***Review article***

# Role of infection in wound healing

*Malik Asif Hussain[1], Flavia Huygens[2]*

**Abstract:**
Chronic wounds, particularly infected wounds are clinically very important due to their significant impact on health budgets as well as patients` health worldwide. Patients with diabetes mellitus, vascular diseases especially peripheral vascular disease and pressure ulcers are major categories of patients presenting with chronic wounds. It is known that there are multiple factors determining chronic wound prognosis. The presence of multiple types of pathogenic bacteria, with specific virulence and adherent (biofilm) properties, contribute a significant role to the development of chronic wounds. This review article is based on the research project entitled" An investigation of the impact of bacterial diversity, pathogenic determinants and biofilms on chronic wounds". The research findings have been published in form of research papers as well as conference posters. The aim of this article is to highlight various important aspects of bacterial impact on wound healing.

**Keywords:** Chronic wounds; wound healing; wound infections.

*Bangladesh Journal of Medical Science Vol. 19 No. 04 October'20. Page : 598-602*
*DOI: https://doi.org/10.3329/bjms.v19i4.46612*

## Introduction:

Chronic infections are clinically very important and huge amount of health budgets worldwide are consumed on managing them[1-3] along with economic burden of diabetes mellitus[4]. These infections usually result after damage or defects in the primary defence mechanisms[5]. One of the major clinical challenges is non-healing wounds like diabetic foot, vascular and pressure ulcers[6,7]. Approximately 17 million people develop chronic wound infections and 550,000 die from these infections worldwide every year[8]. Chronic wounds are estimated to develop in 1–2% of the population of developed countries during their lifetime[9]. Along with local causes various systemic diseases such as hypertension[10] are linked with chronic wounds.

When superficial skin layer (epidermis) is lost along with dermis or deeper layers, we call this a wound or an ulcer. A wound has been described as "a physical break in epithelium integrity and the subsequent host response to repair this break"[11]. A wound lasting for 6 weeks or over is a chronic wound[12]. Chronic wounds are of three categories[12,13]; healable, non-healable and maintenance. The first type heals with proper treatment. Similarly, maintenance wounds could heal and are actually healable wounds. They stay for a long time due to limited resources and care. Once treated properly they progress to healing. Non-healable wounds have extensive tissue damage or damage to blood supply such that it cannot be treated/corrected.

### *Bioburden:*

The actual bacterial load or bioburden, defined as the "metabolic load imposed by bacteria in the wound bed" plays an important role in chronic wounds[14]. The bioburden not only includes total bacterial numbers in the wound but also their metabolic activities, nutrient consumption and production of toxic substances.

1. Malik Asif Hussain, College of Medicine, University of Hail, KSA. School of Biomedical Sciences, Queensland University of Technology, Brisbane, QLD Australia, Wound Management Innovation Cooperative Research Centre, Brisbane, QLD Australia.
2. Flavia Huygens, School of Biomedical Sciences, Queensland University of Technology, Brisbane, QLD Australia, Wound Management Innovation Cooperative Research Centre, Brisbane, QLD Australia.

**Correspondence to:** Dr. Malik Asif Hussain, College of Medicine, University of Hail, KSA; School of Biomedical Sciences, Queensland University of Technology, Brisbane, QLD Australia and Wound Management Innovation Cooperative Research Centre, Brisbane, QLD Australia. Email: hussain_amc2010@yahoo.com

Using a scanning microscopy, observations have been made that reveal that the bioburden is higher in cases of chronic wounds compared to acute wounds. Bioburden, specific organisms and immune response are the main factors contributing to the development of chronic wounds[15,16].

Researchers claim that the presence of a bacterial load of $>10^5$ CFU as a cut off value to determine whether a wound will become infected or not[17]. There is another view which claims that the presence of more virulent organisms can result in infections even if bacterial load is less than $10^5$ Colony Forming Units (CFU)[16]. A recent study has reported that only higher bacterial load ($> 10^5$CFUs/g) is associated with poor healing of wounds, even in absence of infection signs[18].

***Bacterial growth and wound healing:***

Skin acts as a barrier against infections. So, if this protection is lost as in case of wounds bacteria can colonize and cause damage mainly by interfering with normal protective mechanisms like antibacterial secretions[11,19]. Bacteria grow over all chronic wounds[20]. For this we never consider a wound surface sterile[21]. Gontcharova and co-workers found a notable difference in the number of opportunistic pathogens between normal skin (1.54%) and wounds (20%), indicating that the skin harbours the majority of bacteria which are usually harmless[1].

Colonization of a wound bed is characterized by surface growth of bacteria, however, there is no noticeable immune response while critical colonization or infection occurs when bacterial number increases and interferes with the healing process[22]. This difference between colonization and infection should be understood by health care professionals[23]. This is because treatment for infection is recommended while colonization does not need to be treated[24]. It is not clinically possible to assess difference amongst these three (colonization, contamination, infection).

Bacterial infection plays a major role in delaying wound healing by enhancing inflammatory response and tissue damage[6,25,26] and wound healing is enhanced if surface dead tissue is removed as such scabs which act as a reservoir for the majority of microorganisms[27].

Factors affecting the wound microbiome determines the fate of a wound as both acute and chronic wounds are colonized with bacteria, however, the outcomes are different for both types of wounds[11]. Infection control is regarded as an important component of wound bed preparation (WBP)[12,13].

Wound healing is a highly complex and organized process involving many cellular components. These phases are haemostasis, inflammation, proliferation and remodelling[28]. Proposed mechanisms responsible for improper healinginclude a prolonged inflammatory response, the presence of biofilms[29], failures of skin to re-epithelise[30] and an imbalance of micro-molecules[31,32].

The inflammatory response of chronic wound infections is different which results in less prominent symptoms compared to acute infections. This response mainly involves Immunoglobulin G (IgG) antibodies and polymorphic nuclear neutrophils (PMNs) which continuously, but in a controlled manner, keep migrating to the infection site [33]. Keratinocytes play an active part in interactions of the innate immune system[34]. The presence of neutrophils has been reported to slow migration of keratinocytes[35]. Microbial growth, such as *S. Aureus* biofilms, activates apoptosis in keratinocytes[36].

Similarly, bacterial biofilm is now being considered as one of the main factors influencing wound healing[37]. It effects healing processing by various mechanisms particularly by providing a safe environment for bacterial growth[38]. Bacteria growing in form of biofilm have been reported to have ten times higher survival rate compared to their planktonic growth[39].

In a wound bed, there is abundance of nutrients as well as protection against antimicrobials and immune system[40,41]. Similarly, low blood flow and lack of oxygen provides extra protection for these bacteria[42]. It is also being investigated whether the presence of biofilms can reduce surface penetration of oxygen[27,43].

**Prevention, problems and future directions:**

There are many unanswered questions related to bacterial involvement in wound healing. For example, what exactly determines whether a wound will become chronic? What is the exact source of bacteria in various wounds? Why and how do multiple bacterial species survive together? Which species are more pathogenic? Similarly, it is still unclear which treatment strategy is the best[30,44]. More specifically, it is yet to be determined how the biofilm load interferes with the healing process[30]. It is still unclear why and how exactly microorganisms adapt their growth form biofilms [45]. Although it is claimed that biofilm interferes with re-epithelialisation, no model system has proved it yet[46].

Controlling wound care infection is very important as these infection can not only result in systemic infections but can also affect systemic disease[47]. Clinically there are several challenges in avoiding infections and infection transfer. Microbes can

colonize patients and health care workers[48]. They can also grow in substances such as normal saline or some disinfectants[23]. These could act as reservoir for infection spread or cross contaminations. This means it is necessary for health care workers to understand that there is a difference amongst clean, sterile and dirty objects[49].

There is no agreed criterion to determine and differentiate deep infection and critical colonization. If we can diagnose wound infection based on clinical examination, this would be a desirable clinical goal but no single clinical sign has been reported to be able to tell us the difference between superficial colonization or deeper invasion and infection[20]. Sibbaldet al (2006) have presented NERDS (indicating superficial growth) and STONEES (indicating deeper moderate or heavy infection) categories of combination of signs and symptom to differentiate between superficial bacterial colonization and deeper invasion and infection[50]. We need to verify and further develop such criteria.

Currently, there is no 'gold standard' for determining the correlation between bacterial load/bioburden and wound chronicity. It is therefore very important to develop more robust and accurate methods, such as molecular methods, to quantify bacterial load and diversity of chronic wounds. Once this has been developed and validated, we can definitely improve wound treatment plans and healing outcomes51. Similarly, sample collection from wound surface can affect the results. The samples collecting surface materials for bacterial growth do not represent bacterial population invading deep in the wound tissue[52].

The role of the skin microbiome or normal microbial flora in the healing process is still not very well known, though advanced techniques are providing a large amount of data indicating their beneficial role in maintaining normal skin health. It is important to increase our knowledge of the skin microbiometo understand the microbial composition of normal skin as well as the wound microbial composition. This will allow us to set criteria, develop methods to prevent, diagnose and effectively treat such infections[19]. The exact role of microbes in acute and chronic infections requires further research and understanding. It is not fully known which bacterial species specifically contributes to chronic wound and biofilm formation. Acute bacterial infections must be diagnosed and treated quickly as they involve planktonic bacteria but if left untreated, could result in chronic infection[8]. Wound management and treatment will improve by revealing details regarding wound microbial flora[53]. Culture based techniques are not very useful for diagnosis of infection involving biofilm. This also holds true for the identification of many microbes which require special laboratory culture requirements, for example, anaerobes. Culture based methods have limitations, therefore, molecular methods should be developed [16,54]. Diagnosis and confirmation of biofilm presence in chronic wounds is very important and requires further investigation[38,55].

To our understanding, the determination of bacterial types, their virulence and biofilm markers could increase our understanding of the wound environment and this information could be used to propose more effective diagnostic, prophylactic and treatment options.

**Ethical clearance:** No ethical clearance was required for this review article.

***Acknowledgments& Conflicts of Interest:*** There is no specific funding resource for this review paper, but we acknowledge the support of the Wound Management Innovation Co-operative Research Centre (WMICRC) and Queensland University of technology (QUT) for running the original project on chronic wound research. All authors confirm that they have no conflict of interest to declare.

**Author's contribution:** All authors have the contributions of writing and editing of manuscript

## References:


1. Gontcharova, V., et al., *A comparison of bacterial composition in diabetic ulcers and contralateral intact skin. Open Microbiol J*, 2010. **4**: p. 8-19.
2. Scali, C. and B. Kunimoto, *An update on chronic wounds and the role of biofilms. Journal of cutaneous medicine and surgery*, 2012. **17**(6): p. 371-376.
3. James, G.A., et al., *Biofilms in chronic wounds.* Wound Repair Regen, 2008. **16**(1): p. 37-44.
4. Afroz, A., et al., *Cost-of-illness and its determinants for type 2 diabetes mellitus in bangladesh. Bangladesh Journal of Medical Science,* 2019. **18**(3): p. 501-507.
5. Chen, L. and Y.M. Wen, *The role of bacterial biofilm in persistent infections and control strategies. Int J Oral Sci*, 2011. **3**(2): p. 66-73.
6. Ngo, Q.D., K. Vickery, and A.K. Deva, *The effect of topical negative pressure on wound biofilms using an in vitro wound model. Wound Repair Regen,* 2012. **20**(1): p. 83-90.
7. Lu, J., et al., *Manuka-type honeys can eradicate biofilms produced by Staphylococcus aureus strains with different biofilm-forming abilities.* PeerJ, 2014. **2**: p. e326.
8. Wolcott, R. and S. Dowd, *The role of biofilms: are we hitting the right target?* Plast Reconstr Surg, 2011. **127 Suppl 1**: p. 28S-35S.
9. Sen, C.K., et al., *Human skin wounds: a major and snowballing threat to public health and the economy. Wound Repair Regen, 20*09. **17**(6): p. 763-71.
10. Igor, D., et al., *Hypertensive ulcer of lower extremity (Martorell's syndrome): clinical case with the treatment improvement. Bangladesh Journal of Medical Science*, 2017. **16**(2): p. 325-328.
11. Scales, B.S. and G.B. Huffnagle, *The microbiome in wound repair and tissue fibrosis. J Pathol,* 2013. **229**(2): p. 323-31.
12. Sibbald, R.G., et al., *Special considerations in wound bed preparation 2011: an update: wound bed preparation. Wound Healing Southern Africa,* 2011. **4**(2): p. 55-72.
13. Sibbald, R.G., et al., *Optimizing the Moisture Management Tightrope with Wound Bed Preparation 2015©.* Advances in skin & wound care, 2015. **28**(10): p. 466-476.
14. Warriner, R. and R. Burrell, *Infection and the chronic wound: a focus on silver. Adv Skin Wound Care,* 2005. **18 Suppl 1**: p. 2-12.
15. Howell-Jones, R.S., et al., *A review of the microbiology, antibiotic usage and resistance in chronic skin wounds. J Antimicrob Chemother*, 2005. **55**(2): p. 143-9.
16. Bowler, P.G., B.I. Duerden, and D.G. Armstrong, *Wound microbiology and associated approaches to wound management. Clin Microbiol Rev*, 2001. **14**(2): p. 244-69.
17. Robson, M.C., et al., *Maintenance of wound bacterial balance. Am J Surg,* 1999. **178**(5): p. 399-402.
18. Brown, S., *Clinical antimicrobial photodynamic therapy: phase II studies in chronic wounds. J Natl Compr Canc Netw,* 2012. **10 Suppl 2**: p. S80-3.
19. Percival, S.L., et al., *Microbiology of the skin and the role of biofilms in infection. Int Wound J,* 2012. **9**(1): p. 14-32.
20. Woo, K.Y. and R.G. Sibbald, *A cross-sectional validation study of using NERDS and STONEES to assess bacterial burden.* Ostomy/wound management, 2009. **55**(8): p. 40.
21. Olmsted, R., *APIC Infection Control and Applied Epidemiology: Principles and Practice.:Association for Professionals in Infection Control and Epidemiology, Inc.* 1996, Washington, DC.
22. Kingsley, A., *The wound infection continuum and its application to clinical practice.* Ostomy/wound management, 2003. **49**(7A Suppl): p. 1-7.
23. Crow, S. and P.J. Thompson, *Infection control perspectives on wound care. In: Krasner DL, Rodeheaver GT, Sibbald RG, eds. Chronic Wound Care:A Clinical Source Book for Healthcare Professionals.* . 4th ed. 2007, Malvern, Pa: HMP Communications. 323- 330.
24. Bergstrom, N., *Treatment of pressure ulcers*. 1994: DIANE Publishing.
25. Trial, C., D. Morquin, and L. Téot, *[Diagnosis and treatment of chronic wound infections]*. Soins; la revue de reference infirmiere, 2013(776): p. 11-15.
26. Gawande, P.V., K.P. Leung, and S. Madhyastha, *Antibiofilm and Antimicrobial Efficacy of DispersinB®-KSL-W Peptide-Based Wound Gel Against Chronic Wound Infection Associated Bacteria. Current microbiology,* 2014: p. 1-7.
27. Zhao, G., et al., *Time course study of delayed wound healing in a biofilm-challenged diabetic mouse model. Wound Repair Regen,* 2012. **20**(3): p. 342-52.
28. Guo, S. and L.A. Dipietro, *Factors affecting wound healing.* J Dent Res, 2010. **89**(3): p. 219-29.
29. Stephens, P., et al., *Anaerobic cocci populating the deep tissues of chronic wounds impair cellular wound healing responses in vitro. Br J Dermatol,* 2003. **148**(3): p. 456-66.
30. Gurjala, A.N., et al., *Development of a novel, highly quantitative in vivo model for the study of biofilm-impaired cutaneous wound healing. Wound Repair Regen,* 2011. **19**(3): p. 400-10.
31. Menke, N.B., et al., *Impaired wound healing.* Clin Dermatol, 2007. **25**(1): p. 19-25.
32. Edwards, R. and K.G. Harding, *Bacteria and wound healing. Curr Opin Infect Dis*, 2004. **17**(2): p. 91-6.
33. Artini, M., et al., *Staphylococcal IgM enzyme-linked immunosorbent assay for diagnosis of periprosthetic*

*joint infections. J Clin Microbiol,* 2011. **49**(1): p. 423-5.
34. Nestle, F.O., et al., *Skin immune sentinels in health and disease. Nat Rev Immunol,* 2009. **9**(10): p. 679-91.
35. Fazli, M., et al., *Quantitative analysis of the cellular inflammatory response against biofilm bacteria in chronic wounds. Wound Repair Regen,* 2011. **19**(3): p. 387-91.
36. Kirker, K.R., et al., *Differential effects of planktonic and biofilm MRSA on human fibroblasts. Wound Repair Regen, 2*012. **20**(2): p. 253-61.
37. Dowsett, C., *Biofilms: A practice-based approach to identification and treatment.* Wounds UK, 2013. **9**(2).
38. Bowler, P., *A clinical algorithm for wound biofilm identification.* Journal of wound care, 2014. **23**(3).
39. Spiliopoulou, A.I., et al., *Bacterial adhesion, intracellular survival and cytokine induction upon stimulation of mononuclear cells with planktonic or biofilm phase Staphylococcus epidermidis.* FEMS Microbiol Lett, 2012. **330**(1): p. 56-65.
40. Wolcott, R.D., D.D. Rhoads, and S.E. Dowd, *Biofilms and chronic wound inflammation. J Wound Care,* 2008. **17**(8): p. 333-41.
41. Black, C.E. and J.W. Costerton, *Current concepts regarding the effect of wound microbial ecology and biofilms on wound healing. Surgical Clinics of North America*, 2010. **90**(6): p. 1147-1160.
42. Falanga, V., *The chronic wound: impaired healing and solutions in the context of wound bed preparation. Blood Cells Mol Dis, 2*004. **32**(1): p. 88-94.
43. Mathieu, D. and R. Mani, *A review of the clinical significance of tissue hypoxia measurements in lower extremity wound management. Int J Low Extrem Wounds,* 2007. **6**(4): p. 273-83.
44. Gurjala, N., et al., *Animal models of biofilm-infected wound healing. Advances in Wound Care*, 2010. **1**: p. 305-310.
45. Bjarnsholt, T., et al., *Why chronic wounds will not heal: a novel hypothesis. Wound Repair Regen*, 2008. **16**(1): p. 2-10.
46. Schierle, C.F., et al., *Staphylococcal biofilms impair wound healing by delaying reepithelialization in a murine cutaneous wound model. Wound Repair and Regeneration,* 2009. **17**(3): p. 354-359.
47. Barber, L.A., *Clean technique or sterile technique? Let's take a moment to think. Journal of Wound Ostomy & Continence Nursing*, 2002. **29**(1): p. 29-32.
48. Sheff, B., *VRE & MRSA: Putting bad bugs out of business.* Nursing2014, 1998. **28**(3): p. 40-45.
49. Chisholm, D.F., *Asepsis, the Right Touch: Something Old is Now New. Journal of Gerontological Nursing*, 1990. **16**(10): p. 42-43.
50. Sibbald, R.G., K. Woo, and E.A. Ayello, *Increased bacterial burden and infection: the story of NERDS and STONES. Advances in skin & wound care,* 2006. **19**(8): p. 447-461.
51. Grice, E.A. and J.A. Segre, *Interaction of the microbiome with the innate immune response in chronic wounds. Adv Exp Med Biol*, 2012. **946**: p. 55-68.
52. Williams, D., J.R. Hilton, and K.G. Harding, *Diagnosing foot infection in diabetes. Clinical Infectious Diseases,* 2004. **39**(Supplement 2): p. S83-S86.
53. Murphy, E.C. and I.M. Frick, *Gram-positive anaerobic cocci--commensals and opportunistic pathogens. FEMS Microbiol Rev,* 2013. **37**(4): p. 520-53.
54. Davies, C.E., et al., *Use of molecular techniques to study microbial diversity in the skin: chronic wounds reevaluated. Wound repair and regeneration,* 2001. **9**(5): p. 332-340.
55. Hess, C.T. and R.S. Kirsner, *Understanding the Presence of Biofilms in Wound Healing: Opportunities for Intervention.* 2012.