showed a vacuum pressure reduction of 15.8% ± 0.00% at P1 and 13.6% ± 0.00% at P2 (Table 2). However, unlike in the experiments conducted without SWF, collagen/ORC showed an inconsistent P1 and P2 over 48 h in experiments with SWF (Fig. 4G). In line with the experiments conducted at 125 mmHg without SWF, CWD showed a substantial difference in pressure reduction between P1 (68.5% ± 0.00%) and P2 sensors (41.9% ± 0.00%). At a delivered pressure of 75 mmHg and in the presence of SWF, OFMHF again showed the lowest reduction in observed vacuum pressure at P1 (11.9% ± 0.00%) and P2 (12.1% ± 0.00%) compared to OFM (65.5% ± 0.00% and 54.7% ± 0.00%), collagen/ORC (26.6% ± 0.00% and 16.3% ± 0.00%), and CWD (70.8% ± 0.00% and 44.0% ± 0.00) (Table 2).

Fig. 6 illustrates the test articles after 6 and 24 h of incubation at 37 °C in SWF. OFMHF, OFM and CWD largely retained their structural integrity while collagen/ORC (Fig. 4F) showed notable gelling and loss of structural integrity.

4. Discussion

NPWT is a well-established treatment modality for wound closure and its mechanisms and clinical applications have been well documented. In a systematic review of molecular mechanisms of action of NPWT, Glass et al. concluded that NPWT leads to a modulation of cytokines to an anti-inflammatory profile, an increase in the expression of growth factors (VEGF, FGF2, TGF-β, PDGF), and a decrease in proteases MMP-1, -2, -9, and -13 (Glass et al., 2014). These molecular changes were hypothesized to promote angiogenesis and granulation tissue formation, leading to improved healing outcomes. To identify the optimal vacuum pressure of NPWT systems, Morykwas et al. (1997) investigated the effects of different vacuum pressures on blood flow using a laser Doppler probe placed inside porcine wounds. NPWT was applied up to 400 mmHg in increasing 25 mmHg increments with a 15-min dwell time. The authors reported an optimal vacuum pressure of 125 mmHg, that gave rise to a 4-fold increase of blood perfusion (Kairinos et al., 2013).

Similarly, Borgquist et al. (2011) investigated the effect of vacuum pressures on both wound fluid removal and reduction in wound size, demonstrating that a vacuum pressure of 125 mmHg was optimal for wound fluid removal, while 75 mmHg was optimal for reducing wound size. This study led the authors to conclude that any vacuum pressures outside these limits (75 and 125 mmHg) was suboptimal for healing. For example, a vacuum pressure of 70 mmHg was shown to be approximately 20% less effective at wound fluid removal than a vacuum pressure of 125 mmHg, and a vacuum pressure of 50 mmHg was approximately 30% less effective at reducing wound size than a vacuum pressure of 75 mmHg. Scherer et al. (2008) further validated a vacuum pressure of 125 mmHg by showing that wound volume could be decreased by ~80% at this optimal vacuum pressure. Collectively these

Table 1
Results summary without SWF, mean recorded vacuum pressure over the 48 h test period from n = 3 independent experiments. 'P1' and 'P2' refer to the mean central or peripheral vacuum pressures, respectively. 'P1+P2' refers to the mean vacuum pressure of P1 and P2 combined. Relative vacuum pressure reduction (%) is relative to P'. Errors represent SEM.

	P' (mmHg)	P1 (mmHg)	P2 (mmHg)	P1+P2 (mmHg)	P1 reduction (%)	P2 reduction (%)	P1+P2 reduction (%)
125 mmHg							
OFM	125.5 ± 0.02	52.8 ± 0.28	64.9 ± 0.40	58.8 ± 0.27	57.9 ± 0.00	48.3 ± 0.00	53.1 ± 0.21
OFMHF	125.6 ± 0.02	125.7 ± 0.02	125.1 ± 0.02	125.4 ± 0.02	−0.1 ± 0.00	0.4 ± 0.00	0.2 ± 0.01
Collagen/ORC	126.1 ± 0.02	105.4 ± 0.06	106.2 ± 0.07	105.8 ± 0.05	16.4 ± 0.00	15.7 ± 0.00	16.1 ± 0.04
CWD	125.6 ± 0.02	40.4 ± 0.29	81.3 ± 0.15	60.9 ± 0.39	67.8 ± 0.00	35.3 ± 0.00	51.5 ± 0.31
75 mmHg							
OFM	75.6 ± 0.02	33.0 ± 0.29	38.2 ± 0.29	35.6 ± 0.23	56.4 ± 0.00	49.5 ± 0.00	53.0 ± 0.30
OFMHF	75.1 ± 0.01	75.3 ± 0.01	74.5 ± 0.01	74.9 ± 0.01	−0.2 ± 0.00	0.8 ± 0.00	0.3 ± 0.01
Collagen/ORC	76.0 ± 0.03	49.1 ± 0.41	53.8 ± 0.30	51.5 ± 0.26	35.5 ± 0.01	29.3 ± 0.00	32.4 ± 0.34
CWD	75.5 ± 0.04	24.6 ± 0.34	40.3 ± 0.20	32.4 ± 0.24	67.4 ± 0.00	46.6 ± 0.00	57.0 ± 0.31

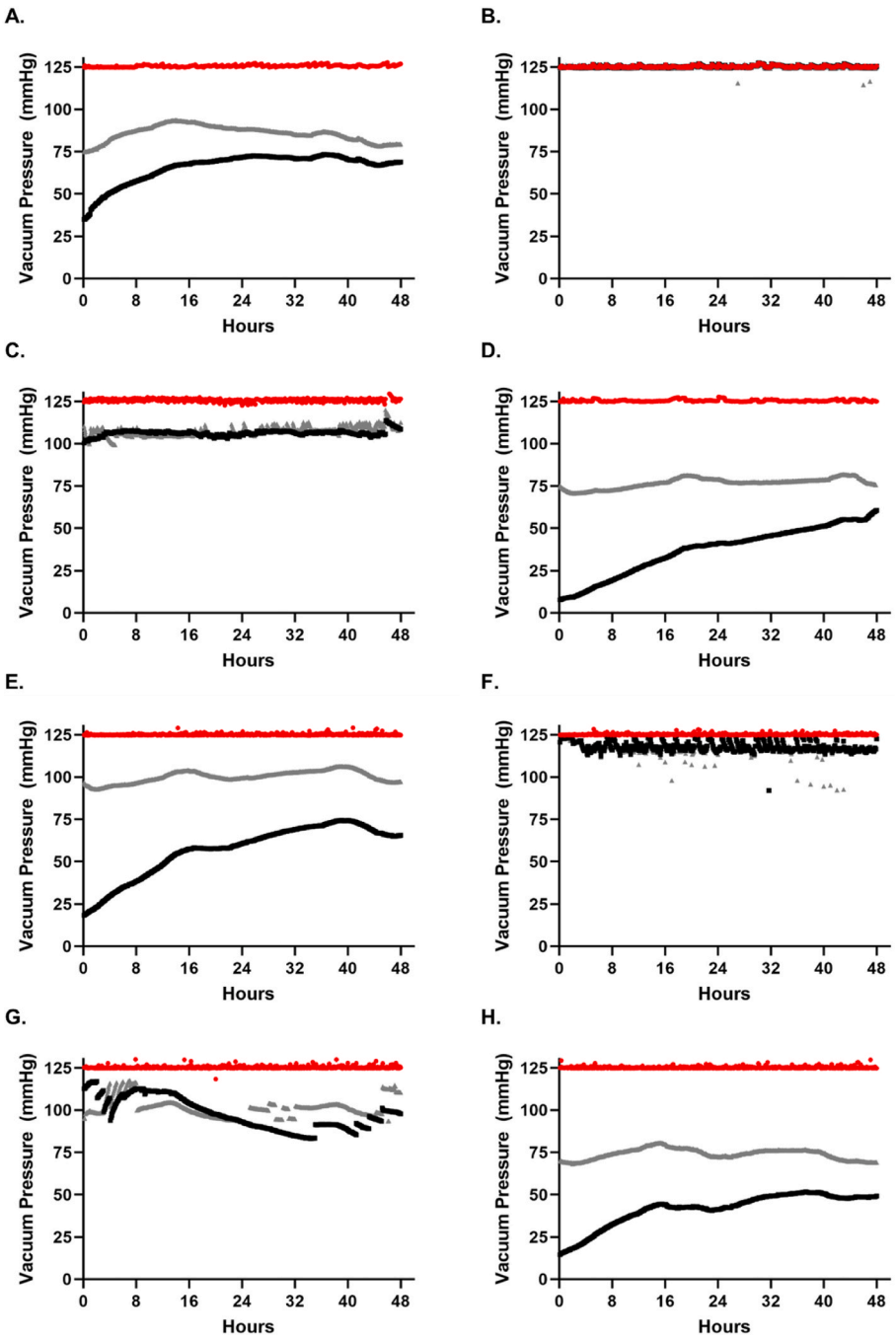

Fig. 4. Representative plots of vacuum pressure versus time for each test sample, **A.** OFM, **B.** OFMHF, **C.** collagen/ORC, and **D.** CWD tested at 125 mmHg, and with the addition of SWF (**E., F., G.** and **H.**). Red circles represent P′, black squares represent P1, and grey triangles represent P2.

studies, among others, were foundational in establishing the current standard of care of 125 mmHg, the most common NPWT pressure as identified from systematic review (Glass et al., 2014).

Based on the findings of this study, the use of a collagen-based dermal matrices under NPWT systems may significantly reduce the effectiveness of the delivered vacuum pressure, thus reducing blood flow, wound fluid removal and wound size reduction. To our knowledge there are no published studies that have investigated the influence of collagen-based matrices on the observed vacuum pressure when used in conjunction with NPWT.

There is one unpublished *in vitro* report characterizing the impact of collagen/ORC on observed vacuum pressure (Westmoreland et al., 2017). The results of this study were consistent with the findings herein, namely that collagen/ORC resulted in a reduction in the observed vacuum pressure (29.57% reduction at 125 mmHg) and the percentage reduction in observed vacuum pressure was inversely proportional to the set vacuum pressure. Consistent with our findings, the authors also noted gelling of the collagen/ORC in the presence of SWF, which likely contributes to the observed vacuum pressure reduction, vacuum pressure fluctuations, and variance across the wound bed. One way to reduce the risk of interference of a collagen-based matrix with the optimal vacuum pressure has been to design NPWT compatibility into collagen-based matrices. For example, OFMHF (Fig. 1), was designed to be compatible with NPWT, and was shown herein to be unobstructive to the delivered vacuum pressure (Fig. 5).

Both NPWT and dermal matrices have similar clinical goals of

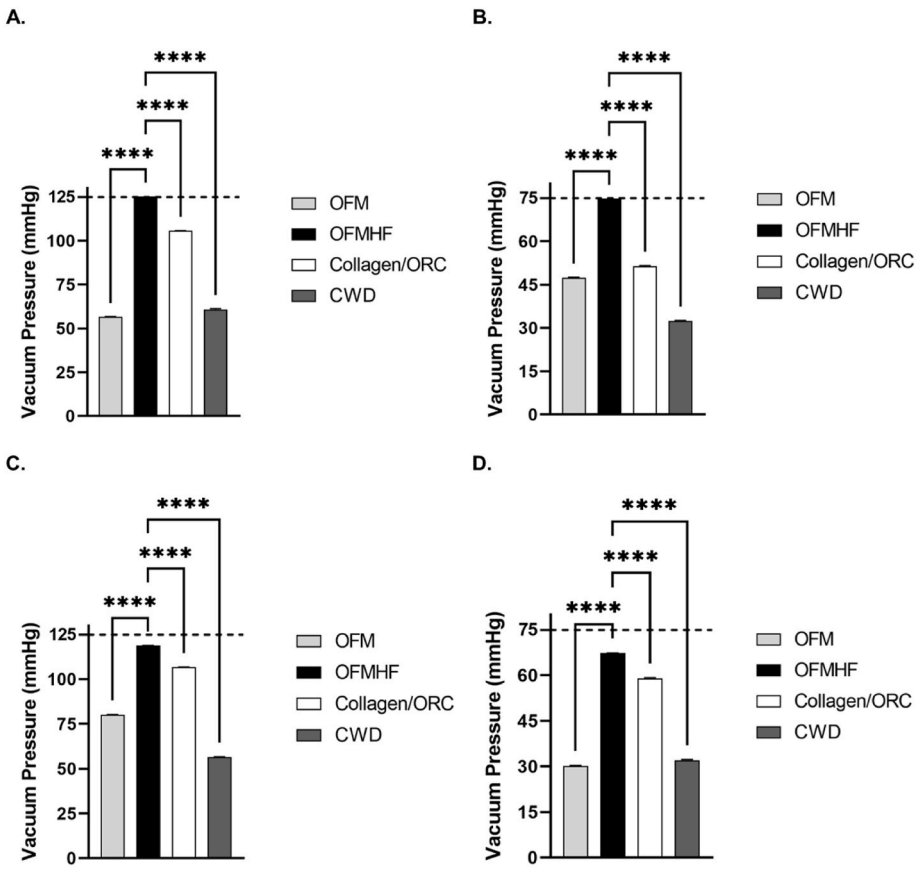

Fig. 5. Observed vacuum pressure (mean over the 48 h test period) measured for each of the test articles tested at 125 mmHg and 75 mmHg, without simulated wound fluid (SWF) (**A.** and **B.**) and 125 mmHg and 75 mmHg, with SWF (**C.** and **D.**). Error bars represent standard error of the mean from triplicate experiments. Horizontal dotted lines represent the set vacuum pressure (125 mmHg or 75 mmHg). Differences between treatment groups determined using ordinary one-way ANOVAs (Dunnett's multiple comparisons test), where; '*' = $p < 0.05$; '**' = $p < 0.01$, '***' = $p < 0.001$, '****' = $p < 0.0001$.

Table 2
Results Summary with SWF, mean recorded vacuum pressure over the 48 h test period from n = 3 independent experiments. 'P1' and 'P'2 refer to the mean central or peripheral vacuum pressures, respectively (n = 3). 'P1+P2' refers to the mean vacuum pressure of P1 and P2 combined. Relative vacuum pressure reduction (%) is relative to P'. Errors represent SEM.

	P' (mmHg)	P1 (mmHg)	P2 (mmHg)	P1+P2 (mmHg)	P1 reduction (%)	P2 reduction (%)	P1+P2 reduction (%)
125 mmHg							
OFM	125.3 ± 0.05	61.7 ± 0.38	98.2 ± 0.18	80.0 ± 0.37	50.7 ± 0.000	21.5 ± 0.00	36.1 ± 0.30
OFMHF	126.2 ± 0.1	118.6 ± 0.12	119.1 ± 0.11	118.9 ± 0.08	6.0 ± 0.00	5.3 ± 0.00	5.7 ± 0.13
Collagen/ORC	125.3 ± 0.03	105.4 ± 0.25	108.2 ± 0.17	106.8 ± 0.15	15.8 ± 0.00	13.6 ± 0.00	14.7 ± 0.12
CWD	125.9 ± 2.0	39.6 ± 10.2	73.2 ± 13.6	56.4 ± 20.6	68.5 ± 8.1	41.9 ± 10.7	55.2 ± 16.4
75 mmHg							
OFM	75.6 ± 0.04	26.1 ± 0.22	34.3 ± 0.22	30.2 ± 0.17	65.5 ± 0.00	54.7 ± 0.00	60.1 ± 0.22
OFMHF	76.5 ± 0.06	67.4 ± 0.11	67.3 ± 0.12	67.4 ± 0.08	11.9 ± 0.00	12.1 ± 0.00	12.0 ± 0.09
Collagen/ORC	75.2 ± 0.02	55.2 ± 0.22	63.0 ± 0.12	59.1 ± 0.14	26.6 ± 0.00	16.3 ± 0.00	21.5 ± 0.19
CWD	75.3 ± 0.02	22.0 ± 0.16	42.2 ± 0.21	32.1 ± 0.22	70.8 ± 0.00	44.0 ± 0.00	57.4 ± 0.29

stimulating cellular processes, increasing tissue perfusion, and building robust tissue. It is unsurprising then that many practitioners have recognized the potential synergistic effects between the two technologies, such that simultaneous use of the two technologies is now widespread (Lehrman, 2020; Loh et al., 2020; Applewhite et al., 2018; Cifuentes, 2022). It is interesting to speculate on the potential synergy between the two technologies. For example, dECM-based dermal matrices are known to contain a variety of pro-angiogenic growth factors (Dempsey et al., 2019), and like NPWT, dECMs lead to improved blood vessel formation in regenerating tissues (Irvine et al., 2011). ECM-based matrices, like OFM, scaffold cellular infiltration, migration and proliferation to build replacement tissue (Overbeck et al., 2020). It is possible then that the local biophysical effects exerted by NPWT may aid cellular infiltration of dermal matrices by drawing cells into the scaffold. Studies have shown that the application of NPWT accelerates processes of cellular invasion of collagen-based matrices, potentially leading to more rapid healing when the two technologies are used in tandem (Baldwin et al., 2009; Potter et al., 2008). Given the biological properties of ECM-based devices (Badylak, 2007), and impact of NPWT on the regenerating wound, further studies are warranted to deconvolute and substantiate the possible synergy between these two technologies.

5. Conclusion

This study demonstrates that the use of collagen-based dermal matrices with NPWT systems may lead to a sub-optimal vacuum pressure within the wound bed that may decelerate healing and fluid management. Limitations of current collagen-based and dECM based dermal matrices can be overcome by thoughtfully designing dermal matrices for simultaneous use with NPWT. A novel presentation of a dECM dermal matrix, OFMHF, did not substantially reduce the observed vacuum

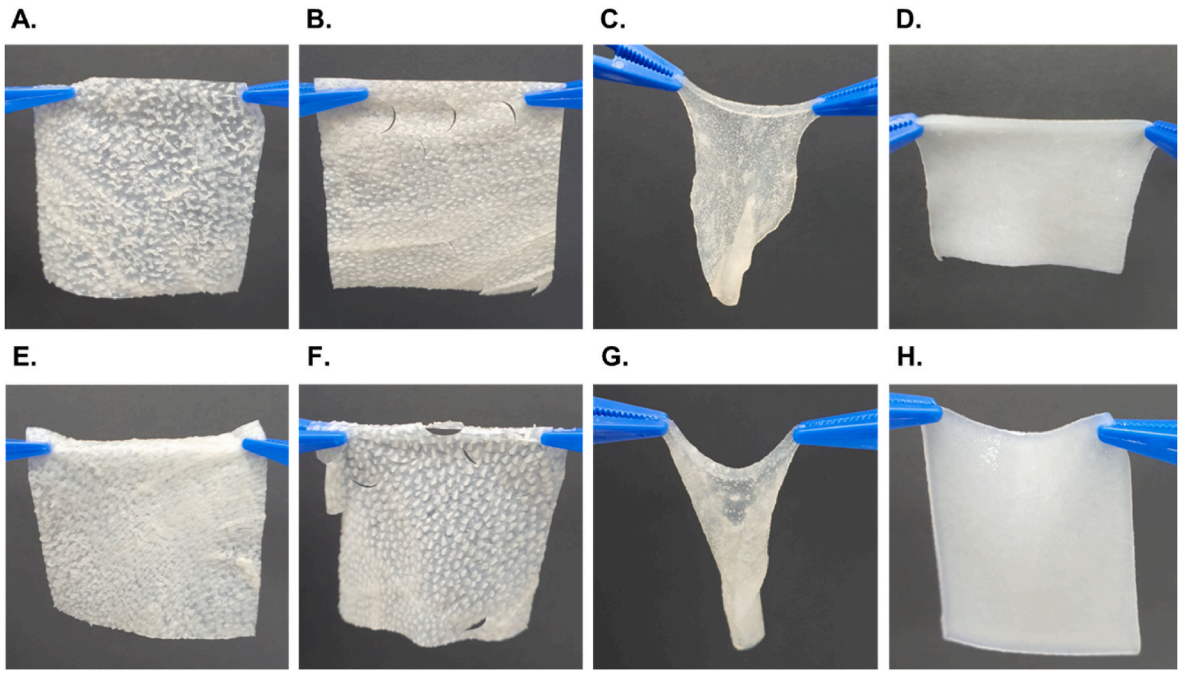

Fig. 6. Representative images of test articles incubated at 37 °C with SWF at 6 h (**A.** OFM, **B.** OFMHF, **C.** collagen/ORC, **D.** CWD) and 24 h (**E.** OFM, **F.** OFMHF, **G.** collagen/ORC, **H.** CWD).

pressure during simulated use. This device presentation may be better optimized for the synergistic use of both technologies to improve healing outcomes.

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CRediT authorship contribution statement

Robert W.F. Veale: Writing – review & editing, Writing – original draft, Visualization, Validation, Resources, Project administration, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Tarek Kollmetz:** Writing – review & editing, Writing – original draft, Visualization, Investigation, Data curation, Conceptualization. **Navid Taghavi:** Writing – review & editing, Writing – original draft, Investigation. **Claudia G. Duston-Fursman:** Writing – original draft, Validation, Resources, Methodology, Investigation. **Matthew T. Beeson:** Writing – original draft, Validation, Resources, Methodology, Investigation. **Dorrin Asefi:** Software, Resources, Methodology, Conceptualization. **Henry D. Chittock:** Software, Resources, Methodology. **Ananth S. Vikranth:** Methodology, Investigation. **Shane G. Dowling:** Writing – review & editing, Supervision, Methodology, Conceptualization. **Sandi G. Dempsey:** Writing – review & editing, Supervision, Project administration, Data curation. **Hamish J. Rose:** Resources, Methodology, Conceptualization. **Isaac T.T. Mason:** Resources, Methodology, Conceptualization. **Barnaby C.H. May:** Writing – review & editing, Writing – original draft, Visualization, Supervision, Project administration, Methodology, Funding acquisition, Formal analysis, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: RWFV, HC, SGD, IM, HJR and BCHM are shareholders of Aroa Biosurgery Limited (Auckland, New Zealand). RWFV, TK, NT, CGD, HC, SGD, SGD, HJR, IM, and BCHM are paid employees of Aroa Biosurgery Limited (Auckland, New Zealand). BCHM is listed as inventor on patent US8415159B2, owned by Aroa Biosurgery Limited.

Data availability

Data will be made available on request.

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