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ORIGINAL ARTICLE

Surfactants: Role in biofilm management and cellular behaviour

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Appropriate and effective wound cleaning represents an important process that is necessary for preparing the wound for improved wound healing and for helping to dislodge biofilms. Wound cleaning is of paramount importance to wound bed preparation for helping to enhance wound healing. Surfactant applications in wound care may represent an important area in the cleaning continuum. However, understanding of the role and significance of surfactants in wound cleansing, biofilm prevention and control, and enhancing cellular viability and proliferation is currently lacking. Despite this, some recent evidence on poloxamer-based surfactants where the surfactants are present in high concentration have been shown to have an important role to play in biofilm management; matrix metalloproteinase modulation; reducing inflammation; and enhancing cellular proliferation, behaviour, and viability. Consequently, this review aims to discuss the role, mode of action, and clinical significance of the use of medically accepted surfactants, with a focus on concentrated poloxamer-based surfactants, to wound healing but, more specifically, the role they may play in biofilm management and effects on cellular repair.

KEYWORDS
biofilms, cell salvage, chronic wounds, surfactants

1 | INTRODUCTION

Wound cleansing is now being considered essential and significant for effective and timely acute and chronic wound healing and biofilm management.[1] Wound cleaning as defined for this paper is "the ability to manage the elimination of exudate, slough, necrotic debris, and associated microbial contaminants, toxins, matrix metalloproteinases (MMPs) and cytokines as well as dressing residue, without adversely impacting cellular activity vital to the wound healing process or colonising the underlying tissue with microorganisms and detached biofilm." Effective cleaning of a wound represents an opportunity to advance wound care due to the possibility that the procedures used will prevent and control biofilm. Whilst biofilms have been identified in non-healing chronic wounds, it is only relatively recently that, because of their recalcitrance to both the immune system and antimicrobials, their presence is being associated with prolonging wound healing, increasing a wound's propensity to infection and delaying wound closure.[2] In acute wounds, the development of biofilms leads to chronic inflammation. This inflammation is because of elevated levels of pro-inflammatory cytokines

leading to an increase in neutrophils, macrophages, and mast cells, which in turn leads to an increase in proteases and reactive oxygen species (ROS).[3] Overproduction of proteases and ROS cause the breaking down of proteins, which are vital to healing.[3]

For this paper, biofilms are referred to as microorganisms that are attached to each other (aggregated or co-aggregated) or to a surface and are encased within an extracellular matrix, referred to as extracellular polymeric substance (EPS).[4] EPS represents the major component of the biofilm (often comprising over 90% of its total volume) and is composed of polysaccharides, proteins, metal ions (particularly magnesium, calcium, and iron), lipids, and extracellular DNA.[5,6] The EPS matrix of the biofilm is often referred to as the "house of the biofilm cells."[7]

Similar to the microorganisms, for effective and enhanced wound healing, the EPS of the biofilm also needs to be reduced and removed from the wound surface and wound bed to help facilitate faster wound healing. However, present strategies and studies on the effectiveness of wound dressings on biofilms have focused only on the ability of these technologies to reduce and kill microorganisms within the biofilm. This constitutes a major concern in a biofilm-based management approach to wound management, particularly as the extracellular components of the biofilm are very inflammatory and help to enhance rapid biofilm regrowth. This was highlighted in a paper published by *Nature* in 2000.[8] The study reported that the toll-like receptor nine recognises the methylated CpG DNA sequences that are common in bacterial DNA but are not present in human (mammalian) DNA. This highlighted that the human innate immune system is "primed" to recognise unique sequences in bacterial DNA that are not present in human DNA. This is a major factor that explains why the exopolymeric matrix of bacterial biofilms is so inflammatory in patients. Consequently, even if the microbes within the biofilms are dead, but the EPS is still present, this can significantly delay wound healing and increase inflammation and therefore increase infection risk.[8]

Wound cleaning involves the use of debridement and desloughing techniques and the use of antimicrobial-based wound irrigating fluids, gels, and wound dressings as they have all shown to help remove slough, devitalised tissue, particulate matter, and planktonic microorganisms.[1,9] Many wound dressings have been shown to help enhance wound cleaning and prepare the wound bed/environment. Often, many of these wound dressings may contain surfactants, particularly in a concentrated format.[1,4] Unfortunately, the role that these surfactants play in wound healing are often not investigated as they are generally considered to be of limited value for helping with wound cleaning, biofilm management, reducing inflammation, and enhancing cellular proliferation and regeneration. However, the use of poloxamer-based surfactants in wound healing is now being shown to have a significant role to play in biofilm management, MMP modulation, and cellular resuscitation/salvage.[10–12]

Key Messages

- wound cleansing is now being considered essential and significant for the effective and timely acute and chronic wound healing and biofilm management
- poloxamers are capable of reducing and preventing cell death by temporarily "jumping in" to replace damaged cell membranes
- poloxamer-based technologies appear to be demonstrating an ability to enhance wound healing

The focus of this paper is on surfactant-based wound dressings, particularly those containing poloxamer. Other agents that warrant discussion will also be briefly reviewed. The role surfactants play in wound and biofilm management, however, has not been discussed and reviewed in great detail despite some recent publications highlighting their growing significance as part of a anti-biofilm strategy.[4,10–13] Consequently, the aim of this review is to discuss wound cleaning and the role, mode of action, and clinical evidence of the use of surfactants in wound healing but, more specifically, the role poloxamer-based surfactants may play in biofilm management and cellular healing.

2 | WOUND CLEANING: WHAT IS IT AND WHY IS IT NECESSARY?

Liquid-based wound cleansers and irrigating solutions are often being used for the purpose of wound cleaning but are routinely used in isolation and for very short contact times.[14] Unfortunately, the administering of cleansers, such as saline, are known to be ineffective for the effective removal of debris and biofilm. Consequently, cleansers with antiseptics, such as polyhexamethylene biguanide (PHMB), chlorhexidine, etc, are now being used to help clean a wound when biofilms are suspected, which is always the case in chronic wounds. However, as with any antimicrobial, performance in a wound environment requires that the antimicrobial be in contact with the microbes and the biofilm for an appropriate period of time to achieve the desired microbicidal effect.[14,15] Presently, antimicrobial-based wound-cleansing agents are not being used with a long-enough contact time to achieve good antimicrobial efficacy.[14] Many factors can affect the efficacy of antimicrobials on microbes and biofilms, including pH, temperature, microbial bioburden, biofilm, slough/-devitalised tissue, and anything that may represent a biological demand that will have an impact on efficacy claims and cytotoxicity because of the reduction of the bioavailability of the active agents.[16]

A well-planned and administered wound-cleaning programme can help remove barriers that are known to

negatively effect wound healing. These barriers to wound healing include, as examples, slough, devitalised tissue, proteases, and biofilm (microorgansims and the extracellular biofilm matrix).[10] As discussed previously, biofilms are considered to be the root cause of the up-regulation of many of the underlying biological processes known to delay wound healing. It is therefore imperative that biofilms are removed from the wound surface, wound bed, wound dressing, and any supportive surface, and understanding them is critical for effective wound cleaning.

The importance of the removal of barriers to wound healing suggests that surfactants, whilst not presently used widely in wound management, represent a recently emergent technology that can have a major and significant impact on wound-healing rates.[11] This has been demonstrated clinically with several clinical studies being reported.[11] Bellingeri et al[17] evaluated 289 patients, comparing propylbetaine-polyhexanide solution (Prontosan, B Braun, Germany) with saline. It was found that the surfactant-based solution resulted in 97% faster healing rates.

The use of concentrated surfactant-based wound dressings has been shown to demonstrate an ability to loosen, soften, and also sequester debris and necrotic tissue, and it is hypothesised that doing so promotes wound healing.[18] Palumbo and colleagues[18] found that a concentrated surfactant-based wound dressing with 1% silver sulfadiazine (SSD) enhanced wound closure and caused a reduction in inflammation, odour, and pain. This suggests that poloxamer-based surfactants may have the ability to function biologically to help remove barriers to effective wound healing.

If surfactants are going to be used as part of a wound-cleaning strategy, it is important to understand the different types of surfactants as they differ significantly in their behaviours and biological effects.

3 | BRIEF BACKGROUND ON SURFACTANTS AND CLASSIFICATION

Surfactants have been used for hundreds of years, with many approved by the US Food and Drug Administration (FDA) and the UK based Medicines and Healthcare Products Regulatory Agency (MHRA) for use in medical conditions and for the delivery of drugs and antimicrobials. Surfactants have the ability to reduce the surface tension between two immiscible agents. They are therefore used in an array of different products, including detergents and cosmetics, emulsions, and paints.[19]

Surfactants are referred to as "surface active agents", which contain both hydrophobic and hydrophilic groups. It is the hydrophobic part that sticks to debris and dirt, and the hydrophilic component enables it to be washed away. Because of their amphiphilic abilities, they are being used in an array of different applications.[20] Many surfactants are also being used as antimicrobial agents that have a broad spectrum of activity against an array of different microorganisms.[21,22]

Surfactants can be classified based on their charge or absence of ionisation of the hydrophilic group, that is, cationic, anionic, non-ionic, and amphoteric or zwitterionic agents.[23] Surfactants such as quaternary ammonium compounds (QACs) are cationic surfactants that are used as disinfectants within the food industry and used in numerous medical situations[24] as they are useful in cleaning and deodorising.[23] Because of the positive charge of QACs, they bind to negatively charged areas on microbes. This results in stress to the cell wall, lysis, and then death. These agents can also cause protein denaturation, affecting cell wall permeability and reducing the uptake of nutrients.[25] Anionic surfactants are strong detergents but are not considered very antimicrobial; however, they have been reported to cause lysis in Gram-negative bacteria.[26] All charged surfactants are generally toxic to wound cells.[27] Non-ionic surfactants include agents such as polyalkylene glycols. The non-ionic surfactants do not ionise in the presence of water and are regarded as having only a low ability to irritate cells. An example of a non-ionic surfactant is poloxamer. Non-ionic-based surfactants, such as ones incorporating poloxamer 188, are considered non-cytotoxic and therefore represent a useful combination in wound care.[28]

When surfactants are mixed in water, as mentioned previously, they reduce the surface tension of the water. In this situation, as the surfactant concentration increases, the surface tension will continue to drop. When a certain concentration of surfactants is reached, the surfactant molecules will form micelles. As the micelles form, any further addition of surfactants will not have any further effect on the surface tension. At this concentration, where the surface tension remains constant, the critical micelle concentration has been formed. Micelles that are formed by surfactants are able to trap hydrophobic molecules at their hydrophobic core and will act as a wetting agent, which therefore makes them very effective cleaning agents. The micelle size that is formed is related to the number of monomers per micelle or the micelles' molecular weight.

4 | POLOXAMER-BASED SURFACTANTS

Poloxamers are tri-block copolymers composed of a central hydrophobic core (polyoxypropylene) that is flanked by two hydrophilic chains of poloxethylene. Poloxamer-based surfactants were historically used in reducing the viscosity of blood before a transfusion, and they are also found in laxatives, mouth washes, and toothpastes. Gels formed by surfactants have been reported, with the formation of a gel through a process known as micellisation.[29] Poloxamers are non-ionic surfactants composed of ethylene oxide and propylene oxide, and many can be found listed in the FDA inactive ingredient guide database for use in formulations used

in the pharmaceutical industry. Poloxamer-based surfactants are water-soluble tri-block copolymers, abbreviated to poly-oxyethylene (POE)-polyoxypropylene (POP)-POE, that have an average molecular weight of 8400 Da. The POE and POP are referred to as poly(oxyethlene) and poly(oxypropylene). The POE chains are hydrophilic, and the POP chains are hydrophobic.

5 | USE OF SURFACTANTS IN WOUND CARE

The main surfactant classes used in wound care and within wound dressings include betaines and poloxamers. However, it is also important to highlight that some antimicrobial agents, such as chlrohexidine, PHMB, and benzylkonium chloride, whilst effective as antimicrobial agents at high concentrations, are being exploited for their surfactant-based abilities at much lower concentrations. Surfactant-based wound dressings are known to have the ability to soften, moisturise, and also loosen cellular debris, with evidence that some have the ability to help in the dispersion/break up of the biofilm and also prevent biofilm formation.[12,30]

Zölß and Cech[28] evaluated the efficacy of a concentrated surfactant gel (CSG) that contained 1% SSD on 226 patients with chronic wounds. Of the patients, 88 were maintained on standard of care and then entered into the study, and the other 138 had been treated with CSG before the study. After a median of 17 weeks, 73% healed or improved. The study also demonstrated a potential improvement of reduced treatment costs compared with standard protocols of care. A study by Ratliff[31] assessed the performance of a concentrated surfactant-based gel on 18 patients with full-thickness wounds over a period of 4 weeks, with positive outcomes with the CSG dressing being shown to be effective in cleaning the wound from slough and necrotic debris.

Numerous studies have indicated a role that combining antimicrobials and surfactants could play, demonstrating that, by combining antimicrobials with surfactants, an enhancement of the antimicrobial could be achieved.[32] Babickaite and colleagues[33] found that, by combining poloxamer and chlorhexidine, biofilm eradication could be achieved but that the dosage of the antiseptic was very important to its antimicrobial activity. An additional study by Demirci and colleagues[34] evaluated the effect of a hydrogel containing poloxamer and boron on wound healing. The formulation was found to be very effective in promoting wound healing. It was reported to stimulate cell migration, growth factors, and vascularisation.[30] A further study by Leyva-Gómez et al[35] combined the properties of chitosan and poloxamer. In mouse models, it was found that the gel, when added to full-thickness mouse wounds, reduced the wound area significantly in a few days. Also observed in this study was the ability of the gel to increase macrophage proliferation and collagen depositions. Leszczyńska et al[36] assessed the in vitro antimicrobial and haemolytic ability of Ceragenin cationic steroid antimicrobial 13 (CSA-13; synthetic mimic of cationinc antbacterial peptides) in the presence of poloxamer. By itself, CSA-13 exhibited antibacterial activity, but in the presence of poloxamer, antibacterial activity was slightly reduced, but haemolytic activity was inhibited. Yanai et al[37] found that the antimicrobial activity of PHMB can be inhibited by NaCl in a concentration-dependent manner, but in the presence of Poloxamer 407 (4%), the activity of PHMB towards *Staphylococcus aureus* and fungi was increased.

6 | SURFACTANTS AND BIOFILMS

Both synthetic and natural surfactants are being used in the management of biofilms.[38] Azeredo et al[39] evaluated the effects of surfactants on biofilm detachment. Comparing the efficacy of surfactants sodium dodecyl sulfate (SDS) and cetyl trimethyl ammonium bromide (CTAB) in detaching *Pseudomonas fluorescens* biofilms from glass surfaces, they found that SDS was able to remove almost all of the attached bacteria relatively quickly. However, CTAB did not cause the promotion of cellular detachment, indicating that different surfactants have different abilities in affecting the detachment of microbes from a surface. Yang et al[12] evaluated a concentrated surfactant-based gel dressing on biofilms on porcine skin. Following daily applications and removal of the CSG dressing after 3 days, biofilms were reduced to undetectable levels. Díaz De Rienzo et al[40] also found evidence that surfactants had the ability to cause the dispersion of *Pseudomonas aeruginosa* biofilms. Satputea et al[38] discussed the various roles that biosurfactants can play in the strategy to combat biofilms in light of the growing need for synthetic-based surfactants. Quinn et al[41] compared various biosurfactants on established biofilms. They found that rhamnolipids and plant-derived surfactants could reduce biofilm biomass. Unfortunately, currently, there are only a limited number of studies that have investigated the role and significance of biofilms in the context of wound healing. These have principally focused on poloxamer-based wound technologies.

The role surfactants play in both the prevention and control of biofilms is presently being investigated, with interesting results now being reported. For example, a recent study by Yu et al[42] found that the surfactants CTAB and SDS demonstrated effects on hyphal development in *Candida albicans.* Hyphal development is known to be involved in biofilm formation.[43] Based on this, the authors hypothesized that the surfactants may have a role to play in affecting biofilm development. Their results demonstrated that the surfactants inhibited biofilm formation and also reduced the activity of pre-formed biofilms. The IC_{50} against biofilm formation and biofilm maintenance was 0.888 and 4.061 ppm for CTAB and 76.092 ppm and > 160 ppm for SDS,

respectively. Biosurfactants are being utilised as anti-biofilm agents with promising results.[44]

Yang et al[12] investigated if wiping on a daily basis combined with daily application of a surfactant (poloxamer) would reduce the level of mature biofilm grown on porcine pig skin explants. The results showed that daily wiping of the pig skin explants with moistened gauze combined with application of poloxamer 188 eliminated Pseudomonas aeruginosa planktonic and biofilm bacteria after three days of daily treatment. Polysorbate 80 is another surfactant that is utilised in pharmaceutical preparations. They are well known to increase the permeability of the membranes in bacteria. This has been documented in *P. aeruginosa.*[45,46] Furthermore, polysorbate 80 has also been shown to inhibit biofilms in *P. aeruginosa* and *Escherichia coli*.[47,48] Malinowski et al[49] investigated the effects of polymyxin B and polysorbate 80, and it was found to inhibit the growth and biofilm formation of *Stenotrophomonas maltophilia.* However, the authors were unable to show how PS80-inhibited biofilms.

7 | SURFACTANTS AND ROLE ON THE BIOFILM MATRIX—EPS

The ability of surfactants to break down the EPS component of the biofilms is presently lacking warranting studies in this area. As mentioned previously, because the EPS represents a very important component of the biofilm, and its prevention and control is significant in biofilm management.

8 | ROLE OF SURFACTANTS IN THE PREVENTION OF CELL DEATH ("CELL SALVAGE")

Whilst many surfactants have been reported to have a broad antimicrobial effect, their mechanisms of action against microbes still remains to be completely elucidated. Although they have traditionally been reported to cause the disruption of biological membranes,[50,51] they have also been reported to inhibit certain enzymes, leading to the enhanced production of ROS, which is also considered part of the antimicrobial ability of some surfactants.[52] ROS increase by the inhibition of certain ROS suppressive enzymes makes the surfactants antimicrobials. Despite these new studies, some surfactants do not affect the plasma membrane, indicating that ROS may not be a major factor in their mode of action.[42,53] With concentrated poloxamer 188 gel it is reported to cause an upregulation of gelatinase and down regulation of collagenase type enymes. This profile of enzyme regulation is suitable for a slough containing wound because gelatin as such is roughly translatable as denatured, necrotic tissue associated collagen. There are also reports of surfactants affecting MMP activity.[54] Yu and colleagues[42] also demonstrated the effects surfactants have on MMPs in *C. albicans.* In their studies, the researchers speculated that the surfactants enter fungal cells and interact with mitochondrial membranes, subsequently leading to the dissipation of the proton gradient across the inner membrane and to a decrease in MMP levels. Surfactants such as cetyl trimethylammonium bromide are reported to promote apoptosis of cancer cells, but it is probable that the ROS leads to a reduction in MMPs. It remains important to investigate the potential relationship between the use of surfactants and oxidative stress.

Surfactants are known to interact with, and therefore affect, microbial proteins, leading to effects on enzymatic stability and activity.[55] It is also well documented that properties such as osmotic pressure, surface tension, and conductivity will either decrease or increase as the surfactant concentration increases.

Some examples of effective surfactants that are showing good potential in wound care are poloxamer-based surfactants. They are documented as being able to incorporate themselves in the phospholipid bilayer of cells, which in turn helps to cause the resuscitation of cells.[56] Studies by Barbee et al [57] and Marks et al[58] have also shown that poloxamers "save" neurons from necrotic death. Yuhua et al[59] and Lee et al[60] have evaluated the use of poloxamers in burn wounds and have shown that poloxamer 188 has the ability to repair cells that are often damaged. During cell death, many cells swell, resulting in the formation of an injured cell that is unable to maintain ionic gradients in the plasma membrane, which then leads to the cell becoming ruptured.[61] Some surfactants have been documented to be able to interact with the lipid bilayer of the plasma membrane with positive outcomes to the cell. For example, studies by Clarke et al[62] and Papoutsakis[63] have found that surfactants have the ability to restore the integrity of cell membranes following stresses caused by electrical[64] and chemical effects.[65] Studies by Philips et al[66] were set up to investigate the ability of poloxamer 118 to "save" cells from necrosis. The study found that P188 significantly increased the percentage of live cells and also increased cellular viability. This study and further studies alike have demonstrated that poloxamer 188 has the ability to repair cells that are damaged.[56,62–65] Furthermore, Kaisang et al[67] found that, when allogeneic non-diabetic adipose-derived stem cells (ADSCs) were added to hydrogel composed of poloxamer and topically applied to a full-thickness cutaneous wound in diabetic rats, angiogenesis was increased together with cell proliferation and enhanced wound closure. In conclusion, the authors suggested that the use of ADSCs in combination with surfactants, in this case poloxamers, may represent a "novel therapeutic strategy" of the treatment of non-healing diabetic foot ulcers.

9 | OVERALL DISCUSSION ON CLEANSING, SURFACTANTS, BIOFILMS, AND WOUNDS

Based on the evidence to date, a well-planned and systemic approach to wound cleaning is warranted to prepare the

wound for interventions and procedures that are going to be utilised when a biofilm-based wound management strategy is required. Antimicrobials and anti-biofilm agents are designed to function at very low concentrations but are known to have reduced efficacy in biological systems as proteins, for example, will reduce the bioavailability of the active components. Therefore, many of these agents are not being delivered to the wound at therapeutic concentrations and above (considered more appropriate in biofilm-related conditions). Consequently, if wounds are not effectively cleaned of barriers to wound healing, that is, slough, devitalised tissue, microorganisms, and biofilms, prior to the administration of a topical antimicrobial, this will affect the antimicrobial's performance and short-term residual activity such that maximum efficacy cannot be achieved to reduce the wound's microbial bioburden and reduce biofilm development and maturation.[1] Accordingly, the likelihood of a positive clinical outcome will be significantly reduced if wounds are not cleaned very early during the wound treatment process.

The management of biofilms in wounds is a complex process principally because of the unpredictability of the biofilm's physiology. In part, this is often because of a wound biofilm's inherent microbiological composition. In long-term non-healing wounds, "a new microbiome" exists, which highlights the microbial complexity now identified in wounds.[68] Changes to this wound microbiome, as with other chronic conditions, can have a significant impact on the biology of wound healing. The phenomena of a potential dysbiosis of an established chronic wound's microbiome may become an issue for infection in wounds, a phenomenon that has been demonstrated in biofilms within the gastrointestinal tract and the oral cavity and causes a shift in disease and infection status.

In the authors' opinion, a significant component of the biofilm management process should involve effective wound cleaning using surfactants.[11] This is, in principal, because of their inherent ability to offer many characteristics that function to support wound healing. Using technology that is known to be effective in other industries with positive outcomes in biofilm prevention and control represents a component that could be used to help advance wound healing in light of the growing need for more effective products and procedures for the management of biofilms in wounds.[69]

10 | CONCLUSION

Surfactants, but more specifically poloxamers, have been used in thousands of patients with no serious side effects and are documented to be well tolerated by patients after topical use[18,70,71] and also intravenous usage.[72] They have also been shown to potentially enhance the "normal" wound-healing process.[18,73–75] Historically, one of the concerns with using poloxamer-based surfactant was that these surfactants have been shown to lack antimicrobial activity.[76] However, there is now growing evidence that poloxamers have a significant effect on biofilms, suggesting a new role of poloxamer-based surfactants in wound cleaning and also in biofilm prevention and control. In addition, evidence suggests that, by combining surfactants with antimicrobial agents, enhanced performance of the antimicrobial could be achieved with positive outcomes. Furthermore, there is growing evidence that poloxamers are capable of reducing and preventing cell death by temporarily "jumping in" to replace trauma or ROS damaged cell membranes until repair has taken place by the cell itself[11] and by also modulating immunological functions.[77] Thus, another mode of action of poloxamer may be its ability to counteract the devastating effects of ROS on cells, therefore enhancing wound healing.

Overall, based on the evidence to date, poloxamer-based technologies appear to be demonstrating an ability to enhance wound healing because of their inherent characteristics to supress and down-regulate many of the detrimental factors known to delay wound healing, that is, biofilms, MMP (collagenase) activity, and inflammation, but in upregulating or promoting cellular integrity leading to cellular proliferation.[75]

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CONFLICTS OF INTEREST

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REFERENCES

1. Percival SL, Suleman L. Slough and biofilm: removal of barriers to wound healing by desloughing. *J Wound Care*. 2015;24(11):498, 500-3, 506-10-510.
2. Percival SL, Hill KE, Williams DW, Hooper SJ, Thomas DW, Costerton JW. A review of the scientific evidence for biofilms in wounds. *Wound Repair Regen*. 2012;20(5):647-657.
3. Mast BA, Schultz GS. Interactions of cytokines, growth factors, and proteases in acute and chronic wounds. *Wound Repair Regen*. 1996;4(4):411-420.
4. Percival SL, Mayer D, Salisbury AM. Efficacy of a surfactant-based wound dressing on a biofilm control. *Wound Repair Regen*. 2017;25(5):767-773.
5. Flemming HC, Wingender J. The biofilm matrix. *Nat Rev Microbiol*. 2010;8(9):623-633.
6. Flemming HC. EPS-then and now. *Microorganisms*. 2016;4(4):E41.
7. Flemming HC, Neu TR, Wozniak DJ. The EPS matrix: the "house of biofilm cells". *J Bacteriol*. 2007;189(22):7945-7947.
8. Hemmi H, Takeuchi O, Kawai T. A toll-like receptor recognises bacterial DNA. *Nature*. 2000;408(6813):740-745.
9. Lammers RL. Principles in wound management. In: Roberts JR, Hedges JR, eds. *Clinical Procedures in Emergency Medicine*. Philadelphia, PA: WB Saunders; 1985:478-530.
10. Percival SL. Restoring balance: biofilms and wound dressings. *J Wound Care*. 2018;27(2):102-113.

11. Mayer D, Armstrong D, Schltz G, et al. Cell salvage in acute and chronic wounds: a potential treatment strategy. Experimental data and early clinical results. *J Wound Care*. 2018;27(9):594-605.
12. Yang Q, Larose C, Della Porta AC, Schultz GS, Gibson DJ. A surfactant-based wound dressing can reduce bacterial biofilms in a porcine skin explant model. *Int Wound J*. 2017;14(2):408-413.
13. Keast D, Swanson T, Black JM, et al. Ten top tips: understanding and managing wound biofilm. *Wounds Int*. 2014;6(2):22-24.
14. Johani K, Malone M, Jensen SO, et al. Evaluation of short exposure times of antimicrobial wound solutions against microbial biofilms: from in vitro to in vivo. *J Antimicrob Chemother*. 2018;73(2):494-502.
15. Castaneda P, McLaren A, Tavaziva G, Overstreet D. Biofilm antimicrobial susceptibility increases with antimicrobial exposure time. *Clin Orthop Relat Res*. 2016;474(7):1659-1664. https://doi.org/10.1007/s11999-016-4700-z.
16. Vestby LK, Nesse LL. Wound care antiseptics - performance differences against *Staphylococcus aureus* in biofilm. *Acta Vet Scand*. 2015;57:22.
17. Bellingeri A, Falciani F, Traspedini P, et al. Effect of a wound cleansing solution on wound bed preparation and inflammation in chronic wounds: a single-blind RCT. *J Wound Care*. 2016;25(3):160-168.
18. Palumbo FP, Harding KG, Abbritti F. New surfactant-based dressing product to improve wound closure rates of non-heaiing wounds: a multicentre study including 1036 patients. *Wounds*. 2016;28(7):233-240.
19. Kumar N, Tyagi R. Industrial Applications of Dimeric Surfactants: A Review. *J Dispers Sci Technol*. 2014;35(2):205-214.
20. Singh P, Cameotra SS. Potential applications of microbial surfactants in biomedical sciences. *Trends Biotechnol*. 2004;22:142-146.
21. Nicoletti G, Boghossian V, Gurevitch F, Borland R, Morgenroth P. The antimicrobial activity in vitro of chlorhexidine, a mixture of isothiazolinones ('Kathon' CG) and cetyl trimethyl ammonium bromide (CTAB). *J Hosp Infect*. 1993;23:87-111.
22. Sutterlin H, Alexy R, Kummerer K. The toxicity of the quaternary ammonium compound benzalkonium chloride alone and in mixtures with other anionic compounds to bacteria in test systems with Vibrio fischeri and pseudomonas putida. *Ecotoxicol Environ Saf*. 2008;71:498-505.
23. McDonnell G, Russell AD. Antiseptics and disinfectants: activity, action, and resistance. *Clin Microbiol Rev*. 1999;12:147-179.
24. Mereghetti L, Quentin R, Marquet-Van Der Mee N, Audurier A. Low sensitivity of *Listeria monocytogenes* to quaternary ammonium compounds. *Appl Environ Microbiol*. 2000;66(11):5083-5086.
25. Cloete TE, Jacobs L, Brözel VS. The chemical control of biofouling in industrial water systems. *Biodegradation*. 1998;9(1):23-37.
26. Glover RE, Smith RR, Jones MV, Jackson SK, Rowlands CC. An EPR investigation of surfactant action on bacterial membranes. *FEMS Microbiol Lett*. 1999;177(1):57-62.
27. Rodeheaver GT, Ratliff CR. Wound cleansing, wound irrigation, wound disinfection. In: Krasner DL, ed. *Chronic Wound Care: The Essentials*. Malvern, PA: HMP Communications, LLC; 2014:47-61.
28. Zölß C, Cech JD. Efficacy of a new multifunctional surfactant-based biomaterial dressing with 1% silver sulphadiazine in chronic wounds. *Int Wound J*. 2016;13:738-743.
29. Raghavan SR. Distinct character of surfactant gels: a smooth progression from micelles to fibrillar networks. *Langmuir*. 2009;25(15):8382-8385.
30. Yang Q, Schultz GS, Gibson DJ. A Surfactant-Based Dressing to Treat and Prevent Acinetobacter baumannii Biofilms. *J Burn Care Res*. 2018;39(5):766-770.
31. Ratliff CR. Case series of 18 patients with lower extremity wounds treated with a concentrated surfactant-based gel dressing. *J Vasc Nurs*. 2018;36(1):3-7.
32. Belfield K, Bayston R, Hajduk N, Levell G, Birchall JP, Daniel M. Evaluation of combinations of putative anti-biofilm agents and antibiotics to eradicate biofilms of *Staphylococcus aureus* and *Pseudomonas aeruginosa*. *J Antimicrob Chemother*. 2017;72(9):2531-2538.
33. Babickaite L, Ramanauskiene K, Grigonis A, et al. Determination of antimicrobial activity of chlorhexidine gel. *Acta Pol Pharm*. 2016;73(6):1623-1630.
34. Demirci S, Doğan A, Karakuş E, et al. Boron and Poloxamer (F68 and F127) containing hydrogel formulation for burn wound healing. *Biol Trace Elem Res*. 2015;168(1):169-180.
35. Leyva-Gómez G, Santillan-Reyes E, Lima E, et al. A novel hydrogel of poloxamer 407 and chitosan obtained by gamma irradiation exhibits physicochemical properties for wound management. *Korean J Couns Psychother*. 2017;74:36-46.
36. Leszczyńska K, Namiot A, Cruz K, et al. Potential of ceragenin CSA-13 and its mixture with pluronic F-127 as treatment of topical bacterial infections. *J Appl Microbiol*. 2011;110(1):229-238.
37. Yanai R, Ueda K, Nishida T, Toyohara M, Mori O. Effects of ionic and surfactant agents on the antimicrobial activity of polyhexamethylene biguanide. *Eye Contact Lens*. 2011;37(2):85-89.
38. Satputea SK, Banpurkar AG, Banat IM, Sangshetti JN, Patil RH, Gade WN. Multiple Roles of Biosurfactants in Biofilms. *Curr Pharm Des*. 2016;22(11):1429-1448.
39. Azeredo J, Pacheco AP, Lopes I, Oliveira R, Vieira MJ. Monitoring cell detachment by surfactants in a parallel plate flow chamber. *Water Sci Technol*. 2003;47:77-82.
40. Díaz De Rienzo MA, Stevenson PS, Marchant R, Banat IM. *Pseudomonas aeruginosa* biofilm disruption using microbial surfactants. *J Appl Microbiol*. 2016;120(4):868-876.
41. Quinn GA, Maloy AP, Banat MM, Banat IM. A comparison of effects of broad-spectrum antibiotics and biosurfactants on established bacterial biofilms. *Curr Microbiol*. 2013;67(5):614-623.
42. Yu Q, Zhang B, Ma F, et al. Novel mechanisms of surfactants against *Candida albicans* growth and morphogenesis. *Chem Biol Interact*. 2015;227:1-6.
43. Finkel JS, Mitchell AP. Genetic control of *Candida albicans* biofilm development. *Nat Rev Microbiol*. 2011;9(2):109-118.
44. Banat IM, De Rienzo MA, Quinn GA. Microbial biofilms: biosurfactants as antibiofilm agents. *Appl Microbiol Biotechnol*. 2014;98(24):9915-9929.
45. Brown MRW, Winsley BE. Effect of polysorbate 80 on cell leakage and viability of *Pseudomonas aeruginosa* exposed to rapid changes of pH, temperature and tonicity. *J Gen Microbiol*. 1969;56:99-107.
46. Brown MR, Geaton EM, Gilbert P. Additivity of action between polysorbate 80 and polymyxin B towards spheroplasts of *Pseudomonas aeruginosa* NCTC 6750. *J Pharm Pharmacol*. 1979;31:168-170.
47. Toutain-Kidd CM, Kadivar SC, Bramante CT, Bobin SA, Zegans ME. Polysorbate 80 inhibition of *Pseudomonas aeruginosa* biofilm formation and its cleavage by the secreted lipase LipA. *Antimicrob Agents Chemother*. 2009;53:136-145.
48. Sloup RE, Cieza RJ, Needle DB, Abramovitch RB, Torres AG, Waters CM. Polysorbates prevent biofilm formation and pathogenesis of *Escherichia coli* O104:H4. *Biofouling*. 2016;32:1131-1140.
49. Malinowski AM, McClarty BM, Robinson C, et al. Polysorbate 80 and polymyxin B inhibit *Stenotrophomonas maltophilia* biofilm. *Diagn Microbiol Infect Dis*. 2017;87(2):154-156.
50. Hamouda T, Baker JR Jr. Antimicrobial mechanism of action of surfactant lipid preparations in enteric Gram-negative bacilli. *J Appl Microbiol*. 2000;89:397-403.
51. Chen C, Hu J, Zhang S, et al. Molecular mechanisms of antibacterial and anti-tumor actions of designed surfactant-like peptides. *Biomaterials*. 2012;33:592-603.
52. Nakata K, Tsuchido T, Matsumura Y. Antimicrobial cationic surfactant, cetyltrimethylammonium bromide, induces superoxide stress in *Escherichia coli* cells. *J Appl Microbiol*. 2011;110:568-579.
53. Percival SL, Chen R, Mayer D, Salisbury AM. Mode of action of poloxamer-based surfactants in wound care and efficacy on biofilms. *Int Wound J*. 2018;15(5):749-755.
54. Jeong S, Schultz GS, Gibson DJ. Testing the influence of surfactant-based wound dressings on proteinase activity. *Int Wound J*. 2017;14(5):786-790.
55. Kamiya N, Inoue M, Goto M, Nakamura N, Naruta Y. Catalytic and structural properties of surfactant-horseradish peroxidase complex in organic media. *Biotechnol Prog*. 2000;16:52-58.
56. Maskarinec SA, Hannig J, Lee RC, Lee KY. Direct observation of poloxamer 188 insertion into lipid monolayers. *Biophys J*. 2002;82(3):1453-1459.
57. Barbee KA, Kilinc D, Serbest G, Horwitz J, Gallo G. Cellular mechanisms of neural injury and the neuroprotective effects of poloxamer 188. *J Biomech*. 2006;39:S580.
58. Marks JD, Pan CY, Bushell T, Cromie W, Lee RC. Amphiphilic, tri-block copolymers provide potent membrane-targeted neuro- protection. *FASEB*. 2001;15:1107-1109.
59. Yuhua S, Ligen L, Jiake C, Tongzhu S. Effect of Poloxamer 188 on deepening of deep second-degree burn wounds in the early stage. *Burns*. 2012;38(1):95-101.
60. Lee RC, River LP, Pan FS, Ji L, Wollmann RL. Surfactant-induced sealing of electropermeabilized skeletal muscle membranes *in vivo*. *Proc Natl Acad Sci U S A*. 1992;89(10):4524-4528.

61. Duke RC, Ojcius DM, Young JDE. Cell suicide in health and disease. *Sci Am*. 1996;275:80-87.

62. Clarke MSF, McNeil PL. Syringe loading introduces macro- molecules into living mammalian cell cytosol. *J Cell Sci*. 1992;102:533-541.

63. Papoutsakis ET. Media additives for protecting freely suspended animal cells against agitation and aeration damage. *TIBTECH*. 1991;9:316-324.

64. Lee RC, Hannig J, Matthews KL, Myerov A, Chen CT. Pharmaceutical therapies for sealing of permeabilized cell mem- branes in electrical injuries. *Ann N Y Acad Sci*. 1999;888:266-273.

65. Hellung-Larsen P, Assaad F, Pankratova S, Saietz BL, Skovgaard LT. Effects of pluronic F-68 on tetrahymena cells: protection against chemical and physical stress and prolongation of survival under toxic conditions. *J Biotechnol*. 2000;76:185-195.

66. Phillips DM, Haut RC. The use of a non-ionic surfactant (P188) to save chondrocytes from necrosis following impact loading of chondral explants. *J Orthop Res*. 2004;22(5):1135-1142.

67. Kaisang L, Siyu W, Lijun F, Daoyan P, Xian CJ, Jie S. Adipose-derived stem cells seeded in Pluronic F-127 hydrogel promotes diabetic wound healing. *J Surg Res*. 2017;217:63-74.

68. Misic AM, Gardner SE, Grice EA. The wound microbiome: modern approaches to examining the role of microorganisms in impaired chronic wound healing. *Adv Wound Care (New Rochelle)*. 2014;3(7):502-510.

69. Schultz G, Bjarnsholt T, James GA, et al. Consensus guidelines for the identification and treatment of biofilms in chronic non-healing wounds. *Wound Repair Regen*. 2017;25:744-757.

70. Edlich RF, Schmolka IR, Prusak MP, Edgerton MT. The molecular basis for toxicity of surfactants in surgical wounds. 1. EO:PO block polymers. *J Surg Res*. 1973;14(4):277-284.

71. Du L, Tong L, Jin Y, et al. A multifunctional in situ-forming hydrogel for wound healing. *Wound Repair Regen*. 2012;20(6):904-910.

72. Grindel JM, Jaworski T, Piraner O, Emanuele RM, Balasubramanian M. Distribution, metabolism, and excretion of a novel surface-active agent, purified poloxamer 188, in rats, dogs, and humans. *J Pharm Sci*. 2002;91:1936-1947.

73. Fowler EB, Cuenin MF, Hokett SD, et al. Evaluation of pluronic polyols as carriers for grafting materials: study in rat calvaria defects. *J Periodontol*. 2002;73:191-197.

74. Kant V, Gopal A, Kumar D, et al. Topical pluronic F-127 gel application enhances cutaneous wound healing in rats. *Acta Histochem*. 2014;116:5-13.

75. Ghatak PD, Mathew-Steiner SS, Pandey P, Roy S, Sen CK. A surfactant polymer dressing potentiates antimicrobial efficacy in biofilm disruption. *Sci Rep*. 2018;8(1):873.

76. Rodeheaver GT, Smith SL, Thacker JG, Edgerton MT, Edlich RF. Mechanical cleansing of contaminated wounds with a surfactant. *Am J Surg*. 1975;129:241-245.

77. Moghimi SM, Murray JC. Poloxamer-188 revisited: a potentially valuable immune modulator? *J Natl Cancer Inst*. 1996;88:766-768.

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