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Effect of a wound cleansing solution on wound bed preparation and inflammation in chronic wounds: a single-blind RCT

• **Objective:** Research into surfactant solutions for the debridement of chronic wounds suggests that surfactants may support wound bed preparation (WBP) in chronic wounds, however their efficacy has not been evaluated in randomised controlled trials (RCTs). Our aim was to assess the clinical efficacy of a propylbetaine-polihexanide (PP) solution versus normal saline (NS) solution in WBP, assessing inflammatory signs and wound size reduction in patients with pressure ulcers (PUs) or vascular leg ulcers.

• **Method:** In a single-blinded randomised controlled trial (RCT) patients were randomly allocated to two groups and treated with either propylbetaine-polihexanide (PP) solution (Prontosan) or NS. Wounds were assessed using the Bates-Jensen wound assessment tool (BWAT). Assessments took place at inclusion (T0), day 7 (T1), day 14 (T2), day 21 (T3), and day 28 (T4). Outcomes were analysed using a two-tailed Student's t-test.

• **Results:** A total of 289 patients were included. Both groups had similar demographics, clinical status, and wound characteristics. Data analysis showed statistically significant differences between T0 and T4 for the following outcomes: BWAT total score, p=0.0248; BWAT score for inflammatory items, p=0.03; BWAT scores for wound size reduction (p=0.049) and granulation tissue improvement (p=0.043), all in favour of PP. The assessment of pain did not show any significant difference between the two groups.

• **Conclusion:** The study results showed significantly higher efficacy of the PP solution versus NS solution, in reducing inflammatory signs and accelerating the healing of vascular leg ulcers and PUs. This evidence supports the update of protocols for the care of chronic wounds.

• **Declaration of interest:** The authors have no conflict of interest regarding this research. This is an investigator initiated trial. B. Braun Milano SpA kindly provided the material under investigation for both treatment groups, and paid the Ethics Committees' application fees in all participating centres.

wound debridement; inflammation; leg ulcers; pressure ulcers; propylbetaine; polihexanide

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Continued overleaf

Hard-to-heal ulcers are frequently due to the presence of debris and tissue that allows the heavy growth of bacteria and the development of biofilm. Cleaning and debridement of the wound bed as well as control of exudate and bacterial load[1] are principles that apply to wound management.[2]

Since 2005[3] international and national guidelines[2,4,5] have incorporated the principles of wound bed preparation (WBP), to promote tissue repair through evidence-based clinical decisions.

Debridement has been traditionally assimilated to WBP. Debridement refers to removing necrotic material, eschar, devitalised tissue or any other type of bioburden from a wound, including wounds with tunnels and/or cavities, to promote wound healing.[1–5] The most important clinical challenge regarding debridement is to select the appropriate debridement method for each individual, maintaining the balance between respecting viable tissue and the speed at which non-viable tissue is removed.[5]

The presence of bacterial biofilm is considered as a barrier to the natural progression of wounds towards healing.[5,6] Biofilms are abundant in chronic wounds as demonstrated by James et al., who reported 60% of the chronic wounds contained biofilm compared with 6% of acute wounds.[7] Biofilm acts as a mechanical barrier reducing antimicrobial contact with bacteria and their effectiveness, and facilitating the bioburden transition from simple colonisation to critical colonisation and infection.[8] Biofilm adhesion to wound bed tissue is stronger than the adhesion of slough, and biofilms are highly resistant to cleansing by irrigation with isotonic solutions.[9,10]

Evidence suggests that debridement is the best method to reduce the biofilm burden in chronic wounds. No form of debridement is likely to remove all biofilm, as its formation is a dynamic process and

any remaining bacteria/biofilm have the potential to reform and grow.[11] Regular debridement decreases time to healing,[12] and makes wound bioburden more susceptible to antibiotics and host defences.[13]

Isotonic solutions such as normal saline (NS) 0.9% NaCl, are used for wound cleansing, but this must be differentiated from debridement.[14] The most frequently used debridement method in clinical practice is autolytic debridement, which is based on the body's capacity to break down necrotic tissues and is carried out using dressings capable of promoting a moist environment.[2–4,15,16]

Some products are suggested to have additional advantages in wound management by supporting the removal of bacteria and debris, and disturbing biofilm, for example a wound cleansing formulation containing the antimicrobial polihexanide and the surfactant component betaine (propylbetaine-polihexanide, Prontosan solution, B.Braun), which reduces surface tension and aids removal of debris and bacteria,[17,18,19] without being cytotoxic.[20]

Experimental trials have investigated the efficacy of surfactants,[17,21,22] and shown that these tensioactive substances destroy the adhesion bridges between biofilm and the wound bed. Kaehn and Eberlin[15] found that PP solution has a higher efficacy than NS to solubilise proteins and other substances. The salt ions present in saline solutions seem to hinder protein hydration and therefore protein solubility, thus ionic solutions are less able to remove proteins from the wound surface.[17] Furthermore, Perez et al.[22] demonstrated that a PP solution was significantly better than NS at reducing the number of meticillin-resistant *Staphylococcus aureus* (MRSA) colony -forming units in a porcine wound model.

Propylbetaine is a surface-active substance that penetrates difficult coatings and helps remove debris and bacteria. The proposed mechanism of action is based on its low surface tension. Polihexanide is a polymerised form of chlorhexidine, which has microbicidal activity and good tissue compatibility. Polihexanide molecules show a high activity against the first layer of the bacterial cytoplasmic bilayer membrane and a minor effect on human cell membranes. The combination of 0.1% polihexanide and 0.1% betaine offers a lower surface tension than the individual substances.[23,24]

A number of authors have investigated the clinical efficacy of PP solution in the management of chronic wounds. Moeller et al. published a retrospective assessment of the management of chronic wounds using PP solution in 953 patients. Wounds progressed towards healing in 97% of patients, and a complete reduction or improvement in wound odour was seen in 65% of patients.[25]

A retrospective study compared two groups of patients with venous leg ulcers (VLUs), reporting that in the PP group wound healing was double that of the NS group ($p<0.0001$).[18] Another study compared the efficacy of a PP solution and NS to control bioburden in the chronic wounds 40 patients. They measured pH variations as a surrogate marker for bioburden, after a 4-week period, the pH values were significantly lower ($p<0.05$) in the PP group, which is correlated with a significant bioburden reduction.[19]

Clinical and experimental studies have suggested that PP solution may be effective in accelerating healing; however, no RCT evaluating the efficacy of PP versus NS in wound bed preparation of chronic wounds have been published. Therefore, we designed an RCT to compare PP to common practice (the use of NS) and to further explore the advantages of using a detergent solution for WBP in chronic wounds.

Objective

To compare the clinical efficacy of a PP solution versus NS solution, assessing inflammatory signs and wound size in patients with pressure ulcers (PUs) or vascular leg ulcers.

Materials and methods

The study was designed as a randomised, controlled, single-blind trial, with patients with chronic wounds, stratified by the type of wound. The study was conducted in six centres in four Italian regions (Lombardy, Piedmont, Tuscany and Apulia), in hospital wards (geriatrics and medicine), and outpatient clinics (phlebology, surgery, and dermatology).

Inclusion criteria

Recruited patients were adults, aged ≥18 years old (inpatients, outpatients or hospitalised at home for at least 24 hours) with the following:

- Presence of at least one PU category II or III as described in the NPUAP/EPUAP classification for PUs[26]
- Braden score of ≥10 for patients with PUs or the presence of a lesion of vascular origin (including the involvement of the subcutaneous tissue, with inflammatory signs and/or biofilm, and/or fibrin on the wound bed)
- Size of the lesion: less than 80cm^2 (able to be covered with a dressing of 10cm x 10cm)

Exclusion criteria

- Terminally ill patients
- Patients treated with systemic or topical antibiotics, and/or antiseptics within 10 days of recruitment
- Patients with a Braden score <10
- Patients under treatment with systemic corticosteroids, immunosuppressants or radiotherapy
- Patients difficult to reposition or impossible to place on a pressure-redistributing mattress

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- Patients with known or suspected sensitivity to any of the components of PP
- Patients with diabetic foot ulcers
- Wounds with necrotic dry eschars
- Patients already included who develop a second chronic wound; the newly developed wound was not considered for a further/extended inclusion.

All patients signed an informed consent form before starting the study, which complied with the Declaration of Helsinki and was approved by the Clinical Research Ethics Committee at each of the participating hospitals. The trial was registered NCT01333670 at the ClinicalTrials.gov database.

Patients were assigned to two groups using a simple randomisation method: a list of numbers generated by proprietary software (available at http://www.randomizer.org). The randomisation list and the sealed envelopes were prepared by an independent third-party research centre. In order to conceal the allocation to treatment, each centre was provided with sealed envelopes to be opened only at recruitment of each individual patient.

The patients received either propylbetaine 0.1% and polihexanide 0.1% (PP) or NS as comparator.

Experimental and control groups interventions

Patients in the experimental group were treated with PP, at inclusion and at every dressing change, wounds were freely irrigated (syringe 20–30 ml, needle 19–20 G), followed by the application of a pack containing PP for at least 10 minutes.

Patients in the control group were treated with NS solution, at inclusion and at every dressing change, wounds were freely irrigated (syringe 20–30 ml, needle 19–20 G), followed by the application of a pack containing NS for at least 10 minutes. After completing the irrigation and packing process, the wounds were covered with dressings following local protocols. Wounds were assessed and data recorded at every dressing change before treatment.

Staff providing wound care were different from the those carrying out wound assessments. Those performing wound assessment were blind to the solution being used for wound irrigation and pack soaking. To minimise the risk of bias, investigators assessing wounds, at each participating centre, were wound care experts belonging to the Italian Nurses' Cutaneous Wounds Association (AISLeC) who received refresher training on how to correctly use the BWAT.

All patients with VLUs, either in the PP group or in the NS group, had the appropriate compressive therapy. All patients with PUs were placed on pressure-redistributing devices depending on individual risk.

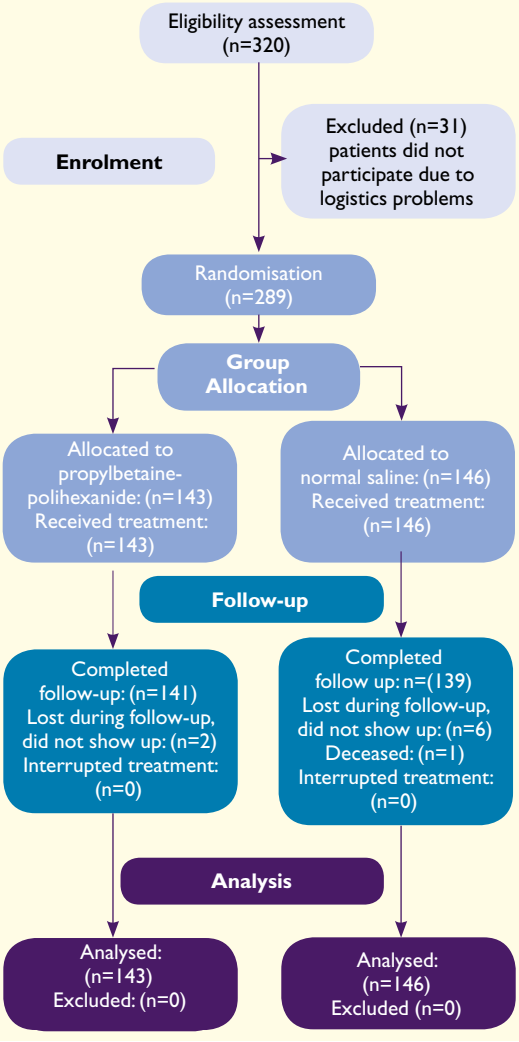
In the event of periwound skin maceration or irritation, a barrier product such as zinc oxide paste or other similar skin-barrier product was applied.

Primary outcome measurement

The efficacy assessment for the primary outcome was made through the recording and analysing the data obtained from clinical signs commonly found in inflamed or infected wounds, as referenced in the scientific literature. The assessment items were those of the Bates-Jensen Wound Assessment Tool (BWAT). This scale was validated in 2010 for a wide range of wounds.[27]

Assessment was performed in all patients at recruitment (T0), at day 7 (T1), day 14 (T2), day 21 (T3) and day 28 (T4). The BWAT contains 13 items that assess wound size, wound depth, wound edges, wound undermining, necrotic tissue type, necrotic tissue amount, granulation tissue, epithelialisation, exudate type, exudate amount, surrounding skin colour, peripheral tissue oedema, and peripheral tissue induration. These items use a modified Likert scale: a score of 1 indicates the healthiest and 5 indi-

Fig 1. Consort flow chart



cates the most unhealthy attribute for each characteristic.[27] The total BWAT score was obtained by adding the individual scores of each assessment item, thus, the total value ranged from a minimum of 13 to a potential maximum of 65.

Assessment of wound inflammation was performed through the analysis of a score obtained from five BWAT items specifically linked to inflammation: exudate type, exudate amount, surrounding skin colour, peripheral tissue oedema, and peripheral tissue induration.

The measurement of the wound size was carried out using sterile rulers and gridded transparent acetate sheets; pictures of the wounds were also taken at each weekly assessment and used to determine changes in the wound, mainly those related to wound size.

Secondary outcome measurement

Pain was assessed with a visual analog scale (VAS: values from 0=no pain to 10=worst possible pain). For all the patients in both study groups, pain assessments were performed at days 0, 7, 14, 21 and 28.

If present, adverse events were recorded. The safety of the study products was assessed through the incidence of adverse events related to the products under evaluation.

Statistical methods

The population size (165 patients per group) was calculated to demonstrate a power of 90% and a significance level of 5% (α=0.05, β=0.10). Significance was calculated from the average differences between the two groups using a two-tailed Student's t-test. No interim analysis was scheduled during the trial.

Results

We recruited 289 patients between June 2010 and December 2013. As evidenced in the consort flow chart (Fig. 1), 31 out of 320 patients screened were not included due to logistic issues related to the ability to attend the follow-up visits. Furthermore, two randomised patients in the PP group and six in the NS group did not attend follow-up visits. There was one death in the NS group related to the patient's comorbidities not the treatment. The follow-up was completed in 141 patients in the PP group and in 139 patients in the NS group; however, their recorded outcomes were included in the overall results assessment in order to respect the principle of intention to treat (ITT).

Patient characteristics

Of the 289 recruited patients, 143 were in the PP group and 146 in the NS group. The populations of both groups had similar characteristics regarding gender, age, Braden score (in patients with PUs), BMI and comorbidities (Table 1).

The majority (67%) of the total number of recruited patients presented vascular leg ulcers (venous and mixed origin), 25% of patients presented PUs (Table 2). The distribution of wounds by type was similar in both groups. The comparative analysis did not find any significant difference.

The statistical analysis of the initial BWAT scores

Table 1. Patients' clinical characteristics

Recruited patients	PP Group n=143		NS Group n=146	
Male	58 (40.6%)		65 (44.5%)	
Female	85 (59.4%)		81 (55.5%)	
	Years	sd	Years	sd
Age (average)	79.8	12.1	77.2	15.3
	Score	sd	Score	sd
Braden Score‡ (average)	18	3.0	20	2.9
Body mass index (BMI)				
BMI normal weight	27 (18.9%)		26 (17.8%)	
BMI obesity	37 (25.9%)		60 (41.1%)	
BMI severe obesity	15 (10.5%)		17 (11.6%)	
BMI overweight	58 (40.6%)		42 (28.8%)	
BMI underweight	6 (4.2%)		1 (0.7%)	
Comorbidities	Number of patients (%)		Number of patients (%)	
Diabetes	13 (9.1%)		20 (13.7%)	
Infectious pathology	9 (6.3%)		4 (2.7%)	
Thrombosis/phlebitis	32 (22.4%)		23 (15.8%)	
Pneumonia	4 (2.8%)		3 (2.1%)	

Standard deviation–sd; PP–propylbetaine-polihexanide ; NS–normal saline; ‡Braden Score evaluated only in patients with pressure ulcers

Table 2. Wounds types by group

Recruited patients	PP group n=143 (%)	NS group n=146 (%)	p-value
Pressure ulcers	37 (25.9%)	35 (24.0%)	0.75
Venous ulcers	74 (51.7%)	66 (45.2%)	0.40
Mixed aetiology ulcers (venous/arterial)	27 (18.9%)	27 (18.5%)	0.80
Traumatic wounds in patients with venous ulcers	5 (3.5%)	18 (12.3%)	0.32

PP–propylbetaine-polihexanide; NS–normal saline

Fig 2. Wound improvement assessed by a reduction of average Total BWAT Scores by group and by visit

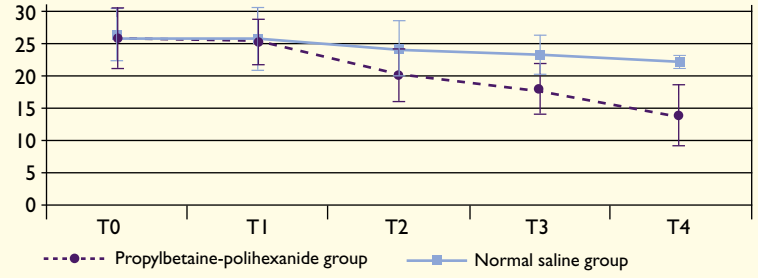
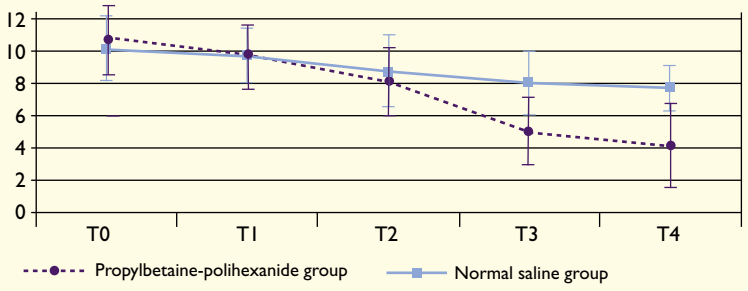


Fig 3. Reduction of inflammatory signs assessed by comparing the BWAT average scores by group and by visit.



did not find any significant difference between the two groups.

Primary outcome results:

The primary outcome (wound improvement), was assessed through the variation of BWAT scores (Table 3). Results from T0 versus the data recorded at T1, T2, T3 and T4 were analysed. This analysis showed a significant difference between T0 and T4 in the following items:

- Total BWAT score of overall wound evolution: p=0.0248 indicating significantly better progression of wounds in the experimental group (Fig 2). The reduction in the average total BWAT score was significantly better at T4 compared with T0 in the PP group.
- BWAT inflammatory score, to assess the change of inflammatory signs: p=0.03 indicates a significantly better progression of inflammatory signs in wounds of patients in the PP group (Fig 3). There was a reduction in the average BWAT scores for inflammatory signs that was significantly better at T4 compared with T0 in the PP group.

Secondary outcome results

Pain scores were similar in both study groups, average score: 3.0, with minimal or no change during follow-up. The study did not find any significant difference in pain associated to the study wounds or to dressing changes or in the pain suffered during the interval of time between dressing changes.

Adverse events

There were no adverse events related to treatment during the study period.

Discussion

Since 2009, Wolcott et al. have been promoting an aggressive approach to treat biofilm,[28] but this does not take in consideration that a large proportion of wound care is provided in the community. Intense mechanical debridement has to be correctly indicated and performed by trained clinicians, and because it may be painful and cause bleeding, it has to be performed in hospital settings. Thus, it is difficult to perform intense debridement at patients' home, where clinicians should provide the best quality care without compromising patients' safety.

Based on published evidence[9,10,12,17,20,22–25] we identified PP solution as a good candidate to replace NS in order to accelerate wound autolytic debridement.

Our study has shown that the use of PP when applied and kept in place for 10 minutes with packing, promotes a quicker reduction of wound size, and inflammatory signs than NS. The difference between PP and the NS was statistically significant, with better results, in the experimental group regarding the reduction of the total BWAT score, inflammatory signs, the reduction of wound size and the improvement of granulation tissue.

The results of this trial confirm that PP looks to be a good option for the management of vascular leg ulcers and category II and III PUs. This is in line with the accepted principle that a better debrided wound has a higher probability of moving towards healing faster.

These results are relevant for the clinical practice because they were obtained from a population reflecting accurately the characteristics of patients commonly treated in wound care centres.

Our results are in line with those published by Durante et al. in 2012 about the efficacy of PP for the treatment of chronic wounds in 124 patients, where they reported a significant reduction in the wound size ($p<0.001$).[29]

The results also showed a trend towards a quicker resolution of inflammatory signs in vascular ulcers when compared with PUs. This may be partly explained by the challenge represented by the anatomic location of some PUs (for example, the sacrum), as in some cases it was difficult to keep in place the pack containing the solutions during the intervention at dressing change. Implementing the best debridement strategy for each individual patient and situation, and having available the appropriate products, are key success factors in wound bed preparation.

The adoption of interventions speeding up the autolytic debridement should be seriously consid-

Table 3. Wound characteristics as evaluated by the BWAT at the initial assessment (T0), are presented by group

BWAT Score	Table 3 BWAT items description	PP (n=143)		NS (n=146)		p Value
		n	%	n	%	
	Size (length x wide)					
1	<4cm^2	0	0	0	0	-
2	4–16cm^2	28	19.6	32	21.9	0.66
3	16.1–36cm^2	14	9.8	9	6.2	0.28
4	36.1–80cm^2	101	70.6	105	71.9	0.89
5	>80cm^2	0	0	0	0	-
	Depth					
2	Partial-thickness skin loss involving epidermis and/or dermis	121	84.6	121	82.9	0.75
3	Full-thickness skin loss involving damage or necrosis of subcutaneous tissue	22	15.4	25	17.1	0.75
	Edges					
1	Indistinct, diffuse, none clearly visible	10	7.0	15	10.3	0.4
2	Distinct, outline clearly visible, attached, even with wound base	52	36.4	41	28.1	0.2
3	Well-defined, not attached to wound base	81	56.6	90	61.6	0.4
	Type of exudate					
3	Serosanguineous: thin, watery, pale red/pink	18	12.6	25	17.1	0.32
4	Serous: thin, watery, clear	120	83.9	119	81.5	0.61
5	Purulent: thin or thick, opaque, tan/yellow, with or without odour	5	3.5	2	1.4	0.27
	Amount of exudate					
3	Scant	18	12.6	26	17.8	0.32
4	Moderate	25	17.5	26	17.8	0.87
5	Large	100	69.9	94	64.4	0.32
	Skin colour surrounding wound					
1	Pink or normal for ethnic group	25	17.5	26	17.8	0.87
2	Bright red and/or blanches to touch	86	60.1	90	61.6	0.37
4	Dark red or purple and/or not blanchable	32	22.4	30	20.5	0.77
	Peripheral tissue oedema and induration					
2	Induration <2cm around wound	66	46.2	81	55.5	0.12
3	Non-pitting oedema extends ≥4cm around wound	77	53.8	65	44.5	0.12
	Granulation Tissue					
3	Bright, beefy red, 25–75% of wound filled	27	18.9	32	21.9	0.56
4	Pink and/or dull, dusky red &/or fills ≤25% of wound	116	81.1	114	78.1	0.56
	Average total initial score BWAT §	**25.9**		**25.45**		**0.75**

Items not observed during the assessment were omitted from this table; PP–propylbetaine-polihexanide ; NS–normal saline; BWAT–Bates-Jensen Wound Assessment Tool

ered. As more evidence becomes available, protocols of care for chronic wounds should be updated accordingly. Our results strongly support the use of surfactant solutions such as PP solution, instead of isotonic solutions in the care of chronic wounds.

Not having any adverse events related to the study device during this trial confirms the safety profile of the solution.

Limitations of the study

Our study explored the efficacy through assessment of items signalling the physical evolution of wounds, however, the observation period was too short to establish an actual rate of healing. It would be useful to confirm these results in an RCT with a longer observation period, and compared with different debridement methods.

Given the characteristics of the solutions under investigation, it was impossible to perform a double-blind trial. To minimise the bias a single-blind study was implemented, where the investigators assessing wounds did not know which product was being used.

Conclusions

The analysis of data collected supports the superiority of PP solution versus NS solution in terms of efficacy, and suggest that when used with best current clinical practice, it promotes WBP, reduces inflammatory signs and accelerates healing, in venous or mixed vascular leg ulcers and PUs. This study did not find any significant difference between the two groups of patients regarding pain reduction. ■

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