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Slough and biofilm: removal of barriers to wound healing by desloughing

S.L. Percival,[1,2] PhD CEO;
L. Suleman,[1,2] PhD, Scientific Development Executive;
1 5D Health Protection Group Ltd, Biohub, Alderley Park, Alderley Edge, Cheshire, SK10 4TG, UK
2 Institute of Ageing and Chronic Disease, University of Liverpool, Liverpool, UK.

Email: Steven.Percival@5Dhpg.com

Slough and biofilm: removal of barriers to wound healing by desloughing

The presence of non-viable tissue in a chronic wound presents a barrier against effective wound healing, hence removal facilitates healing and reduces areas where microorganisms can attach and form biofilms, effectively reducing the risk of infection. Wound debridement is a necessary process in those wounds that have evidence of cellular debris and non-viable tissue. As slough is a form of non-viable tissue we hypothesise that it will support the attachment and development of biofilms. Biofilms are entities that have serious implications in raising the risk of infection and delaying wound healing. In those wounds that contain only slough, high-risk debridement methods are not considered necessary for its removal. The use of mechanical techniques for removing the slough is regarded as posing a much lower risk to the patient and the wound bed. The process of removing slough from a wound is referred to as 'desloughing'. We propose that mechanical desloughing is a low-risk method of debridement to aid the specific removal of slough. Slough in a wound is a recurrent issue for a large majority of patients. Consequently, desloughing should not be deemed a one-off process but an on-going procedure referred to as 'maintenance desloughing'. Maintenance desloughing will help to achieve and maintain a healthy wound bed and aid the removal of wound biofilms, facilitating wound healing.

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biofilm; slough; chronic wound; debridement; desloughing

S.L. Percival,[1,2] PhD CEO;
L. Suleman,[1,2] PhD, Scientific Development Executive;
1 5D Health Protection Group Ltd, Biohub, Alderley Park, Alderley Edge, Cheshire, SK10 4TG, UK
2 Institute of Ageing and Chronic Disease, University of Liverpool, Liverpool, UK.

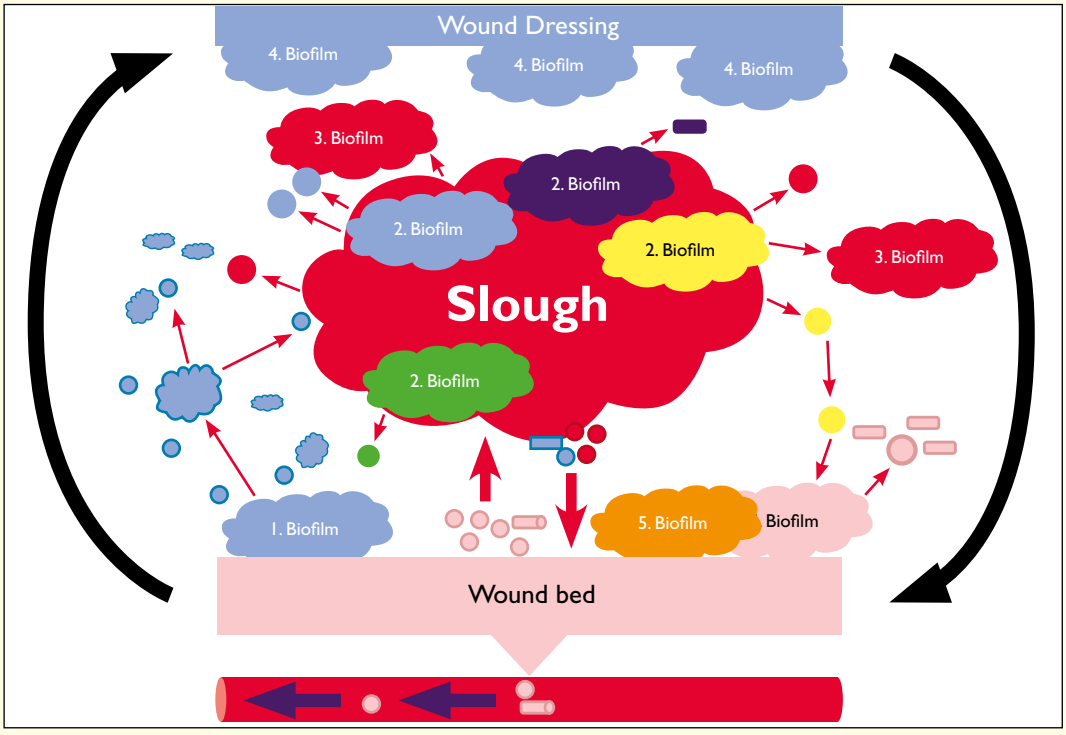
Email: Steven.Percival@5Dhpg.com

There are a number of circumstances where the wound healing process is disrupted, causing a significant delay in wound closure. These wounds are referred to as chronic. Common examples of chronic wounds in humans include pressure ulcers (PUs), venous leg ulcers (VLUs) and diabetic foot ulcers (DFUs), which pose a considerable economic burden, costing the National Health Service (NHS) an estimated £2.3–£3.1 billion per year.[1] Sheehan and Jones, in reference to DFUs, defined these chronic wounds as wounds that fail to show a decrease of 50% of their volume within one month, so that closure could not be achieved within 12 weeks.[2]

Chronic wounds are open voids and are therefore susceptible to microbial colonisation and infection.[3] They need to be carefully managed to ensure healing and prevent the development of further complications. However, the longer a wound remains open, the higher the risk of microbial attachment, proliferation and the formation of a recalcitrant, virulent biofilm. Biofilms are microorganisms that can attach to each other or to biotic (living surfaces such as biological tissues) or abiotic surfaces (non-living surfaces such as a wound dressing). They are encased within a 3-dimensional matrix of extracellular material, called extracellular polymeric substance (EPS).[4] Biofilms are reported to be composed of 10–20% microorganisms and 80–90% extracellular material. EPS is composed of proteins, polysaccharides, anionic and cationic ions and extracellular DNA (eDNA) among other micro and macro components.[5] As a biofilm matures (the word ageing would not be deemed appropriate as there is currently no evidence to suggest that ageing occurs in biofilms as it does in mammals), recalcitrance to the host immune responses and antimicrobial treatments increases significantly.[6] Consequently, it is imperative that once an acute wound forms and the risk of becoming a chronic wound increases (when the interventions used have been unsuccessful in achieving wound repair and closure), then appropriate anti-biofilm therapies and strategies should be used.[7,8]

The presence of non-viable tissue is a prominent feature in many chronic wound types. Clinical observations of necrotic tissue describe necrotic tissue as hard, dry tissue that is black/dark brown in colour and firmly attached to the wound bed. Necrotic tissue has been reported to act as a barrier to wound healing.[9] A study has related the presence of necrotic tissue in burn wounds to high numbers of infiltrating neutrophils and increased levels of the pro-inflammatory cytokine interleukin-8 (IL-8) when compared with post-surgical burn wounds, which were associated with the production of growth factors, fibroblast growth factor (FGF), platelet-derived growth factor (PDGF) and epidermal growth factor (EGF), and enhanced granulation tis-

Fig1. Proposed sites of microbial adhesion and formation of biofilms within chronic wounds. 1. Biofilm formation on wound bed; 2. Biofilms residing in slough; 3. Biofilms suspended as microcolonies within the wound exudate; 4. Biofilms attached to wound dressings/wound dressing fibres/foreign objects, and 5. Biofilms on the surface of necrotic tissue



sue formation and epithelialisation.[10] Debridement of necrotic tissue is vital for a chronic wound to be transformed back to an acute wound.[11]

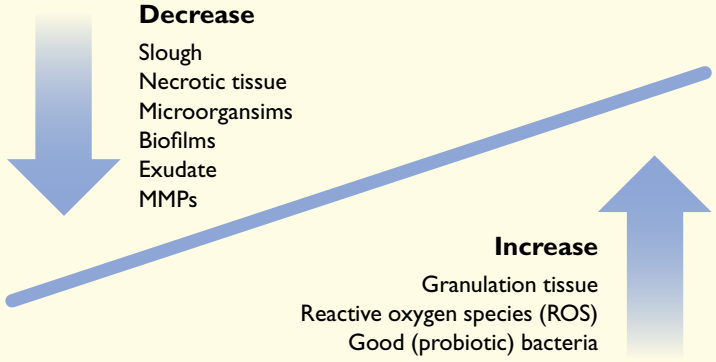
Another common feature in chronic wounds is the formation of slough. Slough within a wound presents as a moist, generally pale yellow entity that is usually tethered to the underlying wound bed. It can be patchy or sometimes semi-confluent over the wound area. Available evidence indicates that slough is composed of fibrin, pus, leucocytes, dead and living cells, microorganisms and proteinaceous materials, essentially a waste product from the immune-related clearance of redundant cellular debris and microorganisms Therefore, in a persistent state of inflammation, as seen in chronic wounds, the over-production of slough is a pathophysiological outcome. The estimated number of wounds that contain slough has not yet been reported. Anecdotal evidence suggests this number to be high however, there is no epidemiological data available. While there are clear phenotypical differences between necrotic tissue and slough, the physical, chemical and biological characterisation of slough has been under-researched (Table 1).

Here, we propose that biofilms may be able to form and thrive in non-viable tissues including necrotic tissue and slough. We believe that necrotic tissue and slough are indeed separate entities and that slough may share similar characteristics to a biofilm itself, although this has yet to be proven. Furthermore, we address the many potential geographical locations for formation of biofilms within

Table 1. The proposed characteristics of necrotic tissue and slough. The fields, which are highlighted in green, are the characteristics which are predominant for either necrotic tissue or slough. Fields highlighted in dark blue are shared characteristics between necrotic tissue and slough

Characteristics	Necrotic tissue	Slough
Black/dark brown	Generally	Not generally
Loosely attached	No	Yes–generally
Very firmly attached	Yes	No–not generally
Dead cells	Yes	Yes
Fibrin	Yes–low level	Yes–high level
Biofilm	Yes–more anaerobes	Yes–complex community
Microorganisms	Yes	Yes
White blood cells	No	Yes
'Houses' exudate	No	Yes
Viscoelastic	No	Yes

Fig 2. The management of factors that both impede and encourage wound healing. Characteristic factors such as increases in secretion of matrix metalloproteinases (MMPs), microbial bioburden, excess wound exudate and the formation of non-viable tissue such as slough and necrotic tissue should be reduced in order to encourage wound healing and restore balance. Similarly, the promotion of granulation tissue formation, reactive oxygen species (ROS) and potentially the use of probiotic bacteria may also lead to effective wound closure



a chronic wound. For instance, biofilms may be found in the:

- Wound exudate, consisting of microcolonies of microorganisms that aggregate together in the planktonic stage or have recently detached from a biofilm and have the same recalcitrant and phenotypic characteristics that are found in biofilms attached to a surface
- Wound bed, whereby aggregates of microorganisms can be found within the wound tissue[13,14]
- Wound dressing
- Necrotic tissue and
- Slough (Fig 1).

With this in mind, we will discuss the debridement techniques tailored towards to removal of necrotic tissue and slough presently used, including an introduction to the method of desloughing. Furthermore we suggest potential treatment pathways to aid an overall reduction in the total wound bioburden. Combining appropriate debridement and desloughing methods with other interventions will form part of an effective anti-biofilm strategy.[15,16]

Wound healing: a brief overview

Cutaneous wound repair comprises several complex and overlapping phases that are orchestrated in order to reduce bleeding, to clear contaminating microorganisms and cellular debris, and to produce new, vascularised tissue with an effective epithelial barrier. There are three major stages of wound healing: haemostasis and inflammation, re-epithelialisation and granulation tissue formation, and finally matrix remodelling.[4] For successful wound repair to occur, there must be a tightly regulated release of growth factors, cytokines and proteases to control cell migration, differentiation and proliferation and to control tissue architecture. Dysregulation of these key cellular and molecular components can lead to chronic wounds. Prominent examples of a chronic wound include DFUs, VLUs and PUs. Chronic wounds are halted in the inflammatory phase of wound repair and present with persistent inflammation.[12] The pathophysiology is unknown, however, a range of factors are considered to increase the risk of developing a chronic wound, including; smoking, diabetes mellitus, reduced mobility, nutrient deficiency, ischaemia and infection.[17] In addition, the presence of necrotic tissue and slough, excessive proteases and microorganisms within a wound can also act as 'barriers' to successful wound healing (Fig 2). Targeting these as part of routine wound management is essential to restore balance within the wound and encourage closure.

Implications of contamination within wounds: biofilms

A proposed characterisation suggests there are four states of microorganisms within a wound.[18] The first is contamination of the wound area with the presence of non-proliferating microorganisms on the superficial tissues, without eliciting a host immune response or affecting wound closure. The second state, microbial adhesion and colonisation, involves the contamination of the wound area with microorganisms, which proliferate and adhere to superficial tissues, giving rise to the formation of microbial microcolonies. Microbial adhesion and colonisation is not thought to induce a host immune response or affect wound closure.

A third state is referred to as 'critical colonisation'; this is a term coined to describe a delay in wound healing without clinical signs of inflammation. Here microorganisms have not managed to invade local tissues, however they are thought to secrete exotoxins and virulence factors that impair wound closure without eliciting an immune response. Critical colonisation is considered to be the point at which the wound can either improve following appropriate treatment, remain in a critical state, or deteriorate to a clinical infection.[18] However, it is important to note that critical colonisation is a theoretical concept that has come up against much scrutiny, and it may be more appropriately referred to as 'sub-clinical infection'.[3,19]

The final state is microbial infection and is characterised by the presence of proliferating microorganisms that have invaded viable tissues and therefore initiated a host immune response. The clinical characteristics of microbial infection include tissue redness (erythema), pain, heat, swelling and excessive exudate at the site. This type of microbial infection is considered to impede wound closure via sev-

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eral mechanisms including increased host protease and pro-inflammatory cytokine production and increased competition for oxygen and nutrients between both host cells and microorganisms.[19,20]

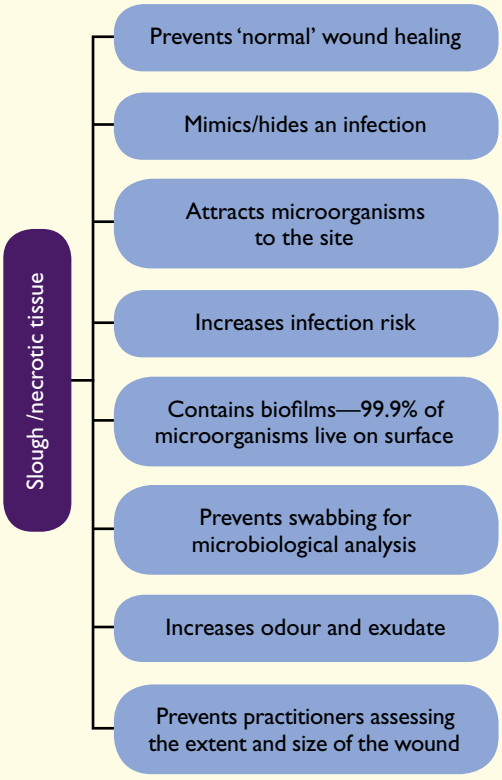
Evidence of biofilms in chronic wounds

It has been proposed that microorganisms within a chronic wound reside within a biofilm. Biofilms can be described as complex communities of microorganisms that reside in a self-synthesised matrix of EPS.[4] Biofilms were first studied in detail in the 1970s and reported to be bacteria encased in a self-synthesised fibrous matrix.[21] One early observation of biofilms in a medical setting was on the sutures of surgical wounds.[22]

Biofilms are a major health concern, primarily due to their increased tolerance, (as opposed to resistance, which implies a genetic effect), to antimicrobial therapies. Recent reviews have reported all the current evidence supporting the presence of biofilms in chronic wounds.[23–25] A particular study by James and colleagues investigated the presence of biofilms in both acute and chronic wounds, using scanning electron microscopy (SEM), and the microbial profile of the wounds using denaturing gradient gel electrophoresis (DGGE). A significant difference in the presence of biofilms between chronic and acute wounds was determined, with 60% of chronic wounds containing a biofilm compared with just 6% in acute wounds. Furthermore, DGGE revealed that these biofilms were polymicrobial.[13] The presence of biofilms in chronic wounds has also been detected using peptide nucleic acid-based fluorescence *in situ* hybridisation (PNA-FISH), ultimately determining the structural organisation of bacteria within a chronic wound. The aggregation of microorganisms into microcolonies within a matrix with very few planktonic cells has been observed within chronic wounds.[26] It is important to note that lack of correlation between the bacterial species identified using traditional culture techniques and PNA-FISH has been highlighted, with wound colonisation results showing >60% *Staphylococcus aureus* and <30% *Pseudomonas aeruginosa* using culture methods, and only 15% *Staphylococcus aureus* and 70% *Pseudomonas aeruginosa* using PNA-FISH.[26] This emphasises the importance of using not only traditional culture techniques, but also molecular methods, in order to achieve an accurate microbial identification and profiling of a wound. As yet, in clinical practice, the detection and diagnosis of a biofilm has not been achieved and microbial profiling usually relies on traditional culture methods.

Furthermore, *in vitro* studies have indeed shown antibiotic resistance in human chronic wound-derived mixed-species bacterial biofilms, indicating the potential resistant phenotype of chronic wound-derived bacteria *in vivo*.[27,29]

Fig 3 Detrimental attributes of necrotic tissue and slough in the wound bed



Biofilms and the host immune response

The presence of biofilms within the viable tissue of wounds does indeed elicit a host immune response. Fazli and colleagues demonstrated the presence of infiltrating neutrophils in chronic VLU biopsies and in addition determined a strong correlation between high neutrophil numbers and the presence of *Pseudomonas aeruginosa*, indicating that *Pseudomonas aeruginosa* biofilms may be one of the factors leading to a persistent inflammatory response.[28] However, *in vitro* research has shown that *Pseudomonas aeruginosa* uses microorganisms components of the hosts immune system to their advantage, more specifically, polymorphonuclear leucocytes (PMNs) that secrete neutrophil-derived polymers, DNA and actin, which provide *Pseudomonas aeruginosa* with a scaffold for biofilm formation.[29] Furthermore, invading bacteria can use mechanisms of immune evasion to avoid bacterial killing and successfully form biofilms. For instance, the EPS matrix of *Pseudomonas aeruginosa* biofilms has been shown to protect against interferon-γ-(IFN-γ)-mediated macrophage killing.[30] In addition, *Pseudomonas aeruginosa* can also secrete proteases such as elastase, which act as virulence factors that can inactivate components of the complement system.[31]

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Biofilms and wound healing

The presence of biofilms within chronic wounds is thought to be an important factor contributing to a delay in wound closure, as demonstrated by *in vitro* and *in vivo* studies. Kirker and colleagues demonstrated the deleterious effects of meticillin-resistant *Staphylococcus aureus* (MRSA) biofilms on dermal fibroblast and *Staphylococcus aureus* biofilms on epidermal keratinocyte viability and wound closure *in vitro*.[32,33] Furthermore, the presence of *Staphylococcus aureus* biofilms in a wounded New Zealand rabbit ear model, resulted in low-grade inflammation, decreased granulation tissue formation and reduced epithelialisation.[34]

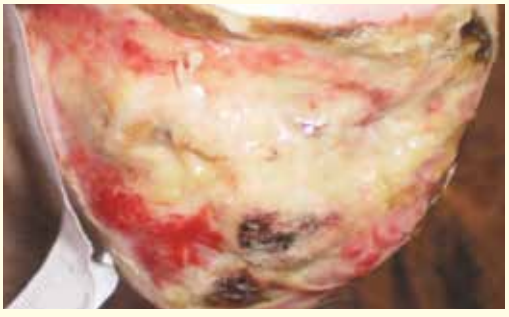
Necrotic tissue and slough

A key focus of this paper is the formation of necrotic tissue and slough within chronic wounds, which are problematic for clinicians (Fig 3). Indeed, biofilms have been shown to develop in viable tissue but evidence supporting the growth of biofilms in necrotic tissue and slough is limited. Harding and Enoch reported that microbial activity within the wound has a correlation to the generation, appearance and regeneration of slough, however they did not report the presence of biofilms within slough.[35]

Necrotic tissue

The tissue that is no longer viable within wounds and is generally observed to be black/dark brown in colour is referred to as necrotic tissue, however, the colour can vary between patients.[36] Necrotic tissue has been described as a fibrous mass of extracellular matrix components including fibronectin, collagen, fibrinogen, elastin and chondroitin sulphate (many of these components have been shown to provide excellent 'surfaces' for microbial attachment).[37] As necrotic tissue begins to dry out it becomes very hard and dry. Necrotic wounds fail to heal and the presence of necrotic tissue can conceal the true size and stage of the wound. Furthermore, as a biological entity it may prevent effective treatment from antimicrobial-incorporated dressings, by acting as a barrier to the release and penetration of antimicrobials.

Fig 4. Slough found within a chronic wound



Slough

Slough is generally pale yellow or yellow/brown in colour and is overall more loosely attached to the wound bed when compared with necrotic tissue, although on occasions it can be very firmly attached (characteristic of dry wounds) to the surrounding tissue (Fig 4).[38] Slough should not be confused with liquefactive necrosis, whereby usually hard and relatively dry necrotic tissue becomes softened and rehydrated. The presence of slough is considered a waste product of the host-immune response to effectively clear cellular debris. Slough is composed of microorganisms, serum proteins such as fibrin, albumin and immunoglobulin, white blood cells and matrix proteins including collagen amongst other components.[38] It potentially provides an ideal support for the attachment and proliferation of microbes and subsequent biofilm formation. Slough, like a biofilm, is not necessarily confluent over the surface but in the majority of cases it is very patchy. Although there are potential similarities to the composition of slough and biofilms, this has yet to be thoroughly researched.

Slough and biofilm: is slough a macroscopic biofilm?

With evidence to suggest that biofilms perpetuate the inflammatory response,[28] the subsequent increase in the production of slough may therefore provide microorganisms within the wound with a focal point of attachment. Consequently, slough may act as a reservoir for biofilms, leading to a hyper-inflammatory response and further production of slough. An early study in 1960 by Colebrook and colleagues demonstrated the ability of Gram-negative and Gram-positive bacteria to proliferate in blister fluid (exudate) and a slough preparation (taken from untreated full-thickness burn wounds).[39] This demonstrated the efficacy of exudate and slough to provide bacteria with the essential nutrients for growth. We propose that slough not only houses microorganisms, which leads to biofilm formation, but also that the slough itself is a macroscopic biofilm. A good example of a macroscopic (visible to the naked eye) biofilm is that of dental plaque, often referred to as slough, which can be clearly visible on the enamel (abiotic surface) after one day without brushing. Dental plaque is reported to be composed of a diverse microbial community embedded onto the surface of the tooth and encased in microbial-derived and salivary-derived polymers.[40] Dental plaque-associated microorganisms have been shown to lead to pathologies such as periodontitis, an inflammatory disease of the tissues supporting the teeth. One could argue that the recommendation of regular,

effective oral hygiene such as mouth-washing (cleansing/irrigating), tooth brushing, use of toothpaste and interdental flossing is not too dissimilar from the removal of slough from a wound, as both require the removal of this material to prevent inflammation of the surrounding tissues.

In relation to chronic wounds, Gethin and Cowman assessed the bacteriological changes in chronic VLUs during a four-week treatment period using either manuka honey or hydrogel wound dressings for the desloughing of the wound.[41] Subsequently, this randomised controlled trial showed a significant eradication of MRSA following desloughing using the manuka honey dressing. However, the authors did not relate MRSA or slough to the presence of a biofilm. Nevertheless, they stressed the importance of controlling infection using antimicrobial desloughing technologies. To date there is insufficient evidence to support the growth of biofilms and their microbial complexities within slough. Simple microbiological techniques and microscopy would confirm the presence of specific microorganisms and their architecture within slough, however more complex assays would be required to determine the presence of biofilms.[42] We are presently investigating the development of biofilms in our slough models.

Slough as an infection risk

The presence of slough, which may act as a macroscopic biofilm within chronic wounds, also has the potential for acting as a reservoir for microorganisms. With this in mind, it is important to consider the potential of slough to facilitate microbial dissemination. Therefore the risk of microbial attachment and proliferation in the underlying viable tissues of the wound bed is extremely high and may lead to the increased bioburden of the wound. However, the wound bed is not the only site of microbial colonisation within the wound, as wound dressings, wound-dressing fibres, wound exudate and necrotic tissue may also house a microbial biofilm (Fig 1). If slough does indeed act as a reservoir for microbial attachment and biofilm formation, the potential for the dissemination of microorganisms from the biofilm, is of great concern clinically. Microorganisms within a biofilm can detach from the biofilm, a process known as 'dispersal', whereby microorganisms within the relatively slow-growing environment of the biofilm become highly motile.[43] However, the mechanical shearing-off of part of the biofilm can further increase the risk of dissemination. An excellent example of the clinical repercussions of microbial dissemination is that of medical device-related infections, whereby the contamination of indwelling medical devices (staphylococcal species) can lead to a systemic infection.[44]

Management of necrotic tissue and slough

Debridement

Debridement is a method used for the removal of non-viable tissue in order to clean and prepare the wound bed, and is an important component in wound management.[45,46] For practitioners to undertake this, it is essential to have a clear understanding of the techniques employed, associated advantages and disadvantages, and reasoning for use. Debridement in isolation of other techniques and methods will not achieve the ultimate goal of wound healing. It would not be feasible to assume that any single debridement technique alone would successfully remove 100% of all non-viable tissue. Like any wound care application, debridement must be used as a structured wound management plan with appropriate milestones to be achieved for the patient. In clinical situations where wounds present with excessive slough, it appears that the production of slough post-debridement is a common occurrence. The reasons for this are unknown, however, it is possible that host responses towards the persistent presence of a biofilm results in the continuous production of slough. Consequently an on-going desloughing procedure needs to be maintained.

As mentioned previously, we theorise that necrotic tissue and slough support the growth of microorganisms and therefore the development of biofilms. Consequently the presence of necrotic tissue and slough can act as barriers to wound healing. The wounds with necrotic tissue and slough often are wounds that are in a state where complex microbiological processes are continuing. There will be a high level of anaerobic bacteria, which indicates complexity of the microbial community, and these generate malodour in the wound. Furthermore, as the microbial community increases antimicrobial efficacy decreases. Therefore effective debridement and desloughing of a wound can help this, significantly. In addition, by reducing the wounds whole microbial bioburden (found in the wound bed, on necrotic tissue, on slough, on the dressing itself and in the wound exudate) (Fig 1) this will help to reduce the hyper-inflammatory responses, which in turn will help the development of granulation tissue. The methods of debridement that are routinely used in wound management and associated risks are described below and in Table 2.

Autolytic debridement

Autolytic debridement occurs when the body uses its own enzymes to break down, soften and liquefy dead and devitalised tissue. This must occur within a moist environment which can be achieved using an array of different wound dressings that support the autolytic debridement. These include hydrogels, hydrocolloids and alginates.[47] Often hydrogels are used for autolytic debridement and are divided into those

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Table 2. Methods of debridement

Debridement method	Method	Advantages	Disadvantages	Risk	Ref.
Autolytic debridement	Not commonly used but products that can facilitate this include hydrocolloids, hydrogels, honey etc. Encourages own patient's enzymes and exudate to liquefy tissue, eschar, slough.	• Useful in the prevention of devitalised tissue and slough • Maintenance of the wound • Does not require specialists • No pain	• Not commonly used. Slow process and can lead to maceration and increases risk of infection	• Important to monitor the moisture levels to avoid maceration and further complications. Increases maceration and Infection	47
Larvae (maggot) therapy	Uses green blowfly, which generate enzymes to breakdown necrotic tissue	• Quick treatment times • Selective to necrotic tissue • Does not require a specialist	• Similar in mode of action to autolytic debridement–enzymes • Comparatively costly • Cannot be used for all patients–adherence		64 65 66 55
Hydrosurgical debridement	Removes necrotic and devitalised tissue using a high-pressure saline cutting technology	• Precisely target the area for debridement. Considered to remove biofilm. Documented to reduce procedure time. • Relatively short treatment	• Requires specialised personnel • Costly	• Potential infection risk-aerosolisation	67 68
Mechanical debridement	Wet-to-dry gauze (dries and adheres)	• Ease of use • Does not require a specialist	• Painful for the patient • Not selective • Requires lots of dressings	• Pain on removal • Can remove healthy granulating tissue	58 56
Sharp debridement	A scalpel or scissors is used to remove the devitalised and necrotic tissue	• Quick • Selective	• Requires a competent practitioner; not appropriate for all • Can be done at bedside	• Risk of damaging nerves, blood vessels and tendons	
Surgical debridement	Possible resection of viable tissue	• Very selective • Maintenance debridement	• Specialised equipment • Cost • Requires skilled personnel • Must be carried out in the theatre.	• Patients refuse procedure due to pain	
Ultrasonic debridement	Debrides using low-frequency ultrasound. This can be direct or indirect contact	• Considered painless for the removal of devitalised tissue. Shown to reduce microbial bioburden • Could be selective • Maintenance debridement	• Expensive • High costs for continued usage • Requires long setup times, sterilisation • Requires specialised/ competent personnel	• Potential for the mist of saline and blood products to be aerosolised.	61
Mechanical desloughing	Specifically removes slough within the wound	• Ease of use • Quick • No pain • Key to maintenance desloughing	• Not considered to have any disadvantages to date	• No risk or very low risk	

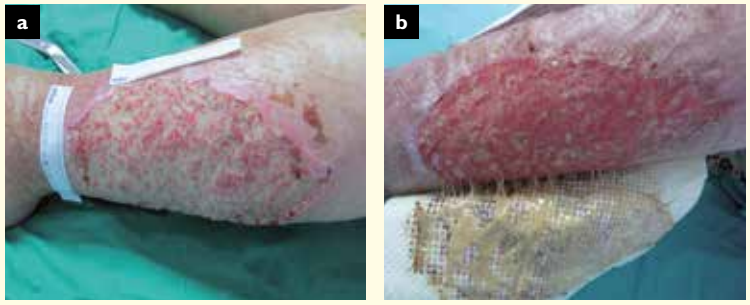
that donate fluid and those that absorb exudate. It is considered a slower process than a number of other techniques.[48] When using this technique some markers are often employed to establish progress is being made within 96 hours. For example, in black necrotic wounds a colour change can indicate some form of progress, when the colour goes from black to grey/brown and then goes yellow (however this may be due to the rehydration of the necrotic tissue), or if separation occurs at the wound margins.[49]

Hydrosurgical

Hydrosurgery combines physical and surgical debridement. It involves the use of a high-pressure

jet of sterile saline. This creates a Venturi effect (the movement of fluid through a constricted opening, resulting in a decrease in pressure and a suction effect) that enables the removal of necrotic tissue.[50] It is considered to cost a lot for the equipment and requires a specialist to carry out the procedure. Also there is a risk of aerosolisation of blood products and microorganisms, suggesting an infection risk.[51]

Fig 5. An example of desloughing a wound by mechanical desloughing



Surgical and sharp debridement

Surgical debridement is performed in the operating theatre and results in a bleeding wound bed. Sharp debridement of non-viable tissue can be performed at the patient's bedside using a scalpel or surgical scissors. The use of sharp debridement may help biofilm management. A clinical study involving three patients with biofilm-infected wounds showed a reduced recalcitrance of the wound biofilm to the antibiotic gentamicin following sharp debridement of the wound.[52] While the methods of surgical and sharp debridement are the faster methods for debriding they require skilled personnel to perform the procedure. However, they are necessary in high-risk patients that are at risk of sepsis or cellulitis.[53]

Larvae (maggot) therapy

Larvae therapy, while a 400-year old method, is still practiced routinely today using sterile larvae called maggots (derived from *Lucilia sericata* species, or the green blow fly).[45] Debridement using these larvae relies on the secretion of enzymes into the wound, which effectively leads to the enzymatic breakdown of necrotic tissue. The process involves adding the larvae directly to the wound or within a bio-bag and then leaving the wound for approximately 3 days. However, there is evidence to show that the use of larvae therapy, although faster in debridement after one week (when compared with autolytic debridement), bears no significant benefits when compared with other conventional methods such as the use of wound dressings.[54] There is, however, evidence to suggest the effectiveness of the larvae to degrade DNA from not only slough and eschar, but also *Pseudomonas aeruginosa* biofilms.[55]

Mechanical debridement

Traditional mechanical debridement uses wet-to-dry gauze dressings or dry gauze dressings. However, the wet-to-dry gauze approach is considered painful for the patient and is not recommended or practised in many hospitals and clinics. Other technologies can be employed for mechanical debridement, that are easy to use and cause little to no patient discomfort.[56]

Ultrasonic debridement

Ultrasound is a relatively new procedure that is being used to debride wounds. It was developed in the early 1950s for use in dentistry and was used for reducing levels of tissue and dissecting bone. There is a growing amount of evidence that supports the use of ultrasonic debridement in VLUs and for the breakdown and removal of biofilms. The procedure is indicated for wounds such as PUs, burns, venous stasis wounds and DFUs.[57] There is however, a high-risk associated with ultrasonic debridement. For example, it has been

Fig 6. Suggested methods of debridement for patients with slough and necrotic tissue

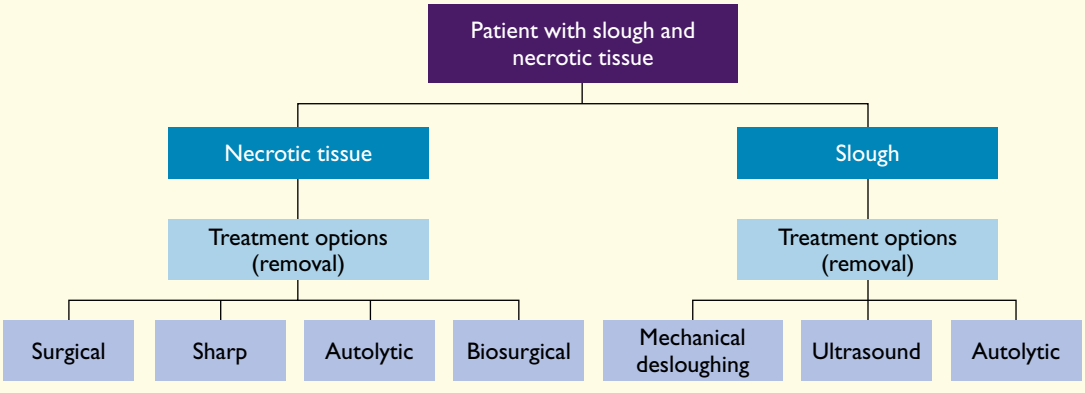


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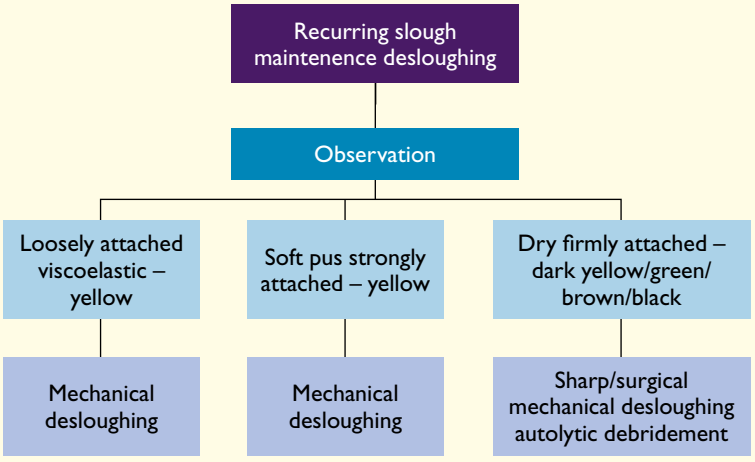
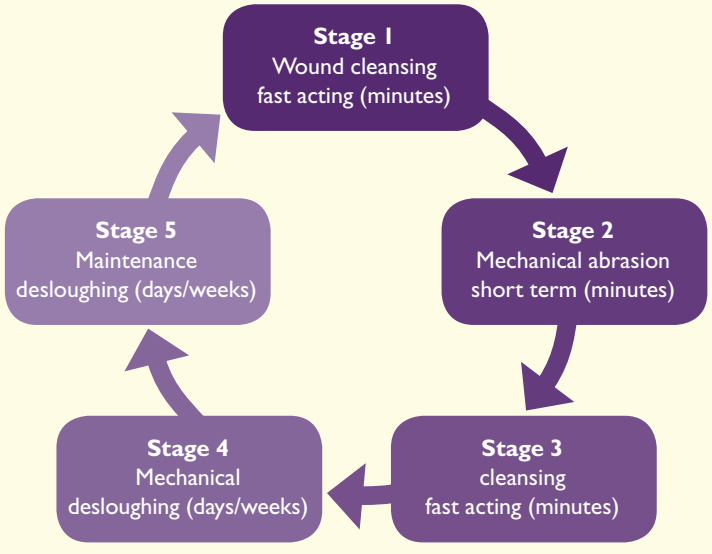
Fig 7. Pathway for the management of recurrent slough



reported that there is a potential for aerosolisation of blood products and microorganisms to occur.[58] Consequently appropriate protective clothing is often advised. The frequency for effective ultrasonic debridement is 20–40 kHz with the mechanism of action reported to have been achieved by two methods, acoustic streaming and cavitation. The acoustic streaming is referred to as the force (mechanical) of the saline, which comes from the ultrasonic probe/tip of the device. Cavitation is the other mode of action that consists of the fluid (saline) released from the sonicating device, forming bubbles of vapour that develop and then break down near the tissue. There are two different sonication methods available, contact and non-contact.[59] The effect of heat generation and therefore thermal concerns are low considering the low frequencies employed. Numerous studies have been conducted on the use of ultrasound to debride wounds demonstrating its effectiveness.[60] Ennis and co-workers coordinated a randomised, double-blind, controlled, multicentre study into the effectiveness of ultrasound therapy in patients with recalcitrant DFUs and subsequently concluded a significant proportion of completely healed cases following 12 weeks treatment compared with the control group, which were treated with a 'sham device'.[61] There are a number of different ultrasonic devices available that are being used specifically for debridement, for example, the Sonoca line (Söring), the SonicOne O.R. system (Misonix) and the Qoustic Wound Therapy System (Arobella Medical) and the non-contact low-frequency ultrasound (MIST Therapy System, Celleration, Inc., Eden Prairie, MN). Ultrasonic debridement has also been reported to be effective for wounds with chronic venous insufficiency and burn wounds.[59,62]

Fig 8. The procedures required to prevent and treat recurrent slough–maintenance desloughing



Risks of debridement

All methods employed for debridement have both their advantages and disadvantages. The method of debridement that is employed is generally decided based on clinical judgement, expertise of the health-care professional, ease of use and importantly, the adherence and accessibility of the patient. A report by Gray and colleagues agreed that when deciding on the debridement method to be used, the decision should be based on the clinical need and not on the skills of the clinician.[58] Therefore there should be particular focus on the method of debridement that is the most effective for the patient as an individual. Furthermore, the choice of debridement technique will also depend on the amount of necrotic tissue, the anatomical site of the wound and the accessibility for debridement tools. The methods used should form part of the overall wound management of that specific wound and also of that specific patient. Importantly, the patient's underlying pathophysiology and comorbidities need to be taken into consideration.

Certain debridement methods that are considered as high-risk, such as surgical debridement, are not necessary for the removal of slough. Instead, lower-risk alternatives can be used and are more patient friendly. This method can be simply referred to as 'desloughing'.

Desloughing

Desloughing is a process that is used to separate slough from the underlying granulation tissue of the wound. Desloughing is a term associated with the removal of slough using wound dressings. In fact, there is much controversy over the differenti-

ation between desloughing and debridement and these terms are interchanged within the literature, causing much confusion. A recent debate took place at the European Wound Management Association (EWMA) meeting, whereby the differences between desloughing and debridement were discussed.[63] It was agreed that slough shares very different characteristics from necrotic tissue.

Although debridement can encapsulate the removal of both necrotic tissue and slough, we propose that the use of 'desloughing' as a gerund may help health-care specialists to decide on the appropriate debridement method. More specifically, the desloughing of slough would involve less aggressive methods of debridement, such as mechanical desloughing (Fig 5, 6 and 7).

Desloughing of recurrent slough

Fig 8 highlights a procedure that may be appropriate for recurrent slough. Stage one requires the use of an antimicrobial (fast-acting) wound cleanser to remove debris and aid in reducing microbial cell numbers and breaking down slough. However, for the antimicrobial to be effective it must be left in the wound for a long time period (at least 5–10 minutes, but this will depend on the antimicrobial). The wound could then be mechanically abraded (this process takes only minutes) to quickly remove the more loosely attached slough (similar to the concept of brushing teeth). It is important then that the dislodged slough and microorganisms are then exposed to the antimicrobial wound cleanser again. After this long-term mechanical desloughing using an appropriate a wound dressing that selectively removes slough could be added to prevent and manage slough build up. It is important that the wound is monitored constantly and wound dressings are removed regularly i.e. maintenance desloughing.

Conclusion

To achieve an environment for effective wound healing debridement of non-viable tissue including necrotic tissue and slough must be carried out. Necrotic tissue and slough represent barriers to wound healing. However, slough is chemically, physically and biologically different from necrotic tissue and may display inherent characteristics similar to that of a biofilm, as seen in dental plaque. The term debridement can be used to encompass the various methods used to remove necrotic tissue and slough, however, slough and necrotic tissue are very different entities and the methods adopted for their removal should differ. The methods necessary to remove necrotic tissue from a wound come with significant risks. The removal of slough should be described as desloughing and these methods should involve lower-risk and less-aggressive forms of debridement such as mechanical desloughing. There are a number of different desloughing technologies available that appear to represent minimal risk.

In this paper, we strongly propose that slough is an ideal environment for biofilm growth and can act as a reservoir for biofilms, therefore by desloughing you are effectively removing two significant barriers to wound healing. Both biofilms and slough have been reported to perpetuate inflammation in chronic wounds, which can lead to a delay in wound closure. Thus the management of slough within chronic wounds should be addressed as an integral part of wound care, which will help reduce the microbial bioburden, the presence of biofilms and help reduce the inflammatory response. ■

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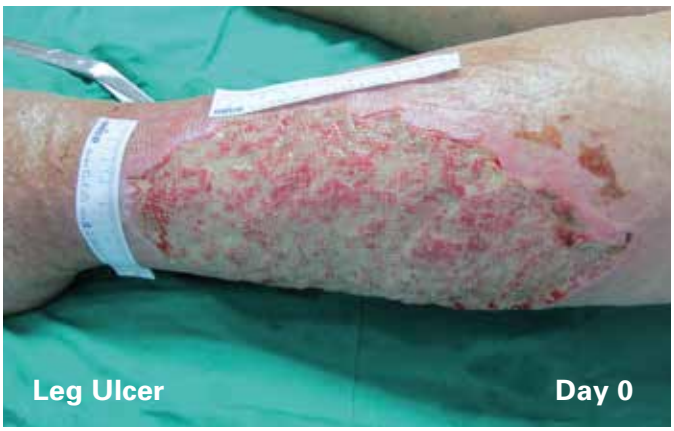

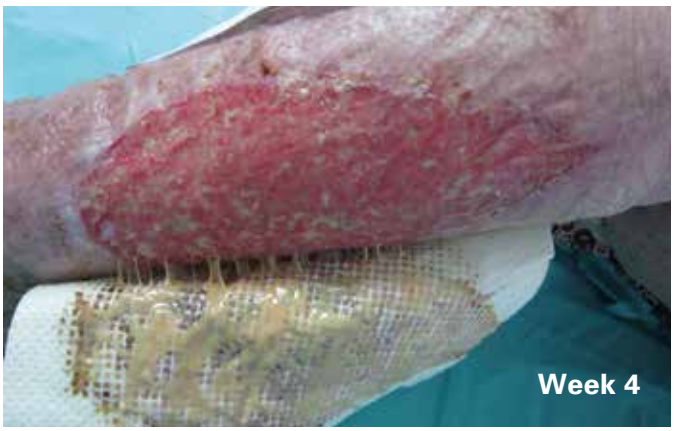

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