The effect of natural matrix biopolymer membrane on hard-to-heal venous leg ulcers: a pilot randomised clinical trial

Objective: The aim of this study was to evaluate the therapeutic effects of natural matrix biopolymer membrane (NMBM) in the treatment of venous leg ulcers (VLUs).
Method: Patients exhibiting one or more VLU were assigned to a test group receiving NMBM or to a control group receiving conventional treatment. Patients exhibiting venous insufficiency-related ulcers within 0.1–170cm^2 were included. Efficacy was assessed based on ulcer size and visual analogue scale (VAS) pain scores at baseline and at weeks one, two and four. Ulcer size and pain were compared between groups using a two-way ANOVA.
Results: In this study, 25 patients with 32 VLUs (NMBM group: 14 patients with 17 ulcers; control group: 11 patients with 15 ulcers) were included in the final analysis. At four weeks after baseline measurements, the mean percentage change in VLU area of patients in the NMBM group was 61.6% (95% CI: 40.3–82.9) compared with 84.1% (95% CI: 56.5–111.7) for control group patients. Additionally, the mean percentage change in VLU volume of NMBM group patients was 51.2% (95% CI: 31.8–70.6) compared with 84.0% (95% CI: 57.0–121.0) for control group patients. The NMBM group patients exhibited a mean decrease of 0.38 (95% CI: –0.85–1.61) in VAS pain score over four weeks, compared with a mean decrease of 0.13 (95% CI: –1.32–1.58) for control group patients. No significant differences in VLU area (p=0.210), volume (p=0.122) or VAS pain score (p=0.460) were shown between groups.
Conclusion: NMBM was found to be as effective and safe as the control group treatments. This pilot study suggests NMBM can be used safely to promote ulcer healing.
Declaration of interest: The authors have no conflicts of interest.

compression therapy • leg ulcer • natural matrix biopolymer membrane • treatment outcomes • venous insufficiency

Ulcers of the lower extremities are widespread among older people, commonly affecting patients over 65 years of age.[1,2] Hard-to-heal venous leg ulcers (VLUs) affect approximately 0.3–1% of the general adult population and 3–4% of patients aged 65–80 years.[3–6] The most common aetiology—applicable to approximately 80% of leg ulcer cases—is chronic venous insufficiency (CVI).[7] Other causes of leg ulcers include peripheral arterial disease, diabetes, recurrent cellulitis, rheumatoid arthritis, lymphoedema, vasculitis and other inflammatory ulcerations.[8–15] Rarer leg ulcer aetiologies include metabolic disorders and haematological disorders.[16–19] The vast majority of VLUs are hard-to-heal ulcers.[20,21]

Compression therapy is widely considered the standard treatment for VLUs, and its ability to heal them is clearly supported by a body of evidence from many studies.[22,23] Accompanied by the use of hydrocolloid, foam or silicone-based wound dressings, pain alleviation can be achieved.[24,25] Moreover, with many compression and dressing therapies, decreases in ulcer size have been shown in several trials.[26,27] However, superiority of one particular compression and dressing regimen over another has not yet been demonstrated in any study, and many studies have been of poor or insufficient quality.[28]

Natural matrix biopolymer membrane (NMBM) is a novel topical cream directed for the treatment of wounds and ulcers. Unlike other dressings, it is not an over-the-counter or prescription product. As NMBM contains no active ingredient, it is classified as a class IIb medical device. NMBM cream comprises a complex of natural waxes, sugars, lipids, amino acids and osmoregulators.[29,30] NMBM is hypothesised to facilitate wound healing by providing a clean, moist antimicrobial environment that supports autolytic debridement.[29]

In this study, the potential efficacy of a combined NMBM and compression therapy in reducing ulcer size and pain, as an alternative therapy for the treatment of VLUs, was evaluated.

Method

Ethical considerations

In accordance with all designated terms and conditions, this clinical trial was approved and registered with ClinicalTrials.gov (NCT01770509); the study was conducted in an ambulatory setting at the outpatient clinic of the Department of Dermatology, Sheba Medical Center, Israel. This study was approved by the Institutional Review Board (IRB) at our institution. All

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patients had signed and dated consent forms that had been approved by our IRB. In addition, all patients had given written and verbal consent to publish non-identifiable photographs of their conditions. Our intended sample size per treatment arm was 15 patients, with a desired allocation ratio of 1:1, to determine safety and basic efficacy of NMBM before expanding to a larger trial.

Inclusion and exclusion criteria

Participants recruited to the study were male and female patients aged 18–90 years who exhibited hard-to-heal VLUs, with an ankle–brachial index >0.7. Only hard-to-heal venous insufficiency-related ulcers, within the range of 0.1–170 cm^2 were included in the final analysis. Hard-to-heal VLUs were considered as a wound that had failed to produce anatomic and functional integrity within 6–8 weeks before enrolment. Only VLUs that were present for at least one month before the beginning of the trial were included in the analysis. In order to normalise the sample in our study, patients exhibiting haemoglobin levels <10.5g/dl, platelet counts <$100x10^9$/l or serum albumin levels <2.5g/dl were excluded from the study. Any patients who had diabetes with haemoglobin A1c (HbA1c) ≥8% were similarly excluded. Other exclusion criteria included: patients with known allergies to compounds used in the study; patients showing evidence of VLUs extending to underlying muscle, tendon or bone; and patients who had used investigational new drugs within 30 days preceding randomisation.

Patient care and clinical assessment

Block randomisation with a block size of one was used for patient allocation into either the NMBM or control group. After patients were randomly allocated on a 1:1 ratio, based on a single sequence of random assignments, they were given a trial cream, with both verbal and written instructions for its use. All patients applied their cream twice daily for four weeks.

The patients' pain and ulcer status were evaluated at baseline and after one, two and four weeks of therapy. Photographs of ulcers were taken with a plain, light background, using a digital camera at a consistent distance. As per standard methods, a disposable paper ruler was used to measure ulcer size (i.e. area and volume) for each patient by determining the longest wound length, width and maximal wound depth.[31] To assess the analgesic effects of NMBM, patients were asked to indicate their pain intensity level as a whole numeric value between 0 (no pain) and 10 (excruciating pain) on the visual analogue scale (VAS) during their weekly visits.[32,33] Stages of venous insufficiency were determined in accordance with the 2009 updated terminology for Clinical Etiological Anatomical Pathophysiological (CEAP) classification.[34] Other ulcer assessments included exudate consistency and presence of erythema, odour, necrosis and infection.

NMBM content, properties and application

NMBM is created to mimic lipid protective surfaces while also providing a low pH, high sugar content and high osmolality[35] to prevent and neutralise bacteria, remove nonviable tissue and encourage healing.[36–39] Its contents are: purified water, beeswax, carnauba wax, glycerin, sorbitol, lauric acid, glyceryl oleate, myristic acid, stearic acid, propylene glycol, sodium polyacrylate, ethylhexyl stearate, trideceth-6, potassium hydroxide, L-lysine HCl, glycine, histidine, arginine, fructose and phenoxyethanol. Phenoxyethanol, a standard preservative commonly used in the pharmaceutical and cosmetic industry, is added to prevent cream contamination. The NMBM product is an emulsion that creates a hypertonic film over wound surfaces that attracts water and other hypotonic liquids into the wound, keeping the wound site hydrated. NMBM also maintains a pH of 6.5–7 when applied to the wound.[40]

The test group patient cohort were provided with standard 40g tubes of NMBM, to be administered topically at the site of the wound twice daily for four weeks. All patients were instructed to wash the ulcer site with water and soap, dry with clean disposable tissue paper, apply the cream to coat the whole volume and cover the wound site with a compression stocking, in compliance with the manufacturers' instructions.[35] Patients were provided with enough NMBM to last the duration of the study.

In the control group, patients were given TenderWet Active (Medline Industries, US), TenderWet (Medline Industries, US), Tielle (Kinetic Concepts, Inc., US), Jelonet (Smith+Nephew plc., UK), Versiva (ConvaTec Group plc., UK), Biatain (Coloplast, Denmark) and Granugel (ConvaTec Group plc., UK). In cases where patients presented with a local infection, the antimicrobial alginate dressing Silvercel (Kinetic Concepts, Inc., US) was used. The dressings for each patient in the control group were selected in consultation with a single wound care nurse specialist. As with the NMBM cohort, patients in the control group were instructed to apply compression garments in accordance with the manufacturers' instructions.

To ensure proper wound care, all patients participated in individual 20-minute sessions where they received training and instructions on applying the NMBM cream or control dressings, along with dry, sterile gauze and compression garments. The nurse who randomised the patients was not blinded to the treatments. The physician performing the pain assessments and ulcer size calculations was blinded to treatment arms.

Statistical analysis

Differences in mean continuous parameters before and after treatment in each group were analysed via a two-way ANOVA (analysis of variance) test. Specifically, percentage changes in ulcer volume after four weeks of treatment time, in comparison with baseline measurements for both the NMBM and control groups, were determined. VAS pain measurements were treated

as interval data, as has been justified by others.[41,42] Accordingly, to assess differences between the NMBM and control group in terms of per-patient mean changes in degree of pain after four weeks of therapy, a two-way ANOVA test was conducted. Descriptive statistics of ordinal and dichotomous data (exudate consistency and presence of erythema, odour, necrosis and nonviable tissue or infection) for the NMBM and control group were also reported. Differences in ordinal and dichotomous data for both the NMBM and control groups at baseline and after four weeks of therapy were analysed using a two-sided Fisher's exact test; p-values of ≤0.05 were considered statistically significant. Statistical evaluation was performed using RStudio (version 3.4.1, RStudio, Inc., US).

Results

Patient recruitment

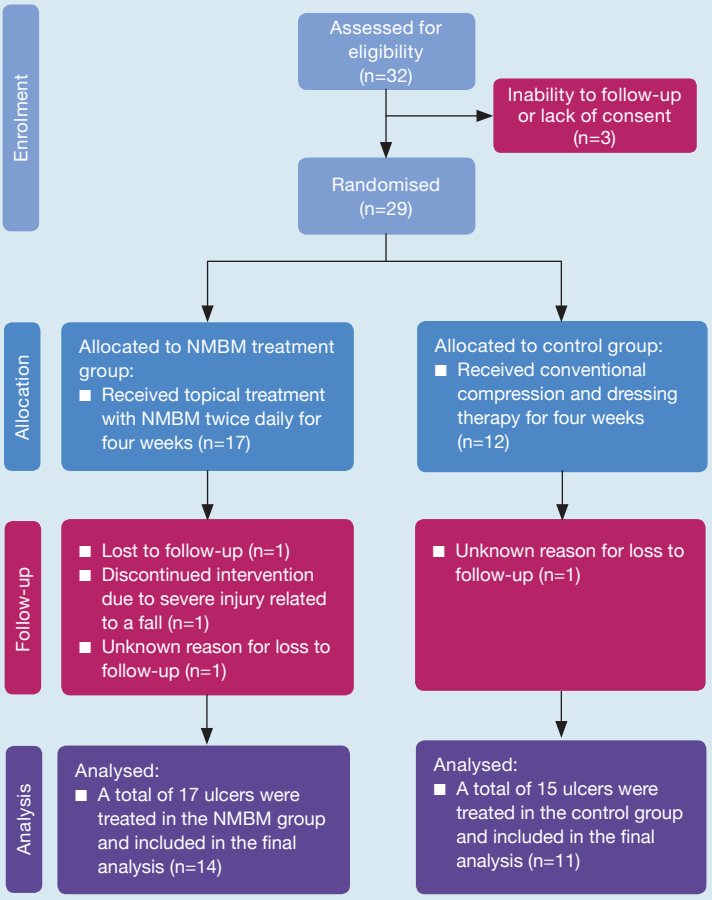
A total of 29 patients were recruited from February 2013 to December 2014 (see Fig 1 for a patient flow-chart providing the conceptual process of patient recruitment and assessment). Of the 29 patients recruited, 17 were randomised into the NMBM treatment group and 12 were randomised into the control group. Due to four patients being lost to follow-up and because of stringent eligibility criteria, only 14 patients in the NMBM group and 11 patients in the control group were included for clinical assessment of leg ulcers. From the NMBM group, three patients dropped out of the clinical trial for the following reasons: lost to follow-up (n=1), severe injury due to a fall (n=1) and unknown (n=1). Within the control group, one patient dropped out of the study for unknown reasons. Patients who dropped out of the study were still incorporated into the list of background and demographic characteristics provided in Table 1.

Analysis of the change in ulcer volume over the course of the study was completed on the remaining NMBM (n=14) and control patients (n=11). For certain patients in the control group, pain measurements were not available; accordingly, only 13 patients in the NMBM group and eight patients in the control group were included for pain measurements.

Ulcer characteristics

The 25 patients who continued with the study had a total of 35 ulcers that were treated over the course of the study. All patients exhibited hard-to-heal venous insufficiency-related ulcers, each within the range of 0.1–170cm^2 and classed as C6 active venous ulcers according the CEAP clinical classification scheme.[34] Of the 35 ulcers, 19 were treated with NMBM and compression therapy (54%) and 16 were treated in the control group (46%). However, in one patient in the NMBM group who had three ulcers, two were too small to meet our aforementioned ulcer inclusion criteria; accordingly, only 17 ulcers treated with NMBM were included in the final analysis. Concerning the control group, one ulcer from a patient with three ulcers was excluded from the final analysis due to measurement errors at the third and fourth follow-up; thus, a total of 15 ulcers in the control group were included in the final analysis.

Fig 1. Flow chart of patient recruitment process. Patient inclusion and exclusion criteria are based on previously published guidelines (NCT01770509). Flow chart was created in accordance with randomised clinical trial requirements with respect to CONSORT guidelines. NMBM—natural matrix biopolymer membrane



Control treatments consisted of various dressings according to the stage of the ulcer. Control group patients were treated with one or more of the following conventional therapies: Tenderwet active (n=3), Tenderwet (n=2), Tielle (n=3), Jelonet (n=1), Versiva (n=2), Biatain (n=3), Granugel (n=2) and Silvercel (n=2; patients with local infections). All patients adhered to their ulcer care instructions and there were no reported variances in wound dressing techniques.

Treatment-related changes in ulcer characteristics

Differences in ordinal scale and dichotomous ulcer characteristics between the NMBM group and the control group after four weeks of therapy are shown in Table 2. For all ordinal scale or dichotomous measurements (exudate consistency, erythema, odour, necrosis and nonviable tissue or infection), there were no significant differences between baseline and four-week follow-up measurements for both the NMBM and control groups.

Table 1. Patient demographics

Characteristic		NMBM (n=17)	Control (n=12)
Mean age, years±standard deviation		68±13.96	78±6.04
Age range, years		39–89	63–87
Sex, n (%)	Male	8 (47%)	7 (58%)
Comorbidities, n (%)	COPD	2 (11.8%)	7 (58%)
	DVT	1 (5.9%)	1 (8.3%)
	Dyslipidaemia	3 (17.6%)	3 (25%)
	HTN	4 (23.5%)	5 (41.7%)
	Hypothyroidism	2 (11.8%)	0 (0%)
	IHD	0 (0%)	1 (8.3%)
	Leg oedema	3 (17.6%)	0 (0%)
	MVR	0 (0%)	1 (8.3%)
	NIDMM	2 (11.8%)	3 (25%)
	Obesity	2 (11.8%)	2 (16.7%)
	PAF	0 (0%)	1 (8.3%)
	Psoriasis	1 (5.9%)	0 (0%)
	Thrombocytosis	2 (11.8%)	0 (0%)
Smoker, n (%)		3 (17.6%)	2 (16.7%)

NMBM—natural matrix biopolymer membrane; COPD—chronic obstructive pulmonary disease; DVT—deep vein thrombosis; HTN—hypertension; IHD—ischaemic heart disease; MVR—mitral valve replacement; NIDMM—noninsulin-dependent diabetes mellitus; PAF—paroxysmal atrial fibrillation

Treatment-related changes in ulcer size

Of the 17 ulcers included in the final analysis that were treated in the NMBM group, 14 (82.4%) showed reductions in size at the end of the four-week course of therapy. Of the 17 ulcers, four (23.5%) showed complete wound closure after four weeks. The mean percentage change in ulcer surface area, as compared with baseline, was 61.6% (95% CI: 40.3–82.9) for the NMBM group after four weeks of therapy (Fig 2a). The mean percentage change in ulcer volume, as compared with baseline, was 51.2% (95% CI: 31.8–70.6) for the NMBM group after four weeks of therapy (Fig 2b).

Of the 15 ulcers treated in the control group, nine (60%) showed reductions in size after four weeks, and two (13.3%) showed complete healing after four weeks of treatment. The percentage change in ulcer surface area, as compared with baseline, was 84.1% (95% CI: 56.5–111.7) for the control group after four weeks of therapy (Fig 2a). When comparing final ulcer volumes with baseline measurements, the mean percentage change was 84.0% (95% CI: 57.0–121.0) for the control group (Fig 2b).

After conducting a two-way ANOVA of the percentage changes of ulcer surface area after four weeks of therapy for the NMBM versus the control group, a p-value of p=0.210 was reported. Similarly, a two-way ANOVA of the percentage changes in ulcer volume after four weeks' therapy for the NMBM versus the control was conducted, yielding a p-value of p=0.122. Fig 3 and Fig 4 show healing of two representative ulcers over time with NMBM treatment.

Treatment-related changes in pain

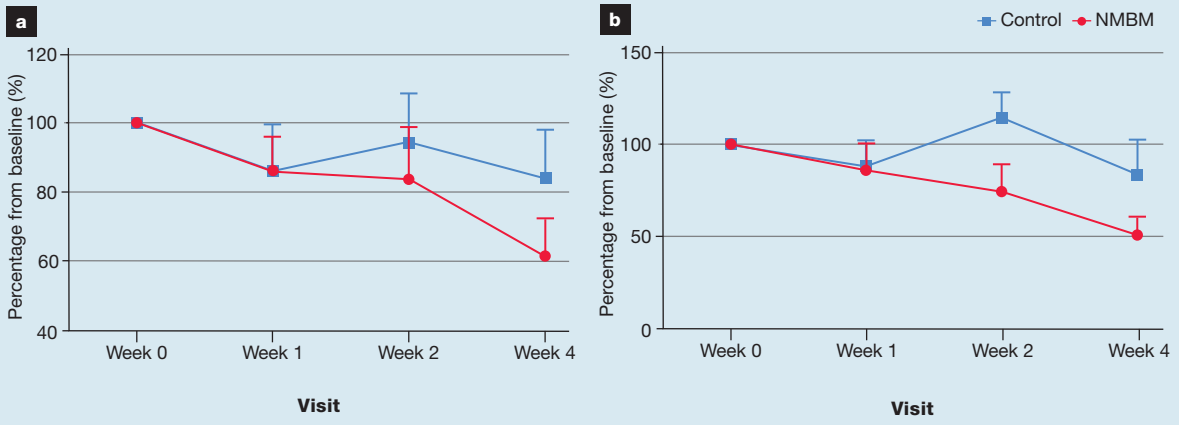
Of the 13 patients in the NMBM group who were followed up on pain reports at weeks 1, 2 and 4, five (38.5%) showed reductions in pain. The mean decrease in VAS pain score among patients in the NMBM group was 0.38 (95% CI: –0.85–1.61) after four weeks of NMBM therapy (Fig 5). Of the eight patients in the control group for whom pain reports were recorded at weeks 1, 2 and 4, five (62.5%) showed reductions in pain after

Table 2. Ulcer characteristics at baseline measurement and four-week follow-up

	NMBM (n=17)			Control (n=15)		
Measurement	Baseline	Follow-up	Difference (p-value)	Baseline	Follow-up	Difference (p-value)
Erythema, n (%)	10 (58.8%)	7 (41.2%)	0.494	8 (47.1%)	4 (26.7%)	0.264
Exudate, n (%)	16 (94.1%)	12 (70.6%)	0.175	15 (100%)	13 (86.7%)	0.483
None	1 (5.9%)	5 (29.4%)	0.175	0 (0%)	2 (13.3%)	0.483
Serous	13 (76.5%)	8 (47.1%)	0.157	13 (86.7%)	10 (66.6%)	0.390
Sanguineous	1 (5.9%)	2 (11.8%)	>0.999	0 (0%)	3 (20%)	0.224
Serosanguineous	0 (0%)	2 (11.8%)	0.485	1 (6.7%)	0 (0%)	>0.999
Purulent	2 (11.8%)	0 (0%)	0.485	1 (6.7%)	0 (0%)	>0.999
Nonviable tissue, n (%)	0 (0%)	0 (0%)	>0.999	4 (26.7%)	3 (20%)	>0.999
Odour, n (%)	0 (0%)	0 (0%)	>0.999	2 (13.3%)	2 (13.3%)	>0.999

NMBM—natural matrix biopolymer membrane; data represent the number and percentage of patients demonstrating each outcome measurement at baseline (0 weeks) and at the final follow-up time (4 weeks). Within-groups significance testing was performed using a two-sided Fisher's exact test

Fig 2. Treatment-related changes in ulcer size. Mean percentage change in ulcer surface area (**a**). Mean percentage change in ulcer volume (**b**). Ulcer sizes for the natural matrix biopolymer membrane (NMBM) and control group were normalised at baseline on a per-patient basis and compared with baseline measurements at designated follow-up times. Wound size was measured in cubic centimetres. Error bars represent the standard error of the mean



four weeks of therapy. The mean decrease in VAS pain score among patients in the control group was 0.13 (95% CI: –1.32–1.58) after four weeks (Fig 5). In a two-way ANOVA concerning mean changes in VAS pain score for the NMBM versus the control group, no significant difference in pain reduction was found (p=0.460).

Discussion

This pilot study investigated the therapeutic effects of NMBM in combination with standard compression therapy for treating hard-to-heal VLUs as compared with conventional compression and dressing therapy. Here, we have shown for the first time that NMBM can be used in the treatment of venous-stasis leg ulcers with

Fig 3. A 79-year-old female patient with a venous leg ulcer (VLU) on her right ankle. VLU before treatment at week 0 (**a**). VLU following two weeks of treatment with natural matrix biopolymer membrane (NMBM); note increased epithelialisation and reduced wound depth (**b**). VLU following four weeks of treatment with NMBM; note ulcer size reduction and epithelial coverage of nearly all of ulcer surface (**c**)

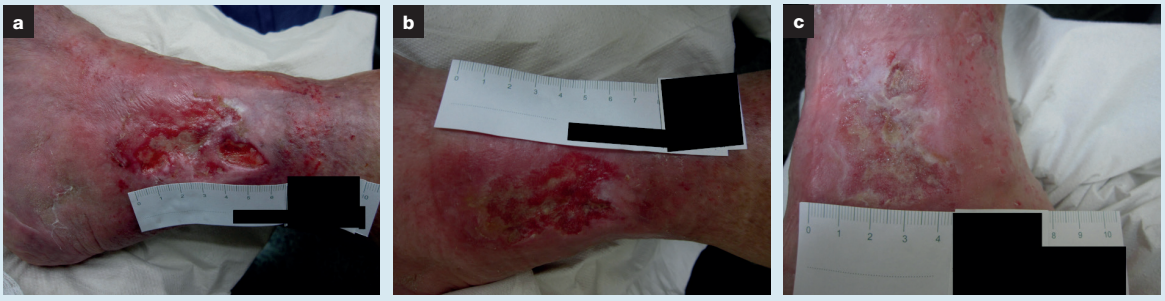


Fig 4. An 85-year-old female patient with a venous ulcer (VLU) on her right leg. VLU before treatment at week 0; note nonviable tissue (**a**). VLU following two weeks of treatment with natural matrix biopolymer membrane (NMBM) (**b**). VLU following four weeks of treatment with NMBM; note complete removal of nonviable tissue, decreased ulcer size and epithelial coverage (**c**)

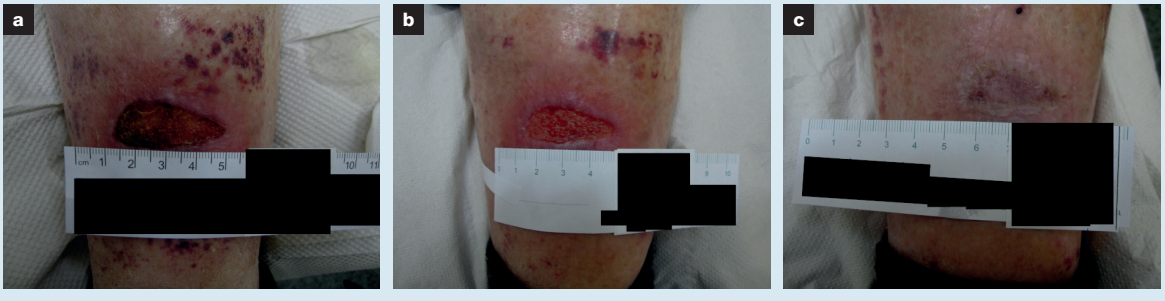
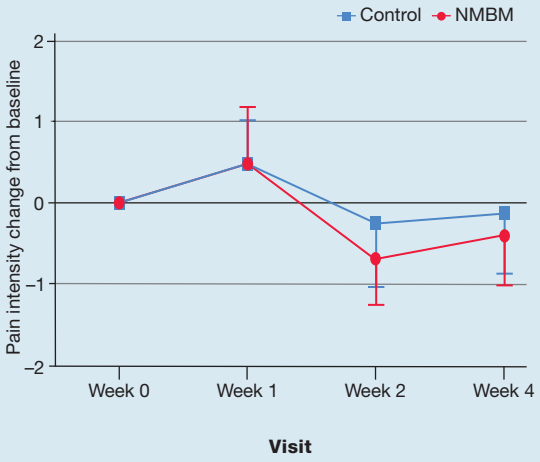


Fig 5. Mean change in pain intensity between treatment arms. Pain intensity was normalised at baseline and compared with baseline measurements at designated follow-up times. Pain intensity was based on a standard 0–10 visual analogue scale. Error bars represent standard error of the mean



efficacy commensurate with conventional modes of wound healing. However, the reductions in ulcer size and pain facilitated by NMBM, even when compared with percentage increases in the control group, were not statistically significant, and it is therefore unclear whether NMBM has a more robust effect on ulcer healing in relation to standard compression and dressing techniques. In addition, no significant differences for any of the secondary ulcer characteristics (exudate consistency and presence of erythema, odour, necrosis or infection) were identified between baseline measurements and four-week follow-up times for either the NMBM or the control group. As this was a small-sample pilot study, this lack of statistical significance may have been due to insufficient statistical power, and larger clinical trials are needed to determine the efficacy of NMBM relative to conventional therapies.

Compression and dressing therapy is regarded as the reference standard in the treatment of VLUs.[22,23] Nevertheless, to the best of our knowledge, this is the first study to demonstrate a class IIb medical device with similar efficacy when coupled with compression treatment. This first pilot study demonstrates that it has promising effects on ulcer healing, which will pave the way toward larger studies using NMBM treatment.

While our pilot study represents the first evaluation of its effectiveness, NMBM was created based on proven mechanisms for mimicking epidermal protection, neutralising bacteria, and maintaining cleanliness and proper moisture. Moreover, NMBM has no contraindications and is hypoallergenic in nature. We previously tested the allergenicity of NMBM through repeat insult patch testing (RIPT) in 50 healthy patients, which demonstrated no cases of contact dermatitis or skin-irritability.[43]

NMBM is also hypothesised to promote autolytic debridement by hydrating the wound surface and lowering the pH of affected skin areas. NMBM maintains a consistent pH of 6.5–7, due to the incorporation of amino acids which act as a buffer and maintain a slightly acidic pH. Lowering the pH of affected wounds has previously been shown to reduce bacterial-induced toxicity by rendering harmful, bacterial byproducts, such as ammonia, inert.[44–47] The ultimate effects of increasing the acidity of wound areas are the destruction of abnormal collagen in ulcer beds, increased angiogenesis, and enhanced activation of macrophages and other immune effectors at sites of ulceration.[37,48,49]

The high sugar content of NMBM also creates a strong osmotic gradient between the skin surface and deep tissue layers, which allows liquid to rise to wound surfaces.[38,50] Accordingly, NMBM's properties as a hydrogel allow it to absorb large amounts of water and maintain a moist environment while maintaining its unique three-dimensional structure. Creation of such osmotic gradients with high-sugar-content agents, such as honey, have previously been shown to reduce antimicrobial growth and rejuvenate nonviable tissue.[51–53] When wounds are kept moist, autolytic debridement is facilitated and nonviable tissue is naturally degraded by endogenous enzymes, such as matrix metalloproteinases.[54] In addition, the viscous topical NMBM cream provides a physical barrier at the wound surface to prevent contamination, and to facilitate granulation and healthy dermis formation.

Ultimately, through a complex and poorly understood mechanism of action, NMBM creates a clean, antimicrobial environment to wounds which accelerates dermal regeneration.

Limitations

Insufficient statistical power was a significant limitation of our study. In addition, though manual methods of ulcer size measurement have been deemed reliable for small and regularly shaped wounds,[55] electronic methods of ulcer measurement, such as digital planimetry, may be more accurate.[55,56] Our use of mild soap to wash the wound site before applying treatment may have confounded the pH-altering properties of NMBM. Because we were focused on determining the efficacy of NMBM on a limited timescale, the long-term effects of the cream on wound healing cannot be reliably inferred beyond four weeks. Sample sizes were also too small to evaluate comparisons of skin irritation between treatments.

Another potential problem was that two patients within the control group presented with local infections and were thus treated with antibiotic dressing; future studies should acknowledge infection as a component of the inclusion and exclusion criteria. Lastly, inclusion of multiple therapies within the control group makes treatment-specific comparisons unreliable in our small sample. Further investigation of NMBM over a longer timespan with a larger sample size should be conducted in order to establish a reliable safety and efficacy profile.

Conclusion

In this study, NMBM was a safe treatment for VLUs, and it reduced both pain and ulcer size. However, as no significant differences in efficacy were shown between NMBM and control treatments, further larger-scale study is needed. JWC

Reflective questions

- What effect(s) does natural matrix biopolymer membrane (NMBM) and combined compression therapy have on the treatment of hard-to-heal venous leg ulcers (VLU)?
- What differences, if any, were there in the effects of treatment between the groups in this study?
- What recommendations can be made regarding use of NMBM cream in future clinical studies?

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