Suprathel-Antiseptic Matrix: In Vitro Model for Local Antiseptic Treatment?

Henning Ryssel, MD; Christian Andreas Radu, MD; Guenter Germann, MD, PhD; Oliver Kloeters, MD; Katrin Riedel, MD; Maximilian Otte, MD; and Thomas Kremer, MD

ABSTRACT

Acetic acid is a traditional antiseptic agent that has been used for more than 6000 years. The main goal of this study was to demonstrate the suitability of Suprathel (PolyMedics Innovations GmbH, Denkendorf, Germany) in combination with various antiseptic agents to create an “antiseptic-matrix” especially designed for problematic microorganisms such as Proteus vulgaris, Acinetobacter baumannii, or Pseudomonas aeruginosa, which are frequently associated with burns. The study was designed to test the in vitro antimicrobial effect of a “Suprathel-antiseptic matrix” (Suprathel combined with acetic acid 3%, povidone-iodine 11% [Betadine], polyhexanide 0.04% [Ivalosept], phenoxethanol 2%/octenidine dihydrochloride 0.1% [Octenisept], mafenide acetate 5%, and chlorhexidine gluconate 1.5%/cetrimid 15% [Hibicet]). As a means to assess the typical bacterial spectrum of a burn unit, the following Gram-negative and Gram-positive bacteria strains were tested: Escherichia coli, P vulgaris, P aeruginosa, A baumannii, Enterococcus faecalis, Staphylococcus epidermidis, Staphylococcus aureus, methicillin-resistant S aureus, and β-hemolytic streptococcus groups A and B. The tests showed a positive bactericidal effect of the Suprathel-antiseptic matrix, particularly with problematic Gram-negative bacteria such as P vulgaris, P aeruginosa, and A baumannii, except for the combination of Suprathel and mafenide acetate. It can be concluded that Suprathel-antiseptic matrix appears to be suitable as a local antiseptic agent, but clinical studies need to be performed to confirm these in vitro observations. The authors’ previous studies have shown that acetic acid demonstrates a wide antiseptic spectrum for microorganisms typically found in burn patients. The combination of Suprathel and acetic acid worked well in this study and appears to be promising for future clinical application.

KEYWORDS: Suprathel antiseptic for burns, Suprathel-antiseptic matrix, antiseptic treatments for burns, burn wound infection

INTRODUCTION

Local infection and burn wound sepsis are still among the most severe problems in the treatment of thermal-injured patients. Morbidity and mortality of burn patients are highly correlated to the incidence of wound infection and its sequelae.1 Subsequent colonization and infection not only cause burn wound sepsis but may also induce immune responses that lead to cytokine release, the entire cascade of acute-phase reaction, and subsequent multiorgan failure.2–4

At present, many different agents may be applied to burn wounds, such as silver nitrate, mafenide acetate, or povidone-iodine in an attempt to reduce bacterial colonization. However, a controversial discussion concerning the selection of the “ideal” antiseptic treatment is still ongoing as topical treatment options are on the rise. Suprathel (PolyMedics Innovations GmbH, Denkendorf, Germany) is a polymer sheet based on polyactic acid technology.5 Initially, it was used to treat superficial and partial-thickness burns to expedite reepithelialization and reduce pain.6 Early clinical experience with the Suprathel product in these applications has been very successful. Given the early success with covering and healing burns, the next topic to focus on was the management of burn-associated infections. This led the authors to evaluate the feasibility and suitability of Suprathel as a carrier substance for different antiseptic agents.6

In a prior study, the authors evaluated the feasibility and suitability of Suprathel as a carrier for different antiseptic agents.6 The first impressions of the polymer matrix were positive in this study.6 With acetic acid as an antiseptic agent, the polymer matrix was easy to handle. The authors put the Suprathel sheet in the acid, and it soaked well with this agent. After 10 minutes, there was no evidence of macroscopic damage of the sheet, nor was it gluey or adhesive. In scanning electron microscopy (SEM), no changes of the structure were visible. With polyhexanide 0.04% (Lavasept; Infectless AG, Basel, Switzerland) as antiseptic agent, the sheets were swimming on the surface of the acetic acid but...
seemed to soak the fluid well. After 10 minutes of uptake, no macroscopic damage of the sheet was observed, nor was it deformed or altered in any way; however, there were some drops of Lavasept on the sheet. In SEM, no changes of the ultrastructure were visible. With mafenide acetate, the sheets were swimming on the surface of the antiseptic but seemed to soak up the fluid well. After 10 minutes, there was no macroscopic alteration of the sheet, nor was it gluey or adhesive, and there were no drops on top as with Lavasept. In SEM, no changes of the structure were visible.

With povidone-iodine 11% (Betaisodona; Mundipharma, Limburg, Germany), there was an easy handling as well. After putting the Suprathel sheet in the agent, it soaked well with Betaisodona without swimming on top. After 10 minutes, there was no macroscopic damage of the sheet, and it was not sticky. In SEM, no changes of the structure were visible.

When the authors placed the Suprathel sheet in octenidine hydrochloride (Octenisept; Schülke & Mayr, Norderstedt, Germany), it soaked well with this agent, but handling of the dressing was difficult. Although there was no macroscopic damage of the sheet after 10 minutes, it was very gluey, sticky, and adhesive. In SEM, the ultrastructure was not destructed.

As the authors tested the uptake and release rates in the pilot study, the following was found: the mean uptake after 10 tests after 10 minutes was 183.53 mg (128.4% of sheet weight) for acetic acid, 215.8 mg (144.6% of sheet weight) for Lavasept, 140.9 mg (97.5% of sheet weight) for mafenide acetate, 350.0 mg (255.1% of sheet weight) for Betaisodona, and 517.97 mg (342.1% of sheet weight) for Octenisept. Focusing on the absolute and relative release rates, whereas the relative release rates are calculated as the percentages related to the initial uptake, the following data were found in the authors’ pilot study:

- 153.8 mg absolute release (83.8% of initial uptake) for acetic acid, 168.25 mg absolute release (77% of initial uptake) for Lavasept, 116.6 mg absolute release (82.7% of initial uptake) for mafenide acetate, 299.5 mg absolute release (85.25% of initial uptake) for Betaisodona, and 422.5 mg absolute release (81.6% of initial uptake) for Octenisept.

In the pilot study, the authors also tested the release of the antiseptics to cadaveric skin from the antiseptic-soaked matrix to imitate a clinical situation and application. Here, the uptake to the cadaveric skin ranged from 30% to 54% of the initial antiseptic amount uptaken by Suprathel, with release of 79% to 93% of the particular antiseptic from the matrix. In summary, the results were positive, especially for acetic acid, which showed favorable uptake and release characteristics.

Therefore, this study was performed to evaluate the in vitro bactericidal power of a Suprathel-antiseptic matrix based on different antiseptics against common bacterial strains found in the authors’ burn patient population. The objective was to create an antiseptic matrix with the regenerative characteristics of Suprathel, which were shown in prior studies, and the antiseptic power of acetic acid, which was also shown in a prior study. Thus, it is important to investigate which combinations of Suprathel and antiseptics are effective.

**MATERIAL AND METHODS**

Following the protocol of the previous study, a Suprathel sheet measuring 5 x 5 cm was soaked in each of the tested antiseptic agents for 10 minutes. The tested bactericidal solutions acetic acid 3%, povidone-iodine 11% (Betaisodona), polyhexanide 0.04% (Lavasept), phenoxyethanol 2%/octenidine hydrochloride 0.1% (Octenisept), mafenide acetate 5%, and chlorhexidine gluconate 1.5%/cetrimid 15% (Hibiet; Mölnlycke Health Care, Norcross, Georgia) are routinely used in the authors’ department; an H2O group functioned as a control.

For the creation of the bacterial test colonies, standard 1-boullion tubes were inoculated with 5 x 106 colony-forming units (CFUs) of the below-mentioned bacteria and incubated at 37°C for 24 hours, per the standard method for the incubation of bacteria. Immediately following innovation, 0.1 mL of each bacterial culture was placed onto standard agar plates. The antiseptic-soaked Suprathel sheets were placed on top of the agar plates and allowed to incubate for 20 minutes. The antiseptic sheet was removed, and the plates were incubated at 37°C for 48 hours and then evaluated for the total number of CFUs. The process of contacting a seeded lawn of organisms with an antiseptic-soaked Suprathel sheet was repeated for each antiseptic. The standards for performing bacterial cultures were based on the recommendations of the Robert Koch Institute and the German Institute of Hygiene. The authors took the Suprathel-antiseptic matrix off after 20 minutes to have a fixed time of application. From the clinical view, the authors know that local antiseptic treatment of 20 minutes is quite effective.

The following Gram-negative and Gram-positive bacteria strains were tested: Escherichia coli (DSM Nr. 498, K12), Proteus vulgaris (Hauer 1885, group 2), Pseudomonas aeruginosa (Schröeter 1872, group 2), Acinetobacter baumannii (Institute Limbach, Heidelberg, Germany), Enterococcus faecalis (Andrews and Horder 1906, group 2), Staphylococcus epidermidis (Winslow 1908, group 2), methicillin-resistant Staphylococcus aureus (MRSA, Institute Limbach), S aureus (Rosenbach 1984, group 2), and β-hemolytic streptococcus groups A and B (Institute Limbach). All bacteria were isolated from patients at the authors’ burn unit. Each combination of each antiseptic agent was tested with each organism 4 times; and the mean CFUs were calculated for each possible combination.

**RESULTS**

Table 1 summarizes the results of the different antiseptic agents put on a Suprathel sheet against various microorganisms; the
data presented represent 4 replicate experiments and also correspond with previous data from the authors’ pilot study. After 20 minutes of contact with the Suprathel sheet–antiseptic matrix, there were no detectable CFUs of *P. vulgaris*, *P. aeruginosa*, *A. baumannii*, *β*-hemolytic streptococci group B, *Staphylococcus epidermidis*, *Staphylococcus aureus*, and *β*-hemolytic streptococci group A, *E. coli*, *E. faecalis*, methicillin-resistant *S. aureus* by 5 × 5 cm Suprathel sheet soaked with acetic acid 3%, povidone-iodine 11% (Betaisodona), polyhexanide 0.04% (Lavasept), phenoxethanol 2%/octenidine hydrochloride 0.1% (Octenisept), mafenide acetate 5%, and chlorhexidine gluconate 1.5%/ cetrimide 15% (Hibicet) and H2O (control group). Standard 1-boullion tubes were inoculated with these bacteria and incubated at 37°C for 24 hours. From the obtained bacterial cultures, 0.1 mL was spread on standard agar plates after and contacted with an antiseptic-soaked Suprathel sheet for 20 minutes and subsequently incubated at 37°C for 48 hours and evaluated for the total number of CFUs. The same process was repeated for each of the antiseptics. Each combination of antiseptic agent and each microorganism was tested 4 times, and the mean CFUs were calculated for each possible combination.

### Table 1.

<table>
<thead>
<tr>
<th>Germ</th>
<th>Antiseptic Agent</th>
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<tr>
<td></td>
<td>Acetic acid 3%</td>
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<tr>
<td><em>Proteus vulgaris</em></td>
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<tr>
<td><em>Pseudomonas aeruginosa</em></td>
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<td><em>Acinetobacter baumannii</em></td>
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<td><em>β</em>-hemolytic streptococci group B</td>
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<td><em>Staphylococcus epidermidis</em></td>
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<td><em>Staphylococcus aureus</em></td>
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<td><em>β</em>-hemolytic streptococci group A</td>
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<tr>
<td><em>Streptococcus</em> group A</td>
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<tr>
<td><em>Escherichia coli</em></td>
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<td><em>Enterococcus faecalis</em></td>
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<td><em>Methicillin-resistant Staphylococcus aureus</em></td>
<td>&gt;10³</td>
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Abbreviation: CFUs = colony-forming units.

Elimination of *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Acinetobacter baumanii*, *β*-hemolytic streptococci group B, *Staphylococcus epidermidis*, *Staphylococcus aureus*, and *β*-hemolytic streptococci group A, *E. coli*, *E. faecalis*, methicillin-resistant *Staphylococcus aureus* by 5 × 5 cm Suprathel sheet soaked with acetic acid 3%, povidone-iodine 11% (Betaisodona), polyhexanide 0.04% (Lavasept), phenoxethanol 2%/octenidine hydrochloride 0.1% (Octenisept), mafenide acetate 5%, and chlorhexidine gluconate 1.5%/cetrimide 15% (Hibicet) and H2O (control group). Standard 1-boullion tubes were inoculated with these bacteria and incubated at 37°C for 24 hours. From the obtained bacterial cultures, 0.1 mL was spread on standard agar plates after and contacted with an antiseptic-soaked Suprathel sheet for 20 minutes and subsequently incubated at 37°C for 48 hours and evaluated for the total number of CFUs. The same process was repeated for each of the antiseptics. Each combination of antiseptic agent and each microorganism was tested 4 times, and the mean CFUs were calculated for each possible combination.

different degrees of resistance with erased mean inhibition concentrations of mafenide acetate for the organisms that were isolates from the authors’ burn patients were also obvious and were consistent with the results from the pilot study. The different degrees of resistance against mafenide acetate indicate that selection of our “clinic-specific germs” has occurred over time. This is possible because mafenide acetate has a sulfonamide group, like antibiotics of the sulfonamide type, which is a well-known target of resistance development.

### Figure 1.

**ZONE OF INHIBITION**

Zone of inhibition around a Suprathel–acetic acid disk on the left and missing inhibition by a Suprathel–mafenide acetate disk on the right. Both tested against methicillin-resistant *Staphylococcus aureus*. 
DISCUSSION

The topical treatment of burns is critical for the overall outcome and survival of burn-injured patients. At present, the occurrence of multiresistant organisms is on the rise; thus, alternatives to current surface treatments need to be investigated. The authors have already conducted studies for the in vitro antiseptic effects of acetic acid alone,\textsuperscript{7} which proved to be very efficient, and the feasibility and suitability of Suprathel as an antiseptic agent carrier,\textsuperscript{6} which also worked effectively. Therefore, this study was a logical progression for the creation of a Suprathel–antiseptic matrix. After 20 minutes of application of the Suprathel sheet–antiseptic matrix, the authors did not detect any CFUs of \textit{P. vulgaris}, \textit{P. aeruginosa}, \textit{A. baumannii}, \textit{\beta}-hemolytic streptococcus group B, \textit{S. epidermidis}, \textit{S. aureus}, and \textit{\beta}-hemolytic streptococcus group A, \textit{E. coli}, \textit{E. faecalis}, and MRSA for all antiseptics put on the Suprathel sheet, except for mafenide acetate and the control group, which both yielded more than $10^3$ CFUs. However, as discovered in the authors’ previous study,\textsuperscript{6} handling of Octenisept was difficult compared with the other antiseptic agents. Although the Suprathel sheet absorbed well with this agent, the combination became very sticky and difficult to handle. However, there was no macroscopic damage of the sheet.\textsuperscript{7} The authors have observed in this study that the Suprathel–antiseptic agent matrix worked effectively overall, with beneficial antiseptic effects except for mafenide acetate. This was due to the resistance of this organism that was isolated from a burn patient from the authors’ burn unit.\textsuperscript{7}

The authors conclude that in an in vitro setting, the Suprathel–antiseptic agent matrix is a powerful alternative antiseptic system. The next phase will be to perform clinical studies to confirm the authors’ in vitro results in a relevant in vivo model and to compare the Suprathel–antiseptic matrix with widely used silver ions that contain surface antiseptics, such as Acticoat and Aquacel. In the initial individual clinical applications, the use of Suprathel, with respect to its regenerative characteristics, has not been altered by soaking the matrix with acetic acid.

REFERENCES

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