Deficits in molecular, physical and biological parameters of healing in the diabetic foot: a literature review

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ABSTRACT

Background Diabetic foot ulcers (DFU) are complex, multifactorial and often complicated by delayed, impaired and uncoordinated wound healing. DFU are associated with devastating outcomes including infection, amputation and premature death.

Aim The aim of this narrative literature review is to obtain a broad perspective on the pathophysiological mechanisms that contribute to delayed and impaired healing in the diabetic foot.

Methods We undertook a review of the literature to critique and synthesise the evidence for pathophysiological factors that contribute to delayed and impaired healing in the diabetic foot.

Findings It is evident from the literature that molecular mechanisms that give rise to impaired inflammation will impact upon healing, whilst physical parameters such as tissue hypoxia, pressure foot-loading, wound PH, temperature and biofilm can all contribute to delayed healing in the diabetic foot.

Conclusions An understanding of the pathophysiology of impaired healing and a focus on controlling these disturbances can facilitate successful healing. To enhance the management of foot disease in diabetic patients, primary care professionals must be made aware of the significance of early referral to a specialised unit. When DFU do not heal adequately after 4 weeks of standard treatment, the underlying pathology should be re-evaluated, and the need for advanced therapy should be considered.

Implications for clinical practice It is important that clinicians involved in treating DFU have an understanding of the pathophysiological mechanisms that cause delayed and impaired healing in the diabetic foot. When a DFU fails to respond to standard care within a 4-week period, the pathophysiology of the wound should be re-evaluated and advanced therapies should be considered.

Keywords diabetes, diabetic foot ulcer, impaired wound healing, diabetic foot, wound healing

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KEY MESSAGES

- Diabetic foot disease is a serious complication of diabetes that is associated with devastating outcomes including diabetic foot ulceration, amputation and premature death.
- Patients with diabetes exhibit delayed, impaired and uncoordinated wound healing due to various pathophysiological factors including impaired molecular mechanisms and abnormal physical parameters.
- It is evident that uncontrolled factors such as inflammation, biofilm, tissue hypoxia, pressure, wound PH and altered temperature contribute to delayed healing. Treatments should focus on addressing these factors to improve patient outcomes.

INTRODUCTION

Diabetes mellitus (DM) is a cluster of metabolic disorders characterised by high levels of glucose in blood. A report by the International Diabetes Federation highlights that 537 million individuals aged 20–79 years are diagnosed with DM and the incidence is expected to be 46% higher in 2045 than in 2021[1]. Diabetic foot disease is a serious complication of diabetes that is associated with devastating outcomes including diabetic foot ulcers (DFU), delayed healing, infection, amputation and premature death. Furthermore, DM patients with chronic ulcers are at increased risk of depression, anxiety and low self-esteem, which are all established risk factors for delayed wound healing[2].

DFU are characterised as a full-thickness wounds that are present at a level distal to the ankle[3]. DFU are the most common cause of non-traumatic lower limb amputation, which has negative consequences for mortality, and high humanistic and financial costs[4]. Patients with DFU have a high mortality rate, which is about twice that of patients without ulceration. The cost of living with DFU is high in terms of direct and indirect costs, estimated at €11.6 billion per year in Europe in 2017 and €7.6–11 billion annually among Medicare

beneficiaries in the United States from 2007 to 2014[5]. In 2019, the international market cost of chronic wound care was US$10.12 billion, with a projected growth to US$16.36 billion by 2027[6].

DFU are prevalent worldwide. Globally, 40–60 million diabetic patients are affected with diabetic foot and lower limb complications, and the likelihood of developing a foot ulcer may be as high as 25%[7]. Australia has the lowest prevalence of DFU at 1.5%, the prevalence of DFU is 3.9% in Ireland, whilst the highest reported prevalence is in Belgium (16.6%); the global average is 6.4%. The rate of DFU complications is higher in male patients than female. In addition, DFU is more predominant among those with type 2 DM when compared to type 1[8]. Both the age and the length of DM increase the incidence of foot lesions[7].

PATHOPHYSIOLOGY OF DFU

The three pathological components – neuropathy, ischaemia and infection – contribute to DFU and its complications, and they often occur together as an aetiologic triad (Figure 1). The initiating causes are neuropathy and ischaemia, which are frequently combined as neuroischaemia, while infection is mostly a resultant[9].

Diabetes-related peripheral neuropathy affects the distal nerves of the limbs, particularly those of the feet. It primarily affects symmetrical sensory function, resulting in irregular sensations and gradual numbness. Such factors make it easier for ulcers to develop as a result of external trauma and/or irregular distribution of internal bone pressure[10]. The incidence of diabetes-related peripheral neuropathy has been estimated to range from 16% to 87%[11].

Patients can also have sensory, autonomic and/or motor neuropathy. Sensory neuropathy causes the loss of defensive control as well the inability to recognise the consequences of repeated trauma. In the lower limb, autonomic neuropathy can give rise to inadequate sweat gland function as a result of sudomotor dysfunction in diabetes, and is associated with dry skin, itching and anhidrosis which can contribute to the development of foot problems, including ulceration[12]. Callus formation is considered a symptom of DFU since the callus point is subjected to 20 times more pressure than the surrounding tissues[13]. Diabetes-related peripheral neuropathy can lead to devastating outcomes; approximately 50% of people with diabetes will develop a foot ulcer during their lifetime and foot ulcers often precede lower limb amputation. In addition, neuropathic pain and decreased sensation can contribute to an array of poor outcomes including falls, impaired quality of life and depressive symptoms[14].

However, patients with DM and advanced peripheral arterial disease (PAD) are more susceptible to sudden ischaemia caused by progressing atherosclerosis, medio-calcinosis, thrombosis, infections and other factors[15]. Evidence of tissue damage becomes more apparent as the disease progresses, more often in the form of chronic non-healing foot ulcers[16]. Using the ankle brachial index to classify PAD, statistics indicate that 20% of patients with DM over the age of 40 years have PAD and the prevalence increases with age[10].

Patients with multiple, longer duration and deeper wounds have a greater risk of infection. Ischaemia in the foot tends to be linked to a rise in infection severity as DM patients have a reduced inflammatory response. A lack of erythema or induration, which are visual indicators of infection, could be caused by reduced blood flow[13]. The consequence of DFU is closely linked to the use of inappropriate antibiotics to treat diabetic foot infections; DFU patients taking inappropriate antibiotics have 2.5 times higher chance of amputation as compared to appropriately treated DFU patients. Antibiotics prescribed incorrectly can also lead to the production of antibiotic-resistant pathogens[17].

Other risk factors for DFU include peripheral vessel medial arterial calcification, altered foot biomechanics and limited joint mobility, skeletal disease, microangiopathy, Charcot arthropathy, trauma, autonomic neuropathy, history of foot ulceration or amputation, increased plantar pressures, prolonged and uncontrolled DM, smoking, diabetic retinopathy, nephropathy and obesity[5,18].

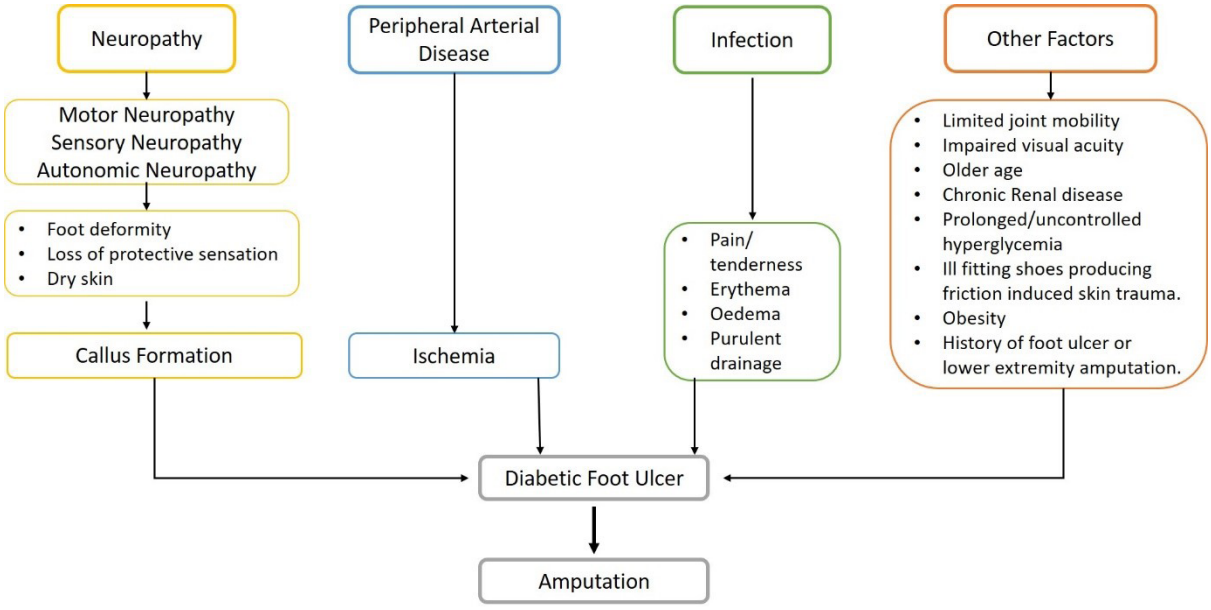

Figure 1. Risk factors associated with DFU [modified]

Over the course of their lives, up to one-third of the world's half-billion patients with diabetes will develop a DFU. More than half of DFU will become infected, out of which 17% will require amputation[19]. It's significant to note that DFU occurs prior to 85% of all lower limb amputations in diabetic individuals[20].

DIABETES AND IMPAIRED WOUND HEALING

Wound healing in the skin is a multifaceted and dynamic process that comprises chemotaxis, inflammation, neovascularisation, and cell division, synthesis of extracellular matrix (ECM) proteins and restoration of anatomic integrity. Wound healing is initiated with haemostasis that controls blood loss and regulates microbe entry to the wound area. An inflammatory phase is followed immediately that cleans up wound debris and prepares the wound site ready for healing. It generally involves three main types of cells – neutrophils, macrophages and mast cells. The proliferative process overlaps the inflammatory phase, during which new tissue, blood vessels and matrix synthesis occurs, allowing tissue regeneration that fills the wound. The ECM's tensile strength is increased and the blood supply to the damaged area is reduced in the final remodelling process[21].

Diabetes-related wounds including DFU are a major concern. Patients with DM exhibit a delayed, impaired and uncoordinated wound healing process. A persistent inflammatory process is observed in DFU healing, which is accompanied by a delay in the development of mature granulation tissue and a decrease in wound tensile strength, subsequently leading to ischaemia[22].

The combined complications of neuropathy, PAD, impaired growth factor (GF) production, keratinocyte and fibroblast migration and proliferation, collagen accumulation, angiogenic response, stability between build-up of extracellular components and their remodelling by proteases, inflammation and hypoxia cause DFU healing to be delayed (Figure 2)[21]. A summary of scientific breakthroughs that shed light on the mechanisms underlying the delayed healing of DFU will be discussed. The key obstacle to the management of chronic wounds must overcome the factors that delay healing as a part of a holistic approach to wound care.

MOLECULAR MECHANISMS

Inflammation

DFU have a chronic pro-inflammatory phenotype, with elevated inflammatory cytokine production. It has been observed that interleukin-1 beta (IL-1β) expression is increased in DFU in both human and mouse tissue samples[23]. Furthermore, high levels of tumour necrosis factor alpha (TNF-α) and monocyte chemoattractant protein-1 (MCP-1), interleukin-1 (IL-1) and interleukin-6 (IL-6) are associated with delayed wound healing in humans[24]. Forkhead box protein M1 (FOXM1), which is involved in the activation and recruitment of inflammatory cells, was also found to be downregulated in DM patients[25]. The roles of various cytokines and GFs involved in wound healing are highlighted in Table 1.

Neutrophils are the first inflammatory cell recruited to the wound site; they function to clear dead cells and infectious microorganisms. Recent research has shown that an increased inflammatory response by neutrophils can have a negative impact on DFU healing[25]. Neutrophil extracellular traps (NETs) are secreted when neutrophils infiltrate a wound to neutralise microorganisms in the decondensed chromatin form by peptidyl arginine deiminase 4 (PAD4)-mediated histone citrullination in a process known as NETosis[26,27]. Wong et al discovered that hyperglycaemia increases neutrophil PAD4 expression, and that the resulting NETs formed in skin wounds are harmful to wound healing[28]. The inflammatory period is also prolonged due to activation of Nod-like receptor protein (NLRP3) inflammasomes in macrophages which stimulate greater production of IL-1β and other cytokines, thus delaying the formation of granulation tissue[29].

Following neutrophils, macrophages are the next cells to migrate to the injury site. Wound macrophages generally transit from a pro-inflammatory (M1) phenotype ($CD14^+CD16^-$ cells in humans) to an anti-inflammatory (M2) phenotype ($CD14^+CD16^+$ cells in humans) during the normal inflammatory stage of wound healing. In the wound bed, this anti-inflammatory transformation stimulates keratinocytes and fibroblasts which proliferate and contribute to the healing process[30]. In DM, however, this phenotypic transition does not occur as easily, and macrophages remain primarily pro-inflammatory, resulting in chronic inflammation[31]. Furthermore, experiments in diabetic mice have shown that pro-inflammatory macrophages' defective efferocytosis of apoptotic neutrophils results in apoptotic cell burden that induces persistent inflammation, preventing macrophages from transitioning to an anti-inflammatory state[32]. Another study in mice demonstrated that a reduction of M2 macrophages with surgical wounds exhibited increased neutrophil count and M1 macrophage infiltration, which helps in extending the duration of inflammatory phase and results in less collagen deposition at the wound bed[33]. These findings indicate that sustained immune activation is an important contributor to delayed wound healing.

PHYSICAL PARAMETERS

Tissue hypoxia

Hypoxia is a condition in which an adequate level of oxygen is not available at the tissue level. In typical wound healing

Figure 2. Factors that delay or inhibit wound healing

situations, local hypoxia stimulates hypoxia inducible factor-1 (HIF-1) which stimulates numerous cellular processes including erythropoiesis, angiogenesis, proliferation and cell survival intended to help adaptive cellular reactions and wound healing[21]. Even with the hypoxia found in diabetic wounds, the amount of HIF-alpha and HIF-1 focused genes are decreased in the wounds of diabetic animal models compared with non-diabetic littermates, causing weakened reactions to cellular hypoxia and prolonging the rate of healing[34]. Prolonged hypoxia, along with hyperglycaemia, is harmful since it exaggerates these early physiological events and causes reperfusion damage as well the production of oxygen free radicals[30]. Hypoxia impairs neutrophil and macrophage activity when combined with hyperglycaemia and other metabolic perturbations[35]. Hyperglycaemia is also linked to the formation of advanced glycation end product (AGE), inactivates HIF-1, and inhibits synthesis of vascular endothelial growth factor (VEGF) and endothelial nitric oxide synthase (eNOS), causing delayed healing. Reactive oxygen species such as hydroxyl radical, superoxide and hydrogen peroxide, as well as reactive nitrogen species, cause increased oxidative stress which causes endothelial damage and slows the rate of healing[36].

Pressure foot-loading

DM patients frequently have increased plantar pressures due to peripheral neuropathy and structural deformities which contribute to the onset of plantar foot ulcers. Increased plantar pressure is an established risk factor for foot ulceration[37]. Subcallus ulcers are caused by the deterioration of the underlying skin and soft tissues caused by persistently high pressures. These ulcers gradually deepen, causing localised deep tissue abscess or osteomyelitis due to prolonged repetitive trauma[38]. Unrelieved pressure, particularly in an insensate foot, results in ongoing mechanical stress and contributes to chronic inflammation in the tissues that, unless addressed, delay wound healing and tissue restoration[39].

pH

In natural conditions, an acidic milieu is found on the surface of skin which is an important feature of the barrier function of skin. A human pathogenic bacterium needs a pH value above 6 to grow which is inhibited by lower pH values of skin. For example, the pathogenic microorganism *Candida albicans* favours increased skin pH, and a more alkaline environment accelerates its overgrowth. Therefore, maintaining an acidic skin pH could help in reducing the microbial overgrowth on the body surface given the fact that they are less resistant to antibiotics. An alteration in the pH value in infected wounds can also change the efficacy of antibiotics. A study has shown that the toxicity of new glycopeptide antibiotic (Oritavancin, LY333328) towards vancomycin-resistant *Enterobacter* species decreases significantly in an acidic milieu with a pH value of 6.4 compared to pH value of 7.4 and 8.4[40]. It has been advocated that the matrix metalloproteinases (MMPs) similar to most other enzymes in the body are very delicate to changes in their instantaneous pH atmosphere[41]. A study by Hart elucidates that creating slightly acidic wound environments would decrease the level of MMPs which, in turn, decreases the inflammatory response[42].

Temperature

The diabetic foot is more prone to ulceration which may in part be due to elevated skin temperature caused by excessive microvasculature blood flow. Using infrared thermal imaging, Long et al discovered that Streptozotocin-induced (STZ) mice had higher wound temperatures which corresponded to slower wound closure[43].

EXTRACELLULAR MATRIX

The ECM, which acts as an interactive scaffold for cells and promotes growth and regeneration in wound tissue, is a significant environmental factor in the healing of DFU. In diabetic patients an imbalance between the synthesis and degradation of the ECM causes a delay in wound healing.

Protease levels in DFU surpass those of their antagonists, resulting in ECM destruction and GF and receptor degradation. The proteolytic degradation of ECM not only stops the wound from progressing into the proliferative stage but also draws in more inflammatory cells, hence accelerating the inflammatory cycle[44].

DM disrupts the equilibrium of MMP concentrations and proteolytic activity. MMPs are a family of zinc-dependent

Table 1. Cytokines and GF involved in wound healing with their expression level

Cytokine and GF	Function	Level of expression during DFU
Interleukin-1 beta (IL-1β)	• Regulates inflammatory mediator production • Stimulates the production of multiple cytokines and chemokines	Decreased
Tumour necrosis factor alpha (TNF-α)	• Regulates activity of fibroblasts, vascular endothelial cells and keratinocytes • Promotes synthesis of ECM proteins and MMPs	Decreased
Transforming growth factor-β (TGF-β)	• Helps initiate granulation tissue formation • Aids in up-regulating the angiogenic GF • Facilitates keratinocytes migration	Decreased
Epidermal growth factor (EGF)	• Aids re-epithelialisation by increasing keratinocyte proliferation and cell migration	Decreased
Fibroblast growth factor (FGF)	• Aids granulation tissue formation, re-epithelialisation and tissue remodelling	Decreased
Platelet-derived growth factor (PDGF)	• Stimulates macrophages to produce and secrete GF • Helps re-epithelialisation by up-regulating the production of Insulin-like Growth Factor 1 (IGF-1).	Decreased
Interferon inducible protein 10 (IP-10)	• Delays re-epithelialisation and prolongs the granulation phase • Inhibits angiogenesis	Increased
Interleukin-8 (IL-8)	• Increases keratinocyte migration and proliferation	Increased

endopeptidases that degrade ECM components involved in tissue remodelling. MMPs digest all matrix proteins, including collagens, elastin, proteoglycans and fibronectin. Although there are 24 different MMP categories, only collagenase (MMP-1 and MMP-8) and gelatinases (MMP-2 and MMP-9) have a role in wound healing[45] (Table 2). The gelatinases (MMP-2 and MMP-9) are the two proteinases that mainly break down type IV collagen from the basic matrix. MMP activity is regulated by tissue inhibitor of MMPs (TIMP) as MMPs are present in an inactive state and need activation to become functional. Thus, it is vital to have a balance between MMPs and TIMPs[46]. Reserved MMPs obstruct wound healing when MMPs are produced in excess during NET formation and cannot be digested to uphold cellular balance[47].

The level of MMPs is 60 times higher in chronic wounds than acute wounds[48]. The increased levels of MMP-1, activated MMP-2, MMP-8, MMP-9, and decreased level of TIMP-1 were found in DFU patients when compared to a wound in non-diabetic patients. Furthermore, high MMP-1 expression is essential for wound healing, but surplus MMP-8 and MMP-9 may slow wound healing in DFU patients, while the MMP-1/TIMP-1 ratio may represent the wound's proteolytic environment[49,50]. Higher MMP-9 expression is associated with poor DFU healing due to poor balance between ECM synthesis and degradation[51].

Hyperglycaemia is linked to lower levels of urokinase plasminogen activator and higher levels of tissue plasminogen activator inhibitor, which may decrease fibrinolysis and impair matrix deposition[24]. Furthermore, in diabetic ulcers, some of the resident cells such as smooth muscle cells and fibroblasts undergo apoptosis due to mitochondrial damage, causing up-regulation of pro-apoptotic proteins and down-regulation of anti-apoptotic proteins, including B-cell lymphoma-2[29]. Fibroblasts isolated from DFU display increased apoptosis, decreased migration ability and reduced proliferative response to GFs such as TGF-β1, platelet-derived growth factor (PDGF) as they become senescent[31]. Fibroblasts are unable to remodel the ECM, causing MMPs, collagenase, serine protease and elastase levels to rise[52]. Type 2 TGF receptor expression is reduced in chronic wound fibroblasts followed by phosphorylation of transduction signals such as Smad2, Smad3 and mitogen-activated protein kinase[53].

BIOFILMS

The study of how unregulated host-pathogen interactions impact healing processes is gaining in popularity. For example, local infection with high levels of replicating bacteria plays a major role in delayed healing and the development of non-healing ulcers[54]. Healing can also be hampered by a high bacterial burden without the classic symptoms of infection[55].

Biofilms can be defined as a complex microbial colony including bacteria and fungi covered in a polysaccharide matrix that can attach to the surface of wounds[56]. This microbial burden is generally polymicrobial and it appears to obstruct host healing. Gram-positive bacteria *Streptococcus agalactiae, Staphylococcus aureus, Enterococcus faecalis and Staphylococcus epidermidis,* as well as gram-negative *Escherichia coli, Pseudomonas aeruginosa, Acinetobacter baumannii* and *Stenotrophomonas maltophilia,* were the most commonly identified pathogens[30]. A report by Trostrup et al showed that VEGF, antimicrobial peptide and neutrophil effector cytokine production is inhibited by *P. aeruginosa*[57]. In addition, the immune system is often ineffective in fighting biofilm-related infections and impairs wound epithelialisation and granulation tissue formation[30].

Polymicrobial biofilms comprising *Bacillus subtilis, S. aureus*, *P. aeruginosa* and *E. faecalis* were found to increase necrosis, delay inflammation and granulation, and hinder ECM production in a porcine model. Upregulation of inflammatory mediators like arginase-1 (ARG-1), IL-8 and chemokine ligand 13 (CXCL13), as well as genes involved in the oxidative stress response like angiopoietin-like 4 (ANGPTL-4) and superoxide dismutase 2 (SOD2), was observed during gene expression analysis[58].

Quorum sensing (bacterial communication mechanism) occurs frequently in biofilms, which influences the chemotaxis towards the surface, availability of key nutrients for biofilm formation, presence of surfactants, bacteria mobility, and surface adhesion[59]. Antimicrobials are unsuccessful at penetrating biofilms, lowering the concentration acting on the bacterial cells within the biofilms and, as a result, biofilms provide a physical barrier to bacteria[60]. These bacterial colonies are frequently multispecies and coated in glycocalyx matrix, making them immune to antibiotics used in topical, parental or oral forms. Within 10 hours of debridement, biofilms can reform. DFU wound bioburden is a notable potentially universal barrier to the healing of chronic wounds due to the diversity of biofilms and their inherent resistance to antibiotics, biocides and host immunity[61,62]. A recent study by Caruso et al indicated a nearly three-fold increase in the risk of antibiotic-resistant infections relative to 2019[63].

ANGIOGENESIS

The inability to rebuild the microvasculature through the process of angiogenesis is a major feature of non-healing

Table 2. Role of metalloproteinases in wound healing

MMP class	MMP subtype	Role in wound healing
Collagenase	MMP-1 (Collagenase-1)	• Aids in remodelling collagen deposition in wound ECM • Promotes keratinocyte survival • Helps keratinocyte migration on type I collagen
	MMP-8 (Collagenase-2)	• Aids in debridement of wound and elimination of damaged type I collagen • Is expressed by neutrophils
Gelatinases	MMP-2 (Gelatinase A)	• Mediates platelet adhesion and aggregation • Aids in cell migration and re-epithelialisation
	MMP-9 (Gelatinase B)	• Is involved in platelet production • Aids in cell migration, mainly keratinocytes and re-epithelialisation

wounds in DM and it mainly affects the proliferative phase[52]. Macrophages are necessary for wound healing because they coordinate the angiogenic response and produce VEGF and other pro-angiogenic mediators in wounds that regulate new blood vessel formation[64,65]. The failure of transition of macrophages during the inflammatory phase adversely affects angiogenesis. In the context of diabetic wound healing, the synthesis of the anti-angiogenic factor, pigment epithelium derived factor (PEDF) was investigated and it has been suggested that increased levels of PEDF could have a deleterious impact on wound healing outcomes[66]. Moreover, the key pathways for maintenance and angiogenesis are angiopoietin-1 (Ang1) and angiopoietin-2 (Ang2), and, in diabetic wounds, Ang2 is dramatically upregulated and the Ang2/Ang1 ratio is dysregulated, disrupting the angiogenesis process[67].

MicroRNAs, often called miRNAs, are another type of molecule that can affect wound healing and the angiogenic process. MiR26-b is highly expressed in diabetic endothelial cells, and neutralisation of this miRNA causes enhanced wound closure and granulation tissue development in diabetes wound models[68]. In diabetic mouse models, restoration of miR27-b regulates angiogenesis in vivo and in vitro in experiments employing local miR27-b,which is thought to impact levels of the anti-angiogenic protein thrombospondin 1 (TSP1) in the wound bed[69].

CONCLUSION

Diabetes frequently affects the healing of wounds which can cause poor outcomes in terms of non-healing wounds, limb threatening infections and amputations. Whilst DFU are complicated to treat, an understanding of the fundamental pathophysiology and a focus on controlling these disturbances may lead to successful wound healing. The main obstacle in the management of chronic wounds is overcoming the factors that lead to delayed healing; these interventions should occur as part of a holistic approach to wound care. Due to severe infection, irreversible ischaemia, imbalance in cytokine, and GF production in the wound bed, patients with DM are more likely to need to have tissue resection. Intensive therapy is required as early as feasible after the development of an ulcer to minimise its chronicity, resultant morbidity and associated mortality.

To enhance the management of foot disease in diabetic patients, primary care professionals must be made aware of the significance of early referral to a specialised unit. Combination approaches involving advanced wound therapies and MMP inhibitors, ECM stimulator, GF, cells combinations or angiogenesis stimulator can be used at different phases of wound healing. Ulcer resolution and ulcer recurrence can be aided by biomechanical examination and treatment planning. When DFU do not heal adequately after 4 weeks of standard treatment, the underlying pathology should be re-evaluated, and the need for advanced therapies that can address molecular and/or physical disturbances should be considered.

IMPLICATIONS FOR CLINICAL PRACTICE OR FUTURE RESEARCH

- Patients with DM exhibit delayed, impaired and uncoordinated wound healing.
- An understanding of the fundamental pathophysiology and a focus on controlling these disturbances may facilitate successful healing.
- When DFU do not heal adequately after 4 weeks of standard treatment, the need for advanced therapy, taking into account the pathophysiological deficits, should be considered.
- Future research to better understand the role of molecular and physical parameters in impaired wound healing in diabetes is needed.
- There is a need for the development of advanced wound products that specifically aim to address the pathophysiological components of impaired healing in DFU, and that are shown through definitive trial designs to improve clinical and patient outcomes.

AUTHOR CONTRIBUTIONS

All authors meet the criteria for authorship. All authors made substantial contributions to this literature review including conception and design, literature searching, drafting of the manuscript, critical revision of the manuscript and final approval of the manuscript.

CONFLICT OF INTEREST

Prof. Timothy O'Brien is a founder, director and equity holder in Orbsen Therapeutics.

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