ORIGINAL ARTICLE

Comparison of bacteria and fungus-binding mesh, foam and gauze as fillers in negative pressure wound therapy – pressure transduction, wound edge contraction, microvascular blood flow and fluid retention

Malin Malmsjö[1], Richard Ingemansson[2], Sandra Lindstedt[2] & Lotta Gustafsson[1]

1 Department of Ophthalmology, Lund University, Lund, Sweden
2 Department of Cardiothoracic Surgery, Lund University, Lund, Sweden

Abstract

Bacteria- and fungus-binding mesh binds with and inactivates bacteria and fungus, which makes it an interesting alternative, wound filler for negative pressure wound therapy (NPWT). This study was conducted to compare the performance of pathogen-binding mesh, foam and gauze as wound fillers in NPWT with regard to pressure transduction, fluid retention, wound contraction and microvascular blood flow. Wounds on the backs of 16 pigs were filled with pathogen-binding mesh, foam or gauze and treated with NPWT. The immediate effects of 0, −40, −60, −80 and −120 mmHg, on pressure transduction and blood flow were examined in eight pigs using laser Doppler velocimetry. Wound contraction and fluid retention were studied during 72 hours of NPWT at −80 and −120 mmHg in the other eight pigs. Pathogen-binding mesh, gauze and foam provide similar pressure transduction to the wound bed during NPWT. Blood flow was found to decrease 0·5 cm laterally from the wound edge and increase 2·5 cm from the wound edge, but was unaltered 5·0 cm from the wound edge. The increase in blood flow was similar with all wound fillers. The decrease in blood flow was more pronounced with foam than with gauze and pathogen-binding mesh. Similarly, wound contraction was more pronounced with foam, than with gauze and pathogen-binding mesh. Wound fluid retention was the same in foam and pathogen-binding mesh, while more fluid was retained in the wound when using gauze. The blood flow 0·5–5 cm from the wound edge and the contraction of the wound during NPWT were similar when using pathogen-binding mesh and gauze. Wound fluid was efficiently removed when using pathogen-binding mesh, which may explain previous findings that granulation tissue formation is more rapid under pathogen-binding mesh than under gauze. This, in combination with its pathogen-binding properties, makes this mesh an interesting wound filler for use in NPWT.

Introduction

Negative pressure wound therapy (NPWT) accelerates wound healing by initiating a cascade of interrelated biological reactions in the wound edge that ultimately lead to wound healing. Initially, the wound is filled with a wound filler

Key Messages

- the aim of this study was to investigate the effects on the wound when using pathogen-binding mesh as the wound filler in NPWT

- sixteen healthy domestic pigs of both sexes, with a mean body weight of 70 kg, were used in the experiments
- short experiments were performed on eight pigs to study the immediate effects of continuous NPWT, including pressure transmission to the wound bed and the effects on microvascular blood flow around the wound edge
- the results of this study suggest that pressure is equally well transmitted through pathogen-binding mesh, foam and gauze
- in this study, the wound contraction was similar for pathogen-binding mesh and gauze, and slightly greater for foam
- the reason why greater wound contraction is seen with foam is probably its open, spongy texture
- the results of this study show that once negative pressure is discontinued, the wounds remain contracted, suggesting that the wound edge is remodelled during the course of the therapy
- the manner in which NPWT is administered should be based on the type of wound and its vascularity
- when applied to a wound cavity, as in this study, NPWT will create a force on the tissue that may open up vascular beds, increasing blood flow
- the use of foam may be beneficial in maximising hypoperfusion, thus stimulating angiogenesis, while pathogen-binding mesh or gauze may be preferable when the vascularisation of the tissue is in doubt, and there is a risk of ischaemia
- the results of this study showed that less wound fluid remained in the wounds treated with hydrophobic mesh than in wounds treated with gauze, which may explain the difference in the amount of granulation tissue formed under these wound fillers
- gauze is often used because of its mouldability and ease of application to large and irregular wounds
- pathogen-binding mesh is also a woven material, and the application techniques are similar to those of gauze
- the use of pathogen-binding mesh as wound filler in NPWT may be especially beneficial when infection prevents wound healing
- in conclusion, pathogen-binding mesh is an interesting alternative wound filler in NPWT owing to its antimicrobial properties, especially in light of the fact that NPWT itself does not reduce the bacterial load, and may, in some instances, even increase bacterial load
- the wound edge blood flow and contraction are similar when using pathogen-binding mesh and gauze in NPWT
- wound effluents contain many factors that are known to inhibit wound healing, and the efficient removal of wound fluid by pathogen-binding mesh, compared with gauze, may explain previous findings of more rapid granulation tissue formation under pathogen-binding mesh than under gauze
- efficient wound fluid removal in combination with its pathogen-binding properties makes hydrophobic mesh an interesting candidate for wound fillers in NPWT

(commonly gauze or foam) to allow pressure to be transmitted to and evenly distributed over the wound bed. The wound is then sealed with an adhesive film dressing and a drain is connected to a vacuum pump. Wound fluid is withdrawn and collected in a canister. One of the fundamental effects of NPWT is believed to be the induction of mechanical deformation of the tissue upon wound contraction (1–3), which in turn affects the microvascular blood flow around the wound edges (2,4–6), stimulating angiogenesis (7,8) and the formation of granulation tissue (2) to expedite wound healing.

There is a common misconception that NPWT controls or reduces the bacterial burden in the wound. In an initial study on pig wounds inoculated with human *Staphylococcus aureus* and *Staphylococcus epidermidis* a reduction in bacterial counts during the course of NPWT was reported (2). However, no clinical studies since then have been able to confirm the early in vivo findings of Morykwas et al. (9–12), and some have even reported an increase in bacterial numbers during NPWT (9,13,14). NPWT has been shown to cause a shift in the bacterial species towards biofilm-producing organisms such as *S. aureus* and *S. epidermidis* (9,10,12). It has been hypothesised that occlusion and negative pressure create relative hypoxia, thus promoting anaerobes and a shift in microorganism populations (12). Furthermore, the gauze used in NPWT has been a particular type of cotton gauze (Kerlix AMD), which may provide pathogen-binding control because it is impregnated with polyhexamethylene biguanide (15). However, the current recommendation is that NPWT should not be used in isolation to control wound infections (www.npwtexperts.com).

Foam and gauze are the materials most commonly used as wound filler in NPWT. It is believed that the type of wound filler used for NPWT can determine the effects on wound healing. Pathogen-binding mesh may constitute an interesting alternative wound filler in NPWT. Such mesh makes use of the hydrophobic interaction to remove pathogenic bacteria. Bacteria have cell surface structures making them hydrophobic, which allows them to adhere to tissue in the initial phase of wound infection. Hydrophobic pathogen-binding mesh is coated with a fatty acid derivative that affords the dressing strongly hydrophobic properties. Wound bacteria are thus irreversibly bound to the dressing when they come into contact with the hydrophobic fibres in the moist wound environment (16). Pathogen-binding mesh also adsorbs and inactivates a wide variety of bacteria, for example, *Staphylococcus aureus* and *Pseudomonas aeruginosa*, as well as fungi, and has been shown to reduce the microbial load in wounds (17,18). The pathogen-binding mesh is primarily used for treatment and prevention of hard to heal, acute traumatic, post-surgical wounds and burn. It is a non-allergic and non-toxic alternative for reducing the microbial load in open wounds. The mesh binds and inactivates bacteria and fungus without the development of resistance among microorganisms. It should be noted that differences in the external structure of microbes result in different degree of cell surface hydrophobicity. The age of the microorganisms and the growth conditions are two factors that affect the hydrophobicity.

The aim of this study was to investigate the effects on the wound when using pathogen-binding mesh as the wound filler

in NPWT. Pressure transduction to the wound bed, wound contraction, the effects on microvascular blood flow in the wound edge and fluid retention by the dressing were measured, and the results were compared with those obtained with gauze and foam.

Materials and methods

Animals

Sixteen healthy domestic pigs of both sexes, with a mean body weight of 70 kg, were used in the experiments. Short experiments were performed on eight pigs to study the immediate effects of continuous NPWT, including pressure transmission to the wound bed and the effects on microvascular blood flow around the wound edge. These pigs were anaesthetised for 12 hours. Longer experiments were performed on the other eight pigs to study wound contraction and wound fluid retention in the filler after 72 hours of NPWT.

Ethics

The experimental protocol for this study was approved by the Ethics Committee for Animal Research, Lund University, Sweden. All animals received humane care in compliance with the European Convention on Animal Care.

Negative pressure wound therapy

The wounds were filled with saline-moistened AMD gauze (Kendall Kerlix™ AMD™, Tyco Healthcare Group, Mansfield, MA), pathogen-binding mesh (Sorbact®, Abigo Medical AB, Gothenburg, Sweden) or black polyurethane foam with an open-cell structure (VAC® Granufoam®, KCI, San Antonio, TX). The AMD gauze is a woven cotton material, whereas the pathogen-binding mesh is a woven acetate material. The foam is made of polyurethane and has an open-cell structure with a pore size of 400–600 μm. The drainage tube was inserted into the upper surface of the wound filler, and an adhesive drape was used to seal the wound. The drainage tube was then connected to a vacuum source.

Experiments for examining the immediate effects of NPWT on pressure transduction and blood flow

Anaesthesia was performed as described previously (19). Circular wounds, 6 cm in diameter, extending into muscle tissue, were created on each pig's back.

Pressure measurements

The negative pressure on the wound bed, underneath the wound filler, was measured using a saline-filled pressure catheter. The tip of the catheter was sutured to the centre of the bottom of the wound (Figure 1). The pressure catheter was connected to a custom-built pressure gauge with which the pressure at the bottom of the wound could be monitored.

Negative pressures of −40, −60, −80 and −120 mmHg were applied and the pressure at the bottom of the wound was measured.

Microvascular blood flow measurements

Microvascular blood flow was measured using laser Doppler velocimetry (Figure 1), using a four-channel Perimed PF5010 LDPM unit (Perimed, Stockholm, Sweden). The laser Doppler filaments were inserted into the tissue, 0·5, 2·5 and 5·0 cm laterally from the wound edge. Blood flow was measured after reaching steady state, which normally took about 1 minute. Laser Doppler measurements were performed before and after the application of negative pressures of −40, −60, −80 and −120 mmHg. The results were recorded as arbitrary perfusion units and then transformed into the percent change in relation to the baseline values (before the application of negative pressure).

Experiments for examining wound contraction and fluid retention during 72 hours of NPWT

Anaesthesia was performed as described previously (20). Circular wounds, 6 cm in diameter, extending into subcutaneous tissue, were created on each pig's back. The wounds were treated at either −80 mmHg, −120 mmHg or not subjected to negative pressure (0 mmHg). After the experiments, the animals were euthanised with a lethal intravenous dose (60 mmol) of potassium chloride. We did not do cultures because this is an acute wound model and we expect it not to be infected.

Wound contraction

The wound was measured in a horizontal and vertical direction (Figure 1). The mean value of these measurements was calculated. Measurements were performed before the application of negative pressure, immediately after the negative pressure was applied (0 hours), every 24 hours during NPWT and after NPWT had been discontinued. Wound contraction by NPWT could thereby be assessed.

Wound fluid retention

The wound filler was weighed before and after each experiment. The difference in weight was calculated as a measure of the fluid absorbed by the filler and thus remaining in the wound.

Limitations

Reduction in bacterial load by pathogen-binding mesh has been shown in previous studies (17,18) and was now the scope of this study. The aim of this study was to determine the suitability for pathogen-binding mesh to be used for NPWT with regard to its physiological properties, compared with presently used wound fillers for NPWT. It has clearly been shown that the acute wound model in the pig is well suited for studying the physiological properties of wound fillers (21). When

A. Measurement of wound bed pressure
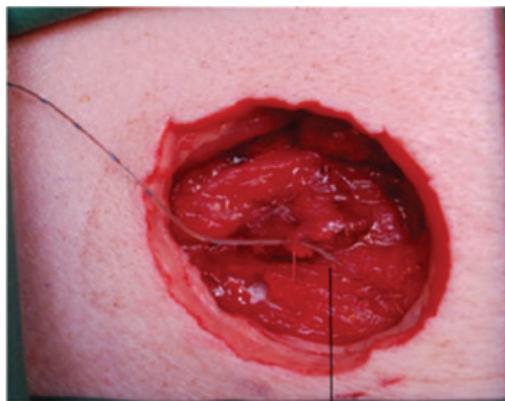
Saline-filled catheter

B. Laser Doppler velocimetry measurements
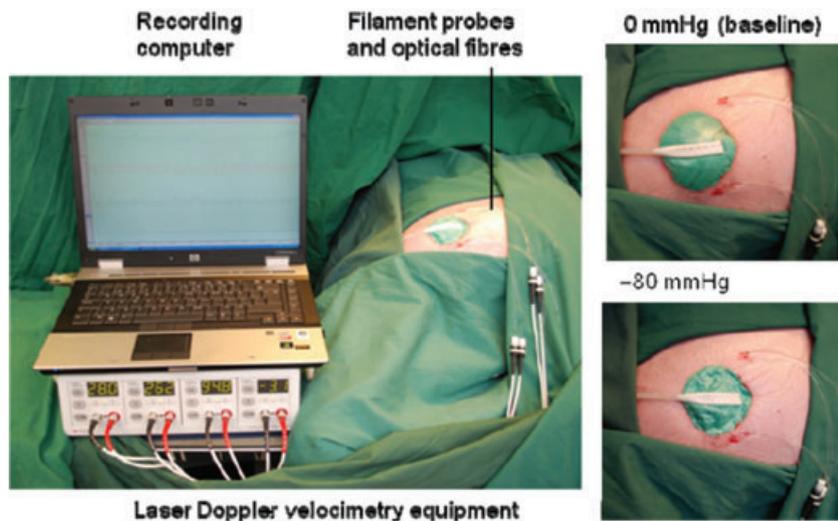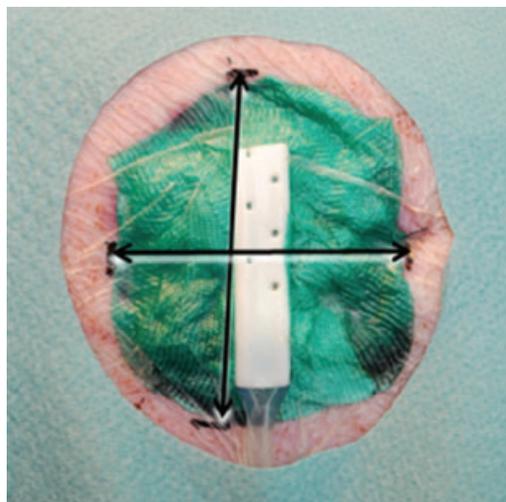

C. Wound contraction measurements

Figure 1 (A) Position of the saline-filled catheter for measurements of wound bed pressure during negative pressure wound therapy (NPWT). (B) Experimental set-up for laser Doppler velocimetry measurements of wound edge microvascular blood flow during NPWT. The laser Doppler filament probes are inserted 0·5, 2·5 and 5 cm into the edge of the wound and the laser Doppler velocimetry unit is connected to a computer in which the data are stored. (C) Measurement of wound contraction during NPWT in a wound on the pig's back. The lines indicate the diameters of the wound, which were measured before the application of negative pressure, immediately after negative pressure was applied (0 hours), every 24 hours during NPWT and after NPWT was discontinued. The wound surface area was calculated from the measured diameters.

these pig wounds have been inoculated with bacteria to study the effects of NPWT on bacterial burden, the results have not been reliable (22).

Calculations and statistics

Calculations were performed using GraphPad 5·0 software (San Diego, CA). Statistical analysis was performed using the Mann–Whitney test when comparing two groups, and the Kruskal–Wallis test with Dunn's post-test for multiple comparisons when comparing three groups or more. Significance was defined as $P < 0.05$. All differences referred to in the text are statistically significant. Results are presented as the means of eight measurements ± standard error of the mean.

Results

Pressure delivery to the wound bed

The pressure on the wound bed increased gradually with increasing level of applied negative pressure from −20 to −160 mmHg. Pathogen-binding mesh, gauze and foam provided similar pressure transduction to the wound bed during NPWT. Detailed results are shown in Figure 2.

Wound contraction

The wounds contracted immediately when negative pressure was applied, and remained contracted during the 72 hours of NPWT. Wound contraction was more pronounced with foam than with gauze or pathogen-binding mesh. Detailed results are shown in Table 1 and Figure 3.

Microvascular blood flow around the wound edge

NPWT induced a decrease in the microvascular blood flow 0·5 cm from the wound edge and an increase 2·5 cm from

Table 1 Wound contraction during NPWT in a porcine peripheral wound model. Wounds were sealed as for NPWT using pathogen-binding mesh, foam or gauze, and then treated at negative pressures of −80 and −120 mmHg for 72 hours. The reduction in wound diameter was calculated as a percentage of the area before negative pressure was applied

	Pathogen-binding mesh	Gauze	Foam
−80 mmHg	93·8 ± 0·4%	92·4 ± 0·9%	90·3 ± 1·6%*
−120 mmHg	93·1 ± 1·4%	94·6 ± 0·7%	89·7 ± 1·3%*

NPWT, negative pressure wound therapy.
*P < 0·05; values are presented as means ± standard error of the mean ($n = 8$).

the wound edge. The decrease in blood flow 0·5 cm from the wound edge was greater with the foam (−33·0 ± 4·1% for foam) than with gauze or pathogen-binding mesh (−22·2 ± 4·5% for gauze and −21·4 ± 5·8% for pathogen-binding mesh, at −80 mmHg, $P < 0.05$). The type of wound filler had no effect on the blood flow response 2·5 cm from the wound edge (increase in blood flow: 57·8 ± 13·7% for foam, 49·6 ± 8·0% for gauze and 60·1 ± 16·6% for pathogen-binding mesh, $P =$ n.s.). The blood flow 5·0 cm from the wound edge was not affected by NPWT. The blood flow changed gradually with increasing level of negative pressure until it reached a steady state. Detailed results are shown in Table 2 and Figure 4.

Wound fluid retention

More wound fluid was retained in the gauze filler than in the foam or pathogen-binding mesh. This means that there is more fluid in the wound during NPWT with gauze. Wound fluid retention was similar when using pressures of −80 and −120 mmHg. Detailed results are shown in Figure 5.

Discussion

Pressure delivery to the wound bed

The wound filler transmits the negative pressure to the wound bed. The results of this study suggest that pressure is equally well transmitted through pathogen-binding mesh, foam and gauze. This is in agreement with a previous study, in which we found the same pressure transmission with gauze and foam (19).

Wound contraction

One of the fundamental effects of NPWT is believed to be the deformation of the wound edge tissue as the wound contracts (7,23,24). These mechanical effects are thought to result in shearing forces at the wound–dressing interface, which affect the cytoskeleton (25), and initiate a signalling cascade that ultimately leads to granulation tissue formation and wound healing. In this study, the wound contraction was similar for pathogen-binding mesh and gauze, and slightly greater for foam. The reason why greater wound contraction is seen with foam is probably its open and spongy texture.
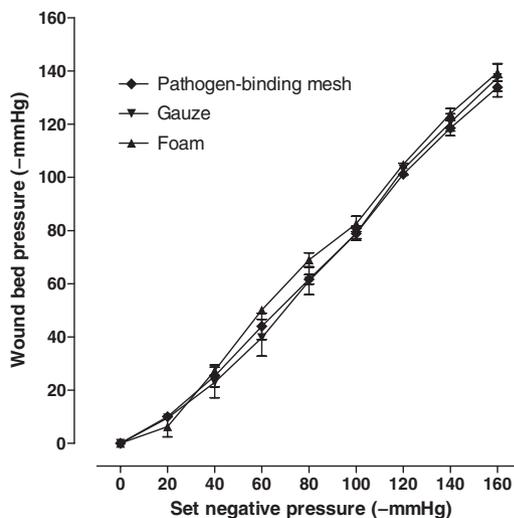

Figure 2 Wound bed pressure underneath the wound filler during negative pressure wound therapy in wounds filled with pathogen-binding mesh, foam or gauze. Results are the means of eight experiments ± standard error of the mean. It can be seen that all wound fillers provided similar pressure transfer to the wound bed.
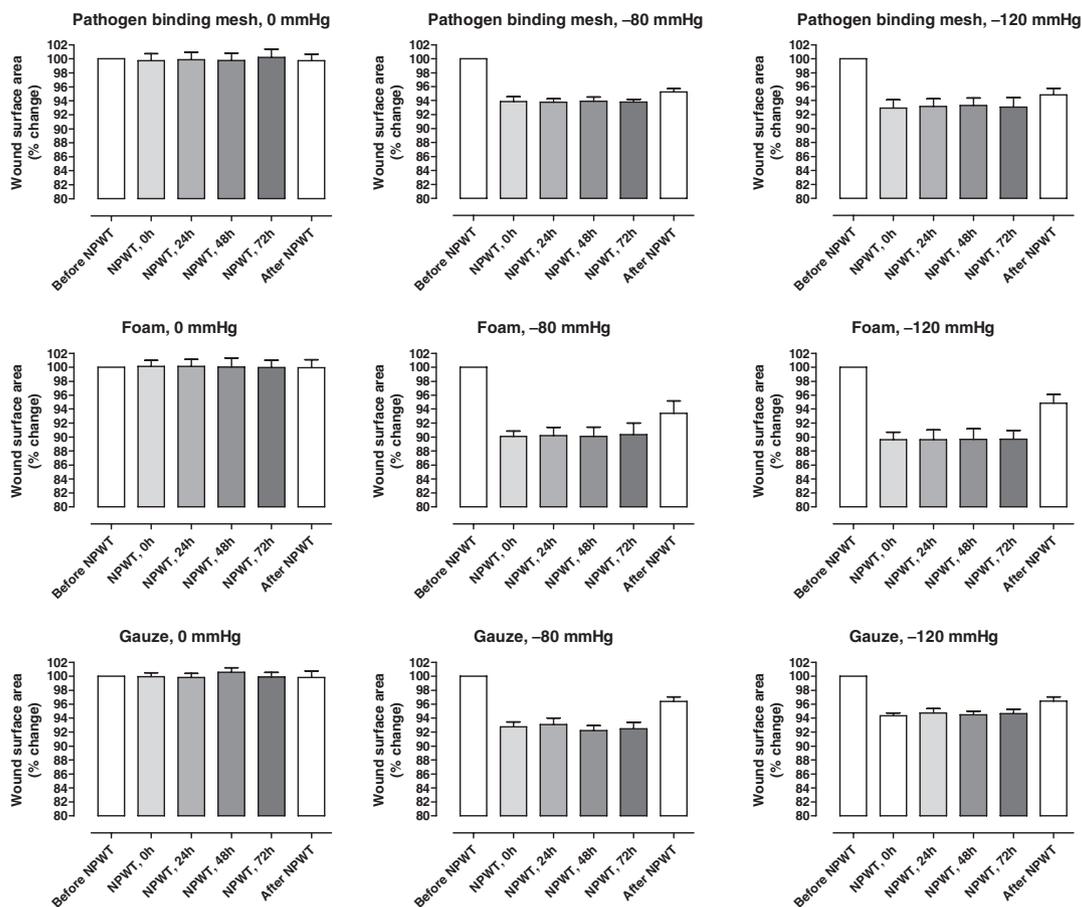
Figure 3 Wound contraction during negative pressure wound therapy (NPWT) in a porcine peripheral wound model. Wounds were sealed as for NPWT using pathogen-binding mesh, foam or gauze, and then treated at negative pressures of −80 and −120 mmHg, or not subjected to negative pressure (0 mmHg). Measurements of wound diameter were performed before the application of negative pressure (before NPWT), immediately after negative pressure was applied (0 hours), every 24 hours during NPWT and after negative pressure was discontinued (after NPWT). The reduction in wound diameter was calculated as a percentage of the area before negative pressure was applied. Values are presented as means ± standard error of the mean ($n = 8$). Note the greater wound contraction with foam than with gauze and pathogen-binding mesh, and the fact that the wounds treated with negative pressure remained contracted after discontinuation of the treatment.

Table 2 Microvascular blood flow in the wound edge during negative pressure wound therapy at various pressures using pathogen-binding mesh, foam and gauze, measured using laser Doppler velocimetry

	−40 mmHg	−60 mmHg	−80 mmHg	−120 mmHg	
Measurements were performed 0·5 cm from the wound edge					
Pathogen-binding mesh	−12 ± 6%	−19 ± 6%	−21 ± 6%	−15 ± 8%	n.s.
Gauze	−13 ± 3%	−17 ± 4%	−22 ± 5%	−24 ± 4%	n.s.
Foam	−27 ± 5%	−31 ± 4%	−33 ± 4%	−36 ± 4%	n.s.
	Foam versus gauze or mesh $P < 0.05$	Foam versus gauze or mesh $P < 0.05$	Foam versus gauze or mesh $P < 0.05$	Foam versus gauze or mesh $P < 0.05$	
Measurements were performed 2·5 cm from the wound edge					
Pathogen-binding mesh	+45 ± 18%	+54 ± 18%	+60 ± 17%	+61 ± 16%	n.s.
Gauze	+38 ± 6%	+50 ± 9%	+50 ± 8%	+54 ± 9%	n.s.
Foam	+40 ± 8%	+58 ± 15%	+58 ± 14%	+54 ± 12%	n.s.
	n.s.	n.s.	n.s.	n.s.	

Microvascular blood flow is expressed as percent change relative to baseline values. Values are presented as means ± standard error of the mean ($n = 8$).
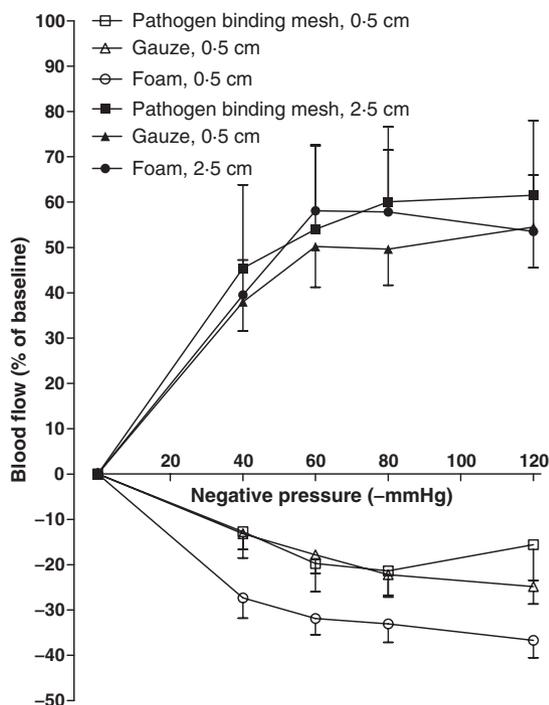
Figure 4 Microvascular blood flow in the wound edge during negative pressure wound therapy at various pressures using pathogen-binding mesh, foam and gauze, measured using laser Doppler velocimetry. Measurements were performed 0·5 and 2·5 cm from the wound edge. Microvascular blood flow is expressed as percent change relative to baseline values. Note that there was no difference in blood flow response between the different types of wound filler 2·5 cm from the wound edge, whereas at 0·5 cm, less hypoperfusion was observed with pathogen-binding mesh and gauze than with foam.
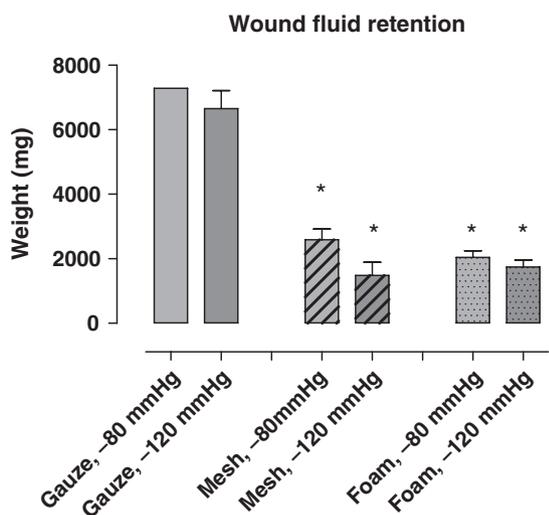

Figure 5 Wound fluid retention in the pathogen-binding mesh, gauze and foam after negative pressure wound therapy for 72 hours. Results are the means of eight experiments ± standard error of the mean. It can be seen that more fluid is captured by the gauze than by the pathogen-binding mesh and foam.

The results of this study show that once negative pressure is discontinued, the wounds remain contracted, suggesting that the wound edge is remodelled during the course of the therapy. It has been shown previously that patients receiving NPWT with foam are approximately 2·5 times more likely to show a reduction in wound area after 1 week of therapy than patients receiving standard moist wound therapy (26). The mechanical effects of NPWT may be important for the entire wound-healing process, as early reduction in the size of the wound has been shown to be correlated with improved final wound healing (26).

Microvascular blood flow in the wound edge

The microvascular blood flow close to the wound edge (0·5 cm from the edge) decreased following the application of subatmospheric pressure with all three fillers, the greatest decrease being seen with foam. The mechanism by which NPWT decreases blood flow in superficial tissue may have been identified in recent work (27,28). When the wound contracts, the tissue of the wound edge collapses towards the suction force, and the pressure around the rim of the wound edge increases (27), probably leading to a decrease in blood flow.

There are both advantages and disadvantages of the hypoperfusion caused by NPWT. It is well known that reduced blood flow stimulates angiogenesis and granulation tissue formation, which in turn facilitate the process of wound healing (1,29). However, several clinical problems are associated with hypoperfusion caused by NPWT. In tissues with already impaired circulation, the further decrease in blood flow may result in ischaemia, and it has been suggested that NPWT should be applied with caution to tissues with compromised vascularity (4). Some advocate that NPWT is contraindicated if there is any doubt about the vascularity of the tissue (30,31). The manner in which NPWT is administered should therefore be based on the type of wound and its vascularity.

A similar increase in microvascular blood flow was observed for all three fillers 2·5 cm from the wound edge. It is believed that the stimulation of blood flow by NPWT may be important in ensuring adequate oxygenation and nutrient supply, and the removal of waste products from the healing wound (32). Increased blood flow may also facilitate the penetration of antibiotics to the otherwise poorly perfused tissue. The mechanism by which blood flow is increased 2·5 cm from the wound edge during NPWT has not yet been fully elucidated. Kairinos et al. showed that the pressure on the tissue was the same as atmospheric pressure at a distance greater than 2 cm from the wound edge, and thus tissue pressure is not a plausible explanation of the present findings (27). The mechanical effects of NPWT cause wound contraction, as shown in previous studies (7,23,24,33,34) and in this study. When applied to a wound cavity, as in this study, NPWT will create a force on the tissue that may open up vascular beds, increasing blood flow. Indeed, previous experimental studies have shown that small arterioles and capillaries in the wound edge tissue open upon the application of negative pressure (7,35).

Clinical implications of the different effects of the different wound fillers on blood flow

Two different strategies can be used to tailor NPWT to alter the degree of hypoperfusion generated in the wound edge: changing the negative pressure level or the type of wound filler. Pathogen-binding mesh and gauze caused less pronounced hypoperfusion than foam, which may be the result of the smaller degree of wound contraction than with foam. The use of foam may be beneficial in maximising hypoperfusion, thus stimulating angiogenesis, while pathogen-binding mesh or gauze may be preferable when the vascularisation of the tissue is in doubt, and there is a risk of ischaemia.

Wound fluid retention

The suction force generated by the negative pressure leads to active drainage of exudate from the wound. Wound fluid was efficiently removed from the wound through foam and hydrophobic pathogen-binding mesh. The reason for this is that both foam and the pathogen-binding mesh are hydrophobic allowing the fluid to be removed by the NPWT. It is probably also due to the more open structure of foam and mesh than gauze. However, more fluid remained in the gauze. The removal of fluid from the wound is advantageous as it reduces cytokines and other compounds that are inhibitory to wound healing, such as proteolytic enzymes and metalloproteinases (36,37). It has recently been shown that granulation tissue formation is greater under pathogen-binding mesh that under gauze (38). One explanation of this may be the difference in texture and thus the imprints made in the tissue by pathogen-binding mesh. The results of this study showed that less wound fluid remained in the wounds treated with hydrophobic mesh than in wounds treated with gauze, which may explain the difference in the amount of granulation tissue formed under these wound fillers.

Ease of application

Gauze is often used because of its mouldability and ease of application to large and irregular wounds. The use of gauze in NPWT has been described by Jeffery when treating wounds caused by landmines and other explosive devices in military personnel (39). Pathogen-binding mesh is also a woven material, and the application techniques are similar to those of gauze.

The use of pathogen-binding mesh in managing wound infection during NPWT

A number of studies have shown an increase in bacterial load during NPWT (9,13,14). Pathogen-binding mesh is known to bind and inactivate bacteria and fungi (16), and therefore constitutes an interesting alternative wound filler in NPWT. Furthermore, there is evidence that NPWT results in a shift in the bacterial community towards biofilm-producing organisms such as *S. aureus* and *S. epidermidis* (9,10,12), which is exactly the kind of bacteria that pathogen-binding mesh is known to counteract (17,18). The use of pathogen-binding mesh as wound filler in NPWT may be especially beneficial when infection prevents wound healing.

Conclusions

Pathogen-binding mesh is an interesting alternative wound filler in NPWT owing to its antimicrobial properties, especially in light of the fact that NPWT itself does not reduce the bacterial load, and may, in some instances, even increase bacterial load. The wound edge blood flow and contraction are similar when using pathogen-binding mesh and gauze in NPWT. Wound effluents contain many factors that are known to inhibit wound healing, and the efficient removal of wound fluid by pathogen-binding mesh, compared with gauze, may explain previous findings of more rapid granulation tissue formation under pathogen-binding mesh than under gauze. Efficient wound fluid removal in combination with its pathogen-binding properties makes hydrophobic mesh an interesting candidate for wound fillers in NPWT.

References

1. Argenta LC, Morykwas MJ. Vacuum-assisted closure: a new method for wound control and treatment: clinical experience. *Ann Plast Surg* 1997;**38**:563–76; discussion 577.
2. Morykwas MJ, Argenta LC, Shelton-Brown EI, McGuirt W. Vacuum-assisted closure: a new method for wound control and treatment: animal studies and basic foundation. *Ann Plast Surg* 1997;**38**:553–62.
3. Morykwas MJ, Simpson J, Punger K, Argenta A, Kremers L, Argenta J. Vacuum-assisted closure: state of basic research and physiologic foundation. *Plast Reconstr Surg* 2006;**117**:121S–6S.
4. Kairinos N, Voogd AM, Botha PH, Kotze T, Kahn D, Hudson DA, Solomons M. Negative-pressure wound therapy II: negative-pressure wound therapy and increased perfusion. Just an illusion? *Plast Reconstr Surg* 2009;**123**:601–12.
5. Wackenfors A, Gustafsson R, Sjogren J, Algotsson L, Ingemansson R, Malmsjö M. Blood flow responses in the peristernal thoracic wall during vacuum-assisted closure therapy. *Ann Thorac Surg* 2005;**79**:1724–30; discussion 1730-1.
6. Wackenfors A, Sjogren J, Gustafsson R, Algotsson L, Ingemansson R, Malmsjö M. Effects of vacuum-assisted closure therapy on inguinal wound edge microvascular blood flow. *Wound Repair Regen* 2004;**12**:600–6.
7. Chen SZ, Li J, Li XY, Xu LS. Effects of vacuum-assisted closure on wound microcirculation: an experimental study. *Asian J Surg* 2005;**28**:211–7.
8. Greene AK, Puder M, Roy R, Arsenault D, Kwei S, Moses MA, Orgill DP. Microdeformational wound therapy: effects on angiogenesis and matrix metalloproteinases in chronic wounds of 3 debilitated patients. *Ann Plast Surg* 2006;**56**:418–22.
9. Khashram M, Huggan P, Ikram R, Chambers S, Roake JA, Lewis DR. Effect of TNP on the microbiology of venous leg ulcers: a pilot study. *J Wound Care* 2009;**18**:164–7.
10. Moues CM, Vos MC, van den Bemd GJ, Stijnen T, Hovius SE. Bacterial load in relation to vacuum-assisted closure wound therapy: a prospective randomized trial. *Wound Repair Regen* 2004;**12**:11–7.
11. Anesater E, Roupe KM, Robertsson P, Borgquist O, Torbrand C, Ingemansson R, Lindstedt S, Malmsjö M. The influence on wound contraction and fluid evacuation of a rigid disc inserted to protect exposed organs during negative pressure wound therapy. *Int Wound J* 2011;**8**:393–9.
12. Chester DL, Waters R. Adverse alteration of wound flora with topical negative-pressure therapy: a case report. *Br J Plast Surg* 2002;**55**:510–1.

13. Weed T, Ratliff C, Drake DB. Quantifying bacterial bioburden during negative pressure wound therapy: does the wound VAC enhance bacterial clearance? *Ann Plast Surg* 2004;**52**:276–9; discussion 279-80.

14. Borgquist O, Gustafsson L, Ingemansson R, Malmsjö M. Micro- and macromechanical effects on the wound bed of negative pressure wound therapy using gauze and foam. *Ann Plast Surg* 2010;**64**: 789–93.

15. Mueller SW, Krebsbach LE. Impact of an antimicrobial-impregnated gauze dressing on surgical site infections including methicillin-resistant *Staphylococcus aureus* infections. *Am J Infect Control* 2008;**36**:651–5.

16. Borgquist O, Ingemansson R, Lindstedt S, Malmsjö M. In process citation. *Lakartidningen* 2011;**108**:2372–5.

17. Vig S, Dowsett C, Berg L, Caravaggi C, Rome P, Birke-Sorensen H, Bruhin A, Chariker M, Depoorter M, Dunn R, Duteille F, Ferreira F, Martínez JM, Grudzien G, Hudson D, Ichioka S, Ingemansson R, Jeffery S, Krug E, Lee C, Malmsjo M, Runkel N, International Expert Panel on Negative Pressure Wound Therapy [NPWT-EP], Martin R, Smith J. Evidence-based recommendations for the use of negative pressure wound therapy in chronic wounds: steps towards an international consensus. *J Tissue Viability* 2011;**20** Suppl 1:S1–18.

18. Mussi C, Salvioli G. Clinical evaluation of Sorbact (bacteria absorbing dressing) in the treatment of infected pressure sores. *ACTA Vulnologica* 2004;**2**:9–11.

19. Malmsjo M, Ingemansson R, Martin R, Huddleston E. Negative-pressure wound therapy using gauze or open-cell polyurethane foam: similar early effects on pressure transduction and tissue contraction in an experimental porcine wound model. *Wound Repair Regen* 2009;**17**:200–5.

20. Borgquist O, Ingemansson R, Malmsjö M. Micro- and macromechanical effects on the wound bed by negative pressure wound therapy using gauze and foam. *Ann Plast Surg* 2010;**64**:789–93.

21. Borgquist O, Ingemansson R, Malmsjo M. Individualizing the use of negative pressure wound therapy for optimal wound healing: a focused review of the literature. *Ostomy Wound Manage* 2011;**57**:44–54.

22. Birke-Sorensen H, Malmsjo M, Rome P, Hudson D, Krug E, Berg L, Bruhin A, Caravaggi C, Chariker M, Depoorter M, Dowsett C, Dunn R, Duteille F, Ferreira F, Francos Martínez JM, Grudzien G, Ichioka S, Ingemansson R, Jeffery S, Lee C, Vig S, Runkel N, International Expert Panel on Negative Pressure Wound Therapy [NPWT-EP], Martin R, Smith J. Evidence-based recommendations for negative pressure wound therapy: treatment variables (pressure levels, wound filler and contact layer)–steps towards an international consensus. *J Plast Reconstr Aesthet Surg* 2011;**64 Suppl**:S1–16.

23. Etöz AÖY, Özcan M. The use of negative pressure wound therapy on diabetic foot ulcers: a preliminary controlled trial. *Wounds* 2004;**16**:264–9.

24. Campbell PE, Smith GS, Smith JM. Retrospective clinical evaluation of gauze-based negative pressure wound therapy. *Int Wound J* 2008;**5**:280–6.

25. Saxena V, Hwang CW, Huang S, Eichbaum Q, Ingber D, Orgill DP. Vacuum-assisted closure: microdeformations of wounds and cell proliferation. *Plast Reconstr Surg* 2004;**114**:1086–96; discussion 1097-8.

26. Lavery LA, Barnes SA, Keith MS, Seaman JW Jr, Armstrong DG. Prediction of healing for postoperative diabetic foot wounds based on early wound area progression. *Diabetes Care* 2008;**31**:26–9.

27. Kairinos N, Solomons M, Hudson DA. The paradox of negative pressure wound therapy - in vitro studies. *J Plast Reconstr Aesthet Surg* 2008;**63**:174–9.

28. Willy C, Gerngross H. Scientific background of the vacuum closure - an abstract. *Zentralbl Chir* 2004;**129** Suppl 1:S6.

29. Petzina R, Gustafsson L, Mokhtari A, Ingemansson R, Malmsjö M. Effect of vacuum-assisted closure on blood flow in the peristernal thoracic wall after internal mammary artery harvesting. *Eur J Cardiothorac Surg* 2006;**30**:85–9.

30. Venturi ML, Attinger CE, Mesbahi AN, Hess CL, Graw KS. Mechanisms and clinical applications of the vacuum-assisted closure (VAC) Device: a review. *Am J Clin Dermatol* 2005;**6**:185–94.

31. Attinger CE, Janis JE, Steinberg J, Schwartz J, Al-Attar A, Couch K. Clinical approach to wounds: debridement and wound bed preparation including the use of dressings and wound-healing adjuvants. *Plast Reconstr Surg* 2006;**117**:72S–109S.

32. Jonsson K, Jensen JA, Goodson WHI, Goodson WH 3rd, Scheuenstuhl H, West J, Hopf HW, Hunt TK. Tissue oxygenation, anemia, and perfusion in relation to wound healing in surgical patients. *Ann Surg* 1991;**214**:605–13.

33. Borgquist O, Ingemansson R, Malmsjö M. Micro- and macromechanical effects on the wound bed by negative pressure wound therapy using gauze and foam. *Ann Plast Surg.* 2010;**64**:789–93.

34. Malmsjo M, Ingemansson R. Similar biological effects of green and black polyurethane foam in negative pressure wound therapy: green foam facilitates monitoring of wound status, bleeding and exudate. In 20th Conference of the European Wound Management Association; 26–28 May. Geneva, Switzerland, 2010.

35. Ichioka S, Watanabe H, Sekiya N, Shibata M, Nakatsuka T. A technique to visualize wound bed microcirculation and the acute effect of negative pressure. *Wound Rep Reg* 2008;**16**:460–5.

36. Yager DR, Nwomeh BC. The proteolytic environment of chronic wounds. *Wound Repair Regen* 1999;**7**:433–41.

37. Armstrong DG, Jude EB. The role of matrix metalloproteinases in wound healing. *J Am Podiatr Med Assoc* 2002;**92**:12–8.

38. Malmsjo M, Ingemansson R, Lindstedt S. Use of hydrophobic pathogen-adsorbing mesh in negative pressure wound therapy provides thick significant granulation tissue without tissue ingrowth. *Eplasty.* In press.

39. Jeffery SL. Advanced wound therapies in the management of severe military lower limb trauma: a new perspective. *Eplasty* 2009;**9**:e28.