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Consensus document on treatment of infections in diabetic foot

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ABSTRACT

Diabetic foot infection, particularly if it is associated to ischaemia, is the most common cause of lower limb amputation, in the general population, of hospital admissions, and a decrease in the quality of life in diabetics. Of all diabetics, 15% of them are going to suffer from a foot infection during their life, with an annual incidence of 1-4%, preceded by a foot ulcer in more than 80% of cases. They are complex infections and the prognosis is influenced by many factors, depending on the ulcer (location, extension, whether chronic or not, previous amputation, ischaemia grade), and the patient (age, renal impairment, time of onset of diabetes, associated comorbidity). All these must be taken into account when establishing its treatment.

The infections must be classified according to their severity (mild, moderate-mild, moderatesevere, and severe). Their treatment is complex and must be multidisciplinary and must include debridement, discharge, adequate antibiotic therapy, revascularisation, and treatment of the ulcer.

In this consensus document, produced in collaboration with the Spanish Angiology and Vascular Surgery Society (SEACV), the Spanish Society of Internal Medicine (SEMI), the Spanish Chemotherapy Society (SEQ), the Spanish Surgeons Association (AEC), the Spanish Society of Urgent Medicine and Emergencies (INFURG-SEMES) and the Spanish Society of Intensive and Critical Medicine and Coronary Care (SEMICYUC), the guidelines are developed based on the best available evidence on diabetic foot infections, aimed at achieving greater clinical efficacy.

Key Words: Diabetic Foot, Diabetic Foot Ulcer, Diabetic Foot Infection, Antibiotics

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Documento de consenso sobre el tratamiento de las infecciones en el pie del diabético

RESUMEN

La infección del pie diabético, sobre todo si se asocia a isquemia, es la causa más frecuente de amputación de extremidad inferior en la población general y de ingreso hospitalario y disminución de la calidad de vida en los diabéticos. EL 15% de los diabéticos van a sufrir a lo largo de su vida una infección del pie, con una incidencia anual del 1-4%, precedida en más del 80% de los casos de una úlcera en el pie. Son infecciones complejas en cuyo pronóstico influyen muchos factores, dependientes de la úlcera (localización, extensión, cronicidad, amputación previa, grado de isquemia) y del paciente (edad, insuficiencia renal, tiempo de evolución de la diabetes, comorbilidad asociada), lo que hay que tener en cuenta a la hora de plantear su tratamiento. Las infecciones deben clasificarse en función de su gravedad (leves, moderadas-leves, moderadas-graves y graves). Su tratamiento es complejo y debe ser multidisciplinar. Debe incluir desbridamiento, descarga, antibioticoterapia adecuada, revascularización y cura de la úlcera.

En este documento de consenso, fruto de la colaboración de la Sociedad Española de Angiología y Cirugía Vascular (SEACV), Sociedad Española de Medicina Interna (SEMI), Sociedad Española de Quimioterapia (SEQ), Asociación Española de Cirujanos (AEC), Sociedad Española de Medicina de Urgencias y Emergencias (INFURG-SEMES) y Sociedad Española de Medicina Intensiva, Crítica y Unidades Coronarias (SEMICYUC), se desarrollan las pautas, basadas en la mejor evidencia disponible, de las infecciones de pie diabético, encaminadas a obtener la mayor eficacia clínica

Palabras Clave: Pie diabético, Ulcera pie diabético, Infección pie diabético, antibióticos

RATIONALE

Diabetes is a health problem of the first order, as shown by its high prevalence and numerous consequences. One of the most common complications during the life of a diabetic is the development of an ulcer in the foot. An important European

study[1] states that half of these ulcers are associated with ischemia (49%) or infection (58%), or the combination of both in one third of cases (31%). This aggravates the condition, increasing in the rate of amputations and mortality in these patients.

Conscious of the problem of diabetic foot (DF), the Spanish Society of Angiology and Vascular Surgery (SEACV), commissioned in 1996 an "ad hoc" committee to develop a consensus on consensus on DF[2]. Years later (2005), the SEACV, based on the importance for the specialty of all aspects related to DF, created the specific DF group, now Section, called Pie Diabético - SEACV[3]. Only one year later, members of the SEACV and of that Section were asked to be part of an expert panel, on behalf of several medical societies, to develop a consensus document on the antimicrobial treatment of DF infections, where they have provided treatment regimens based on the best available evidence for achieving the greatest clinical efficacy[4]. The treatment of infections of the DF, of great complexity, requires multidisciplinary care because of the multiple factors involved in its development[5].

This document is primarily designed to give continuity to and update the previously mentioned consensuses. The level and quality of clinical evidence is not the most desirable due to the lack of homogeneity of the available information (clinical trials), so many of the decisions presented are based on expert opinion.

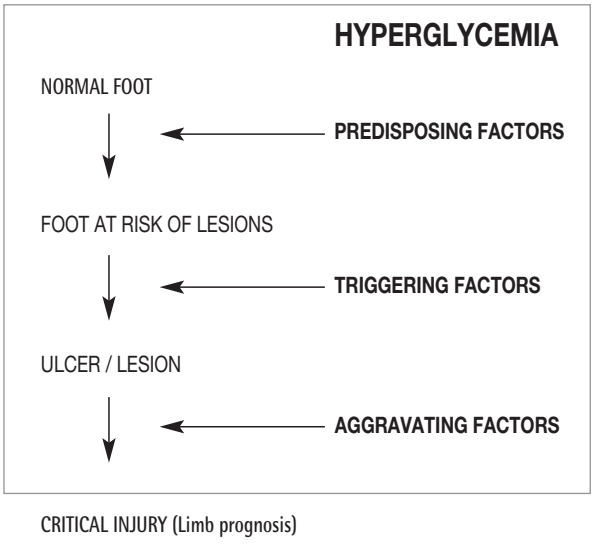

Figure 1 General pathophysiology of diabetic foot ulcer.

EPIDEMIOLOGY

Diabetes is a highly prevalent disease (6% of the population), with a similar proportion of undiagnosed patients who have the disease, which has multiplied by 6 the number of diabetics in the past 40 years[6]. In addition, there is an increase with age, reaching 11% in the persons over 65[7]. In developed countries, it is the 7th cause of death as a direct cause, without taking into account its role in cardiovascular mortality, the leading cause of early death in diabetics[7-9].

Diabetic patients, as a consequence of their extended life expectancy, have many problems, including DF. The main late complications of diabetes (atherosclerosis, neuropathy, retinopathy, etc.) are vascular (macro and microangiopathy) and metabolic in their pathogenesis. Foot ulcer is one of the most common complications in the lower extremities of diabetics. It appears during the course of disease in approximately 15% of cases[10-12]. Its annual incidence is 2-3% and 7% in patients with neuropathy, and its prevalence is 2-10%[13,14].

Foot infections affecting the skin and soft tissues, and bone, with or without systemic impact, are the most common reason for hospitalization of diabetics (25%), with prolonged stays[11].

Diabetes is the most common cause of lower extremity amputation in Europe and the U.S.[15]. The annual rate of amputations adjusted for age is 82 per 10,000 diabetics. These patients have a 15 to 40-fold greater risk of requiring an amputation than the nondiabetics and men at least 50% more than women[8,16]. Diabetics with a foot ulcer will require an amputation in 14-20% of cases and in turn foot ulcer is the precursor of more than 85% of lower extremity amputations in these patients[17,18]. After amputation of a lower limb, the incidence of a new ulcer, and/or contralateral amputation at 2-5 years is 50%[11,19]. Survival of diabetic patients undergoing amputations is significantly worse than the rest of the population and even less if they have experienced another prior amputation[11]. Only 50% to 40% of patients survive 3 and 5 years from an amputation, respectively, and prognosis worsens as the level where it is performed increases[19,20].

Although the costs derived from DF ulcers and other infections are not accurately known, in the U.S. it is estimated that an ulcer episode costs from $4,500 to $28,000 at two years after diagnosis, with a mean of $5,500 per patient per year[21,22]. Although mean hospital stay of an amputation has decreased, it remains a costly procedure, ranging from $20,000 to $40,000 depending on the level of amputation, hospital stay, or patient comorbidities[11,23]. More up-to-date and similarly high values are available for Europe[24].

Finally, we should mention that recent Spanish epidemiological studies are available[25,26] which report along the same lines (prevalence, frequency of amputation, mortality, etc).

PATHOPHYSIOLOGY

Understanding of the pathophysiology of DF is essential for optimal care, since modifying the factors that influence its development can restore or keep the foot intact, conserving the limb and maintaining a healthy foot so that the patient can lead a completely normal life. Although DF lesions may seem different, the path leading to a foot ulcer and its complications is very similar, and is determined by various factors. Neuropathy, present in more than 90% of ulcers, plays a major role in the development and progression of DF. It causes an in-

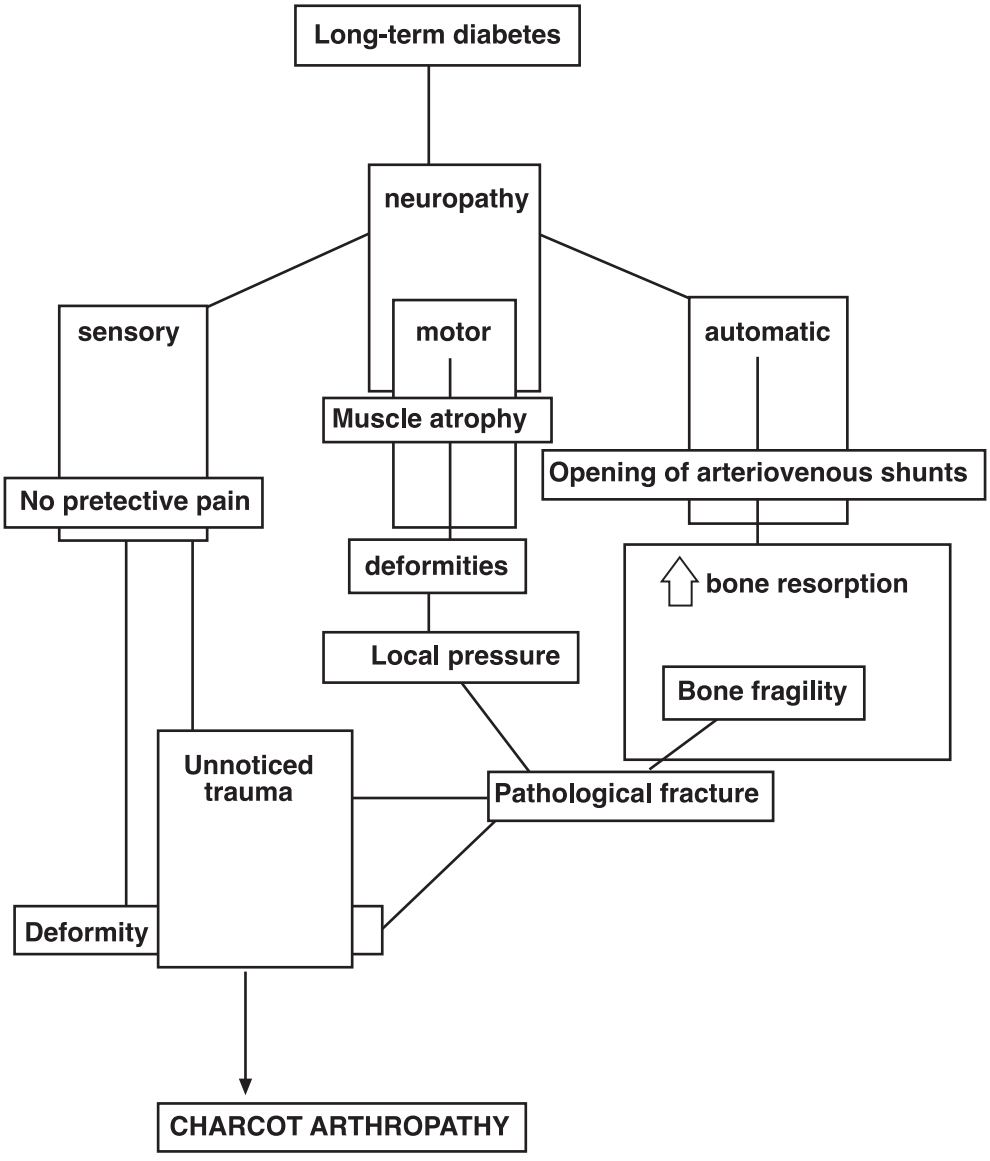

Figure 2 | **Pathogenesis of charcot neuroarthropathy.**

sensitive and deformed foot, altering gait biomechanics, developing hyperkeratosis (callosities) where plantar pressure is concentrated, and where an ulcer occurs because of a small lesion. If the patient is not aware of them because of the loss of sensitivity, he/she continues walking, causing healing to be altered. Ischemia due to arterial obstruction, which is present in 50% of ulcers, and infection, are the factors that will determine the prognosis of the ulcer and the limb.

Schematically, there are predisposing factors, neuropathy associated with a greater or lesser degree of macro- and microangiopathy, which cause a high-risk, vulnerable foot, precipitating or triggering factors, generally a mechanical injury, causing an ulcer or necrosis, and aggravating factors, which determine the prognosis of the extremity and include infection, which will induce extensive tissue damage, ischemia which will delay healing and neuropathy which will prevent recognition of both the injury and the triggering factor[27] (figure 1).

The most common form of neuropathy is metabolic polyneuropathy, a condition characterized by symmetrical, distal, chronic, insidious onset, somatic (sensory-motor) and autonomic dysfunction. It predominantly affects the lower ex-

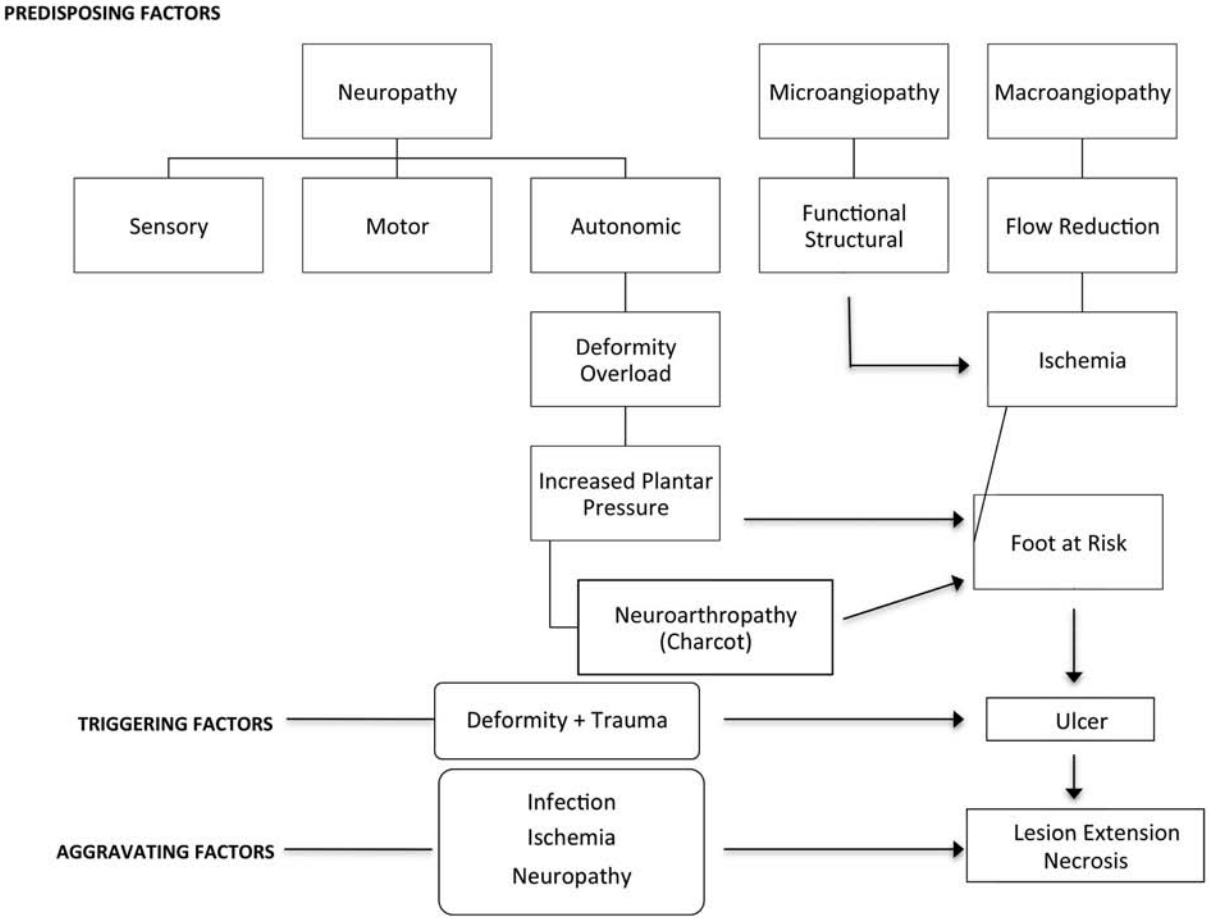

Figure 3 | **Mechanism producing diabetic foot ulcer.**

tremities[28]. It is found in approximately 30% of diabetics[29], and increases in prevalence with increasing duration of disease[30]. Sensory involvement is usually asymptomatic. It initially causes loss of sensation of pain and temperature, and subsequently of perception of vibration and superficial sensitivity. Because of this, diabetics are unable to detect changes in temperature, excess pressure produced by tight-fitting shoes or any other continued trauma. Motor involvement causes atrophy and weakening of the intrinsic muscles of the foot, with loss of the stabilizing function of metatarsophalangeal and interphalangeal joints, causing a dynamic contracture of flexors and long extensors inducing hammer toes and claw toes, which leads to protrusion of the metatarsal heads and an abnormal distribution of loads in the foot[28]. Autonomic neuropathy[31] results in anhidrosis, causing from dry skin or fissures forming callus tissue in load areas to opening of cutaneous arteriovenous shunts, which in the absence of obstructive arterial disease, decrease perfusion of the capillary network and increase skin temperature, causing a postural disturbance in flow regulation and an abnormal inflammatory response to tissue insult[32] and neuropathic edema[33]. All this, by increasing osteoclast activity and via an interleukin-mediated inflammatory reaction, can result in Charcot neuroarthropathy, one of the worst consequences of diabetic foot[34] (figure 2).

Neuropathy, with or without associated ischemia, is implicated in the pathophysiology of DF ulcer in 85 to 90% of cases[35-37], and ischemia due to diabetic macroangiopathy in 40 to 50%, usually associated with neuropathy[35,36]. Diabetic macroangiopathy is simply atherosclerosis in diabetic patients, without differences in the type of pathological damage. However, it appears at a younger age, with a similar incidence in both sexes[38], but with a different location of the lesions, which is usually multisegmentary, bilateral and distal[39].

There is a wide controversy about the true importance of diabetic microangiopathy in the pathophysiology of DF[40]. There is no decrease in lumen diameter, but a thickening of the capillary basement membrane secondary to hyperglycemia,

Table 1 **Etiology of diabetic foot infections.**

Infection	Microorganisms
Cellulitis	*Staphylococcus aureus*
Erysipelas	Beta-hemolytic streptococci (A, B, C and G)
Ulcer untreated with antibiotics	*Staphylococcus aureus*
	Beta-hemolytic streptococci (A, B, C and G)
Ulcer treated with antibiotics or long-term (generally polymicrobial)	*Staphylococcus aureus*
	MRSA
	Coagulase-negative staphylococci
	Streptococcus spp.
	Enterococcus spp.
	Enterobacteriaceae
	Pseudomonas aeruginosa[1]
	Other nonfermenting gram-negative bacilli[2]
	Corynebacterium spp.[2]
	Candida spp.[2]
Necrotizing fascitis or myonecrosis (generally polymicrobial)	Anaerobic gram-positive cocci
	Enterobacteriaceae
	Nonfermenting gram-negative bacilli
	Anaerobes

MRSA: Methicillin-resistant *Staphylococcus aureus*

nonenzymatic glycosylation of collagen and protein glycans and genetic susceptibility[41]. There are functional abnormalities at the capillary level, because the ultimate cause of tissue necrosis is failure of microcirculatory function, which in diabetics is due to an interaction of the effects that neuropathy, macroangiopathy, and microangiopathy itself on the microcirculation.

Neuropathy in particular, combined in some cases with ischemia, together with the other factors described, are the factors that place the diabetic foot at risk of ulceration. However it is its combination with the precipitating or triggering factors which causes the ulcer. The principal factor is trauma[42], mechanical, thermal or chemical[27], the most common being mechanical, usually by poor fitting shoes in the neuroischemic ulcers or pressure overload in callosities formed in neuropathic ulcers[35,43].

Infection does not usually causes ulcer, except in specific cases of fungal infections (tinea pedis, candidiasis) in the interdigital spaces. However, it will largely determine both the prognosis and treatment for any foot lesion, particularly when associated with ischemia. The break or opening in the skin caused by a foot ulcer is a port of entry for microorganisms. There is an impairment of the immune system, both cell-mediated and humoral, specifically of the granulocytes, affecting diapedesis, leukocyte adherence, chemotaxis, phagocytosis and intracellular lysis. These aspects are aggravated by poor control of blood glucose[44], which on the other hand causes nonenzymatic protein glycosylation, affecting their function and structure. This together with loss of the sensitivity induced by neuropathy allows patients to walk on infected tissue without being aware of it, thus facilitating spread of infection to deeper levels, increasing its severity[45]. Traditional signs of inflammation may be due to another cause such as acute Charcot osteoarthropathy, or be absent in the case of ischemia or underlying osteomyelitis[46].

The complex pathophysiology of DF can be summarized in that chronic hyperglycemia induces biophysical and biochemical changes in different body organs and systems. In the case of the foot, primarily neuropathy and macroangiopathy and to a lesser extent microangiopathy, turn the diabetic foot into a high-risk foot, in which chronic and sustained trauma usually causes an ulcer. Prognosis of the ulcer and therefore of the limb will depend on the greater or lesser extent of neuropathy, ischemia and infection associated with the ulcer[4] (figure 3).

ETIOLOGY

The microorganisms involved in the etiology of DF infection vary depending on the type of infection and specific patient situations (antibiotic therapy, previous manipulation or hospitalization) (table 1)[4,47].

Superficial infections, such as erysipelas and cellulitis, are usually caused by gram-positive organisms, particularly Group A, B, C and G beta-hemolytic streptococci, and *Staphylococcus aureus*, respectively[4,47].

Ulcer infections are generally polymicrobial and mixed, the most common isolates being facultative and anaerobic gram-positive and gram-negative bacteria, and *Candida* spp[48,49]. The complexity of the flora found increases with hospital admissions, clinical duration of the ulcer[48,49], depth/severity of the lesion and history of antimicrobial treatments[48]. Occasionally, cultures are negative (6-12%)[48,50]. This may be due, among other circumstances, to the fact that microbiological studies are performed on samples while the patient is receiving antibiotics, the samples are not representative of the infection, the microbiological methodology used is inadequate or because techniques with adequate sensitivity are not available.

In recent infections, cultures are more commonly caused by a single organism and the most widely found bacteria include *S. aureus* followed by different species *Streptococcus*[49]. In long-term infections, the role of *S. aureus* and streptococci is still important although the percentage of their recovery is lower. There is an increase in coagulase-negative staphylococci (CNS), *Enterococcus* spp, gram-negative bacilli -particularly *Pseudomonas aeruginosa*- and anaerobes[49]. In India, in recent studies, the percentage of isolates of some species of *Enterobacteriaceae* and *P. aeruginosa* equal or even exceeds the number of *S. aureus*[51,52].

In moderate or severe ulcer infections, using very demanding microbiological technology and in patients who did not receive antibiotics, anaerobic isolation has been reported

in almost half of positive samples, in most cases in association. From 1 to 8 bacterial species were isolated per sample with a mean of 2.7 microorganisms per culture. By order of frequency, aerobic and facultative organisms recovered included methicillin-susceptible and -resistant *S. aureus* (18.7%), CNS (15.3%), *Streptococcus* spp (15.5%), *Enterococcus* spp (13.5%), Enterobacteriaceae (12.8%), *Corynebacterium* spp (10.1%) and *P. aeruginosa* (13.5%). Distribution of anaerobes was as follows: gram-positive cocci (45.2%), *Prevotella* spp (13.6%), *Porphyromonas* spp (11.3%), and different species from the *Bacteroides fragilis* group (10.2%). The differences observed in the bacteriology of DF ulcers were related to the type of sample used, the quality of microbiologic processing, the presence or absence of infection, severity, prior antimicrobial therapy and geographic and temporal variations[48]. In a study using molecular techniques to investigate the etiology of infected chronic ulcers, *S. aureus* was the bacteria most detected[53]. Other aerobic and facultative organisms found were *Morganella morganii*, *Enterococcus faecalis*, *Citrobacter* spp and *Haemophilus* spp. Among anaerobes, the most common were: *Anaerococcus* spp, *Bacteroides fragilis*, *Finegoldia magna*, *Peptoniphilus spp*, *Clostridium* spp, and *Veillonella* spp.[53].

In complicated infections, a recent study conducted in Spain has shown results that are similar except for the fact that there was a higher proportion of monomicrobial etiology (59%). Most patients had received antibiotics in the previous month. Gram-positive organisms were the most recovered both in monomicrobial and polymicrobial cultures. Among gram-negative bacilli dominated *Enterobacteriaceae* on non-fermenting gram-negative bacilli. Anaerobic bacteria were mainly isolated in polymicrobial cultures. By order of frequency, the most recovered species were *S. aureus* (33%), *P. aeruginosa* (12%), *Escherichia coli* (8%), and *E. faecalis* (8%). Thirty-eight percent of *S. aureus* strains were methicillin resistant (MRSA) meaning that this bacterium was present in 12% of clinical samples analyzed[50].

In necrotizing fascitis and gangrene, the most comon isolates are facultative gram-positive cocci, enterobacteria, non-fermentative gram-negative bacilli and anaerobes[4,47].

In osteomyelitis, a large number of samples taken by biopsy or aspiration are sterile. In those in which bacterial growth is obtained, few bacterial species are usually found, often only one. The microorganisms isolated are similar to those found in chronic ulcers. In a recent study, in about 50% of all cases gram-positive bacteria were isolated, particularly methicillin-susceptible and -resistant *S. aureus* followed at a distance from CNS, group B streptococci, enterococci and corynebacteria. Gram-negative bacilli were recovered in almost 40% of cases with Enterobacteriaceae exceeding non-fermenting bacilli. In approximately 10% of cases, bacteria were anaerobic[54-56].

The bacteria isolated from DF infections may be multiresistant. Previous antibiotic treatment, duration of antimicrobial treatment, frequency of hospital admissions for the same wound, duration of hospital stay(s), presence of osteomyelitis[57], neuropathy and ulcer size[58] have been reported as significant risk factors.

In terms of specific bacterial species, *S. aureus* is the most recovered, both in mild and severe infections, as well as in recent and long-standing infections. In 20% of cases, it is isolated as a pure culture[48]. This organism hinders cure and persistently colonizes ulcers[59], particularly in the deep part and surrounding tissues[60]. It is thought that in most infections, methicillin-susceptible *S. aureus* (MSSA), and (MRSA) and streptococci are the primary pathogens and targeted treatment to them would cure them regardless of the associated bacteria. This has lead to formulation of the "Head of the Snake" concept (gram-positive cocci) has been formulated, according to which by destroying the head of the snake the body would be killed (gram-negative bacilli and anaerobes)[61]. There have been reports of the existence of genomic differences between the colonizer strains and those causing infection. In the latter, resistance genes to aminoglycosides are more common[59] and those that encode some virulence determinants[59,62]. A significant number of *S. aureus* produces a mucosal layer and polysaccharide intercellular adhesin[63] and a variable percentage are methicillin resistant. In a review of Eleftheriadou[64], MRSA strains represented from 15 to 30%, and were detected in both the hospital and community. In Spain and in diabetic foot with complicated infections, it was reported in 38%[50]. Furthermore, it should be noted that its presence increased over time[65]. The risk of resistance to methicillin is increased by previous hospital admission, ulcer duration, chronic kidney failure[49], presence of osteomyelitis, nasal colonization, previous use of antibiotics and ulcer size[64]. MSSA and MRSA infection is a significant predictive factor for amputation in an extremity[66]. Knowledge of the local prevalence of methicillin resistance is important for starting empirical antimicrobial therapy. Sensitivity of MRSA to vancomycin is a controversial. The minimum inhibitory concentrations (MICs) have increased (strains with moderate and high resistance), there are tolerant isolates[67], and there are even many therapeutic failures in strains with MICs within the sensitivity range[68].

Among CNS, isolation of *S. epidermidis, S. lugdunensis*, *S. haemolyticus* and less commonly of *S. auricularis, S. capitis, S. caprae, S. cohnii, S. hominis, S. schleiferi, S. sciuri, S. simulans, S. warneri*, and *S. xylosus* has been reported. *S. epidermidis* is usually most common and its isolation is not usually associated with *S. aureus*[48]. Most strains of *S. epidermidis* recovered from DF produce a mucosal layer and polysaccharide intercellular adhesin[63]. *S. epidermidis* is usually isolated more in neuroischemic than in neuropathic ulcers[69].

Streptococcus agalactiae is isolated in DF infections, even in severe forms, particularly if there is chronic renal failure, severe arterial disease, alcoholism, overweight and/or immunosuppression[70].

The role of enterococci is controversial as evidenced by the good clinical response of ulcers with this microorganism when they were treated with ertapenem, an antibiotic to which they have natural resistance[71].

In a significant percentage of patients, a variety of species of *Corynebacterium* are isolated[48]. Usually considered poorly pathogenic, recent genomic studies have shown that they are a significant part of the characteristic biofilm of chronic DF infections[72].

P. aeruginosa, a bacterium producing a mucosal layer, is isolated most commonly and significantly in chronic long-term ulcers[49]. Traditionally, the circumstances promoting maceration of ulcers are considered risk factors[4]. In many cases a history of antimicrobial treatment in the previous month is documented[50]. Its pathogenic role, as in the case of *Enterococcus*, is not clear as in mixed infections in which it was isolated a similar clinical response was reported with ertapenem and piperacillin-tazobactam[71]. As in other infections, carbapenemase-producing strains resistant to imipenem and/or meropenen are isolated[73]. Less commonly, other non-fermenting bacilli such as *Stenotrophomonas maltophilia, Alcaligenes faecalis*[48] or *Acinetobacter* spp are recovered[51].

Among the Enterobacteriaceae isolated, *E. coli* and *Klebsiella* spp are predominant and may be extended-spectrum beta-lactamase-producing (ESBL), a trend that in some countries such as India is growing[52,58,74,75], where it reaches up to 44.7%[58]. In Spain, 29% of *E. coli* strains were resistant to amoxicillin-clavulanate and ciprofloxacin[50]. *Enterobacter cloacae*, *Serratia marcescens* and *Citrobacter freundii* are also recovered[48,50], species that produce chromosomally encoded inducible AmpC beta-lactamase, and *Proteus* spp, *Providencia* spp and *Morganella morganii* [48,50] with natural resistance to tigecycline.

Data on involvement of fungi in DF infection are limited and discordant. Yachts et al only isolated fungi in 0.4% of the isolates[49]. Prospective studies aimed at the search for fungi in DF ulcer infections found that geographical differences–associated with other factors–are marked. A study conducted in Croatia estimated isolation of *Candida* spp at 4.3%. The most recovered species was *C. parapsilosis*, usually associated with different bacteria in the setting of a severe mixed infection76. In contrast, another study conducted in India found fungi in 65% of patients. In 77% yeasts were isolated, mostly from the genus *Candida* (93%), particularly *C. albicans* (49%), *C. tropicalis* (23%), *C. parapsilosis* (18%), *C. guillermondi* (5%) and *C. krusei* (5%). The other species of yeasts isolated were *Trichosporon cutaneum* and *T. capitatum*. *Trichophyton* spp was the only dermatophyte recovered. Moulds were isolated in 38% of patients, especially *Aspergillus* spp (72%). *Fusarium solani*, *Penicillium marneffei* and *Basidiobolus ranarum* were also cultured[77].

Biofilms are more common in chronic skin ulcers, in which are included many bacteria as shown genomically[72]. The biofilm explains some of the characteristics of these infections such as chronicity, polymicrobial etiology, importance and difficulty of sample collection for diagnosis, limitation of traditional microbiological techniques that only recover some bacteria, utility of genomic techniques and problems in treatment because it is essential to remove it (debridement).

CLASSIFICATION

It would be ideal to have a classification system of DF lesions that standardized the definitions of the different clinical scenarios. This classification is necessary to know the course of different lesions, to compare the different treatment results and to improve interdisciplinary communication. A great variety of classification systems has been developed though none has been universally accepted.

Shea proposed to classify pressure wounds by grades according to the depth of the wound and the structures that were progressively exposed at the base of the wound[78]. Meggitt established a classification of DF ulcers applying this concept[79], a classification popularized by Wagner, which has become classical and is probable the most commonly used in the world[80].

It gives great importance to depth of the lesion. It starts

Table 2 **IDSA classification of severity of diabetic foot infection (adapted by SEACV).**

IDSA (adapted by SEACV) Severity of Infection	Clinical Signs of infection	IWGDF PEDIS grade
NO INFECTION	No inflammatory signs and effusion	GRADE 1
MILD INFECTION	No systemic signs of infection Evidence of purulence or 2 or more signs of inflammation	GRADE 2
MODERATE-MILD INFECTION	No systemic signs of infection. Cellulitis >2cm. Deep tissue infection (crosses subcutaneous cellular tissue, no abscess, lymphangitis, arthritis, osteomyelitis, myositis or critical ischemia)	GRADE 3
MODERATE-SEVERE INFECTION	No systemic signs of infection. Cellulitis >2cm. Deep tissue infection (crosses subcutaneous cellular tissue, no abscess, lymphangitis, arthritis, osteomyelitis, myositis or critical ischemia)	GRADE 3
SEVERE INFECTION	Any infection associated with systemic toxicity (fever, chills, vomiting, confusion, metabolic instability, shock)	GRADE 4

IWGDF: International Working Group on the Diabetic Foot

PEDIS System: Perfusion, Extension, Depth, Infection, Sensitivity.

with grade 0 ("foot at risk" with unpenetrated skin), progresses to grade 1 (destruction of skin barrier reaching the subcutaneous cellular tissue) and then to grade 2, where the tendon, joint capsule, or bone is exposed. If this deep ulcer is associated with septic arthritis, osteomyelitis, or abscess, it is grade 3. Grades 0 and 1 are usually treated on an outpatient basis, 2 and 3 requires hospital admission. Grades 4 and 5 imply ischemic gangrene, the former limited to the forefoot and the latter to all the foot. In neuropathic ulcers, there is a decreased rate of healing and increased rate of amputation as ulcer depth increases and reaches tendon, bone, or joint[81,82]. In fact, all subsequent classifications include the depth of the wound as a basic parameter. The limitation of this classification lies in that grades 4 and 5 are not a more advanced phase than 1, 2 and 3, it does not differentiate whether or not there is ischemia in grades 1, 2 and 3, which is essential to determine prognosis, and it does not specify if there is associated infection and to what extent.

Ischemia and depth have been analyzed separately[83], the most comprehensive classification being probably that of the Texas University[84]. It assesses depth, ischemia and infection. It relates grades 0, I, II and III of lesion depth, similar to those previously described in the above classifications, with a stages A, B, C and D: No infection, no ischemia (A); Infection with no ischemia (B); Ischemia with no infection (C); Ischemia and infection (D). It has been validated by its authors demonstrating that as grade and stage of the lesion increase, its prognosis worsens, and probability of amputation is much higher[85].

For categorizing ulcers into subgroups, severity scoring systems based on standardized clinical parameters of the wound have been developed, such as: a) the S(AD)SAD system, assessing size with area and depth, sepsis, arterial disease and denervation, grading each parameter from 0 to 3 in increasing order of severity[86]; b) the PEDIS system (Perfusion, Extension, Depth, Infection and Sensation)[87]; c) the DUSS (Diabetic Ulcer Severity Score) based on the presence or absence of four severity criteria, assessed with one point if this criterion is present and zero points if not present: absence of distal pulses (1 point), positive probing to bone (1 point), ulcer located at any area of the foot other than toes (1 point) and the presence of more than one ulcer (1 point)[88]; and d) the Strauss wound scoring system, assessing 5 parameters and grading each of them from 0 to 2[89].

A key aspect in the management of DF infections is to assess the severity of infection, which determines the prognosis and the therapeutic strategy. They can be classified by their depth into superficial (skin and subcutaneous tissue–SCT-) and deep (penetrating superficial fascia and deep structures) or by clinical signs as mild (without risk for the limb, superficial, with cellulitis under 2 cm in size), moderate (threatening to the limb, deep, with frequent osteomyelitis and more extensive cellulitis, usually requiring hospitalization) and severe (life-threatening, associated with sepsis, usually presenting massive cellulitis, deep abscesses, necrotizing fascitis and/or myonecrosis and usually requiring emergency surgery)[90].

Table 3 Differential diagnosis between the neuropathic and neuroischemic ulcers.

Neuropathic ulcer	Neuroischemic ulcer
Painless	Painful
Normal pulses	Absent pulses
Punched-out appearance	Irregular margins
Located on the sole of the foot	Usually located on the toes
Presence of callosities	Callosities absent or uncommon
Loss of sensitivity, reflexes, and vibratory sense	Variable sensory findings
Increased blood flow (arteriovenous shunts)	Decreased blood flow
Dilated veins	Collapsed veins
Hot, dry foot	Cold foot
Reddish appearance	Pale, cyanotic appearance
Bone deformities	No bony deformity

The International Consensus on the Diabetic Foot (2003) classified infection into 4 grades (PEDIS): 1 (no signs of infection), 2 (mild infection with involvement of skin and subcutaneous cellular tissue only), 3 (moderate infection with extensive cellulitis and/or deep infection), and 4 (severe infection with presence of systemic inflammatory response syndrome). It is similar to the IDSA (Infectious Disease Society of America) classification91 and has been validated as having prognostic value in DF infections[92]. The main advantage of this classification is probably that it calls attention to the signs of systemic toxicity as markers of the severe infection in these patients. However, the absence of such signs does not exclude a severe infection that could be life-threatening since actually more than half of these patients do not present systemic signs of infection. On the other hand, if ischemia is associated with infection, irrespective of the grade of the latter, severity is increased.

For practical purposes, the important thing is to know which infections can be safely treated on an outpatient basis, those requires hospital admission because they threaten the lower limb and which are life-threatening and require expedited diagnostic/therapeutic decision-making process. It is generally considered that there is mild infection (PEDIS grade 2) if there are two or more clinical signs of local inflammation (suppuration, erythema, pain, sensitivity, heat and induration), but they do not extend beyond 2 cm. around the ulcer, the depth of the ulcer does not exceed the subcutaneous cellular tissue and it is not associated with other complications or systemic involvement. No threat for the limb, although there a risk of osteomyelitis (10-20%). Infection is moderate (PEDIS grade 3) when in addition to presenting 2 or more clinical signs of inflammation, it has one or more of the following: cellulitis >2 cm around the ulcer, signs of local dissemination (lymphangitis and lymphadenopathy), or reaches deep tissues:

fascia (necrotizing fascitis), muscle (myonecrosis), joint (arthritis), bone (osteomyelitis), or tendon; without systemic and metabolic stability. In these cases there is the risk of losing the extremity. However, this is a very large group that covers a wide spectrum of infections with different prognosis, distinguished also by the presence or absence of associated ischemia. In this group, two subtypes of infection can also be distinguished: 1) mild-moderate, defined by the presence of cellulitis >2 cm, limited to the dermis, without lymphangitis or critical ischemia, which would not require hospital admission, but early reassessment 2) moderate-severe, if cellulitis >2 cm is associated with lymphangitis or critical ischemia or extends deeply, which would require hospitalization. Severe infection (PEDIS grade 4) is a moderate infection associated with systemic toxicity or metabolic instability (fever, chills, tachycardia, hypotension, confusion, vomiting, leukocytosis, metabolic acidosis, severe hyperglycemia or uremia). They are life-threatening for the limb and the patient (table 2).

DIAGNOSIS

A) INFECTION

A diabetic foot ulcer is considered to be infected when it has suppuration or two or more inflammatory signs are present (erythema, heat, pain, induration or tenderness). Diagnosis of underlying osteomyelitis requires compatible imaging tests.

1. Port of entry

DF infection almost invariably occurs in patients who suffer an ulcer more or less time since diagnosis or have suffered an acute lesion in the foot with rupture of the skin barrier[93]. This means that almost always we can identify what was site of entry of the organisms developing the infection. The first phase of diagnosis is to establish if the ulcer is neuropathic or neuroischemic, which is essential to determine the prognosis and propose the treatment, and may be done by performing a clinical examination (table 3)[94]. A major cause of infections, often severe, of the lower limb is overinfection by gram-positive cocci from skin fissures in the interdigital spaces in turn infected by dermatophytes[95]. The diagnosis of the cause of the ulcer is essential in order to plan treatment and enhances healing of the ulcer.

Ulcers appearing on the back of the interphalangeal joint of claw or hammer toes, on the most prominent area of bunions on the 1st or 5th toes, on the lateral margins, or on the back of the foot, are due to pressure and repeated friction in this area usually induced by footwear. In the areas where a bone protuberance has occurred as in neuropathic ulcers that appear on the sole of the foot (plantar perforating disease), on the tip of the claw or hammer toe, in the area of the prominence of the metatarsal heads, in the midfoot after it has incurred a "rocking chair foot" in Charcot joint disease, ulcers occur due to the increased pressure generated while walking.

In diabetic patients with neuropathy and loss of protective sensation, penetrating wounds in the sole of the foot can also occur and go completely unnoticed, until signs of infection appear later. These lesions occur mainly due to mechanical and thermal aggressions or by iatrogeny (podological interventions on skin callosities). They may also be due to interdigital lesions from onychomycosis or onychocryptosis.

Ischemia is another cause of diabetic foot ulceration, often affecting the toes and heel. Infection of these ulcers progresses rapidly and often poses a serious risk of amputation. The inflammatory reaction is inadequate, since its capacity of vasodilatation, edema formation and leukocyte infiltration is decreased. The microorganisms that develop in the necrotic foci proliferate uncontrolled because of the low leukocyte phagocytic activity, so rapidly progressing infections are particularly common with a trend to necrosis. In addition, ischemic ulcers have a very low tendency to healing because this process entails metabolic needs much higher than those needed to maintain skin integrity, so its natural tendency is to gangrene.

Depending on the initial lesion and its anatomical location, infection may evolve according to several clinical forms. On the skin and soft tissue two forms are distinguished: 1) superficial (dermis and epidermis) such as cellulitis and erysipelas; and 2) deep, such as necrotizing fascitis located in the subcutaneous cellular tissue, and myonecrosis and abscesses occurring in the deep fascia and muscle. They also can extend to the underlying joints (arthritis) and bone (osteomyelitis).

In the presence of a hot and swollen DF with ulcerated lesions, it is necessary to establish the differential diagnosis between acute Charcot neuroarthropathy and an infectious conditions, including cellulitis, plantar abscess, and osteomyelitis[96]. Other diagnostic possibilities to rule out would be gout attack, arthritis, and deep venous thrombosis. In addition to clinical assessment, imaging tests such as plain X-ray, CT and particularly MRI may be required to establish the diagnosis, as well as echo-Doppler to rule out venous thrombosis.

2. Colonization *vs* infection

All chronic ulcers over time and irremediably end up being invaded by microorganisms forming part of the microbiota of the surrounding skin (*Staphylococcus* spp and *Streptococcus* spp), and later from any other source.

The simple presence of bacteria or any other pathogen is called contamination. However the ulcerative bed, rich in protein and other nutritive substances, constitutes a good broth for the microorganisms to reproduce in, leading to the phenomenon of colonization. The following step after colonization is infection. It is the tissue invasion of the microorganisms that triggers an inflammatory response with the appearance of the classical local signs and purulent secretion with or without systemic clinical manifestations. The reason why colonizing bacteria acquire the ability to invade tissue is not fully understood[97]. The bacterial burden appears to be involved. It is has been noted that there may be a critical point ($\geq 10^5$ cfu/g of tissue), which may be influenced by the type of microorganism and the status of the host (degree of immunosuppression), from which the change from colonization to infection would

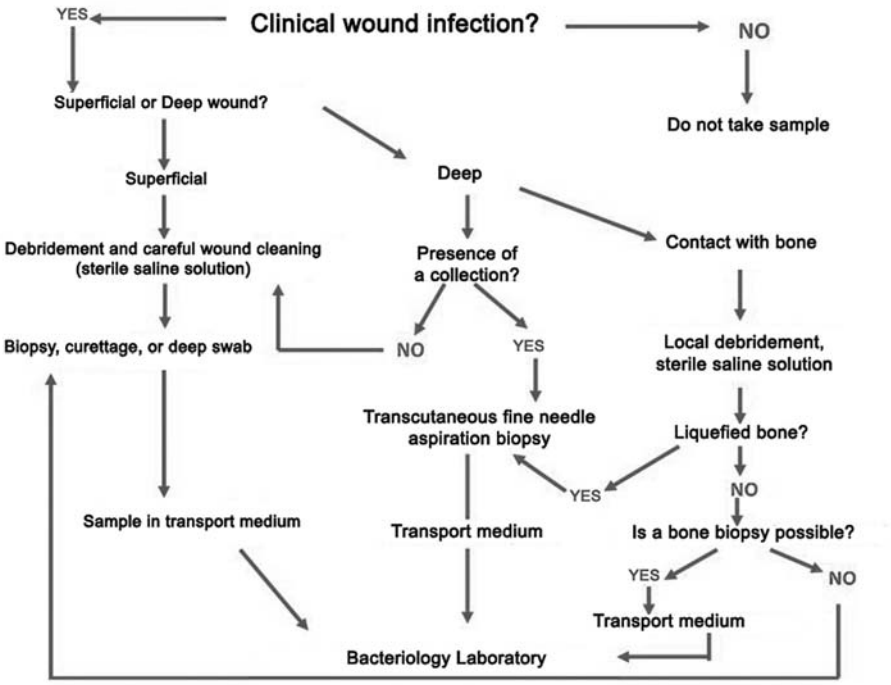

Figure 4 | **Sample collection in diabetic foot infections[102].**

be made. This is the so-called critical colonization[98].

The clinical significance of bacterial colonization has always been questioned[99]. However, the persistence of a subinfective bacterial density in an ulcer and toxins caused by microorganisms may delay tissue healing and cause some signs not attributed to infection such as the presence of friable granulation tissue and a serous type secretion[100]. In cases where the classical signs of infection are not seen but healing does not follow a favorable course, the performance of quantitative cultures from ulcer tissues may be indicated, with the purpose of detecting that critical colonization which would explain the inadequate course[101]. This novel microbiological concept of critical colonization, with all its current limitations, has also changed the prescription of antimicrobial treatment of chronic ulcers. To date, the use of antibiotics was only indicated for infections as such, but today it may advisable to consider it in ulcers with delayed healing not explainable by other causes, provided that the quantitative cultures are significant, even at cost of overtreating some patients. In short, critical colonization would be a state between colonization and infection, that would explain certain situations up to now unclear and that may influence the indications of antimicrobial treatment.

3. Sample collection

Standard colonization of chronic ulcers makes microbiological diagnosis only indicated when there are criteria for clinical infection. Collection of an adequate sample is decisive for microbiological diagnosis to be useful. Culture of nonrepresentative samples provides results without value that can lead to inadequate treatments. Tissue sampling should include those involved in infection and avoid surface material that can reveal only colonizing flora. The available options are a biopsy, curettage, percutaneous aspiration, and collection with a swab (figure 4). If infection us severe, with systemic manifestations, blood cultures should be taken and processed[47].

The biopsy is performed from the base of the ulcer, removing as much as possible superficial colonizer bacteria. For this, after performing surgical debridement, cleansing with a gauze soaked in normal saline is done. Optionally, after debridement, disinfection can be achieved with a disinfectant which is then removed with normal saline[102]. In osteomyelitis, bone biopsy is the reference sample. It can be taken by open surgery or by imaging-guided percutaneous aspiration (fluoroscopy or computed tomography). It is performed outside wounds or ulcers to avoid contamination by the flora colonizing them. The first method provides fewer negative results. Patients should have ideally been without receiving antibiotics from 2 to 4 weeks before taking the biopsy, because antimicrobials may remain for prolonged periods in bone. However, sometimes the clinical condition prevents treatment discontinuation for so long, so the culture should be interpreted in the context of the clinical condition[55].

Curettage is performed by preparing ulcer as described for the biopsy. If it is chosen to disinfect the base of the ulcer, a 50% povidone-iodine solution may be used which is removed with normal saline solution. After leaving to dry, a tissue sample (1-1.5 mm^3) is taken from the base of the ulcer with a sterile curette. This procedure is superior to the swab, avoids colonizers, and allows for recovery of more anaerobes[103].

Percutaneous fine needle aspiration may be used in cases of cellulitis, purulent collections, and osteomyelitis when liquefied bone is present (requires imaging guiding). It requires skin disinfection and subsequent cleaning with normal saline before being performing.

Although it would be desirable not to collect samples with swabs, it is he most commonly used method because of its simplicity and wide availability. It may be used in ulcers and cavities that can be opened. Prior preparation is the same as for biopsy. The sample is taken from the base of the ulcer rotating the swab with a certain pressure to extract the tissues. If is desired to make a quantitative or semiquantitative study, rotation is done in an area of 1 cm^2 for five seconds[104]. Tissue samples are initially preferred as more microorganisms are recovered[47,56,105]. When agreement between various types of collection are compared quantitatively, curettages give better results than swabs or aspirates. Swab cultures tend to overestimate and aspirate cultures to underestimate the number of isolates present in deep tissues[106]. Cultures taken with swabs compared to those performed with deep tissue samples have shown a low sensitivity (49%), specificity (62%), positive probability (1.1), and negative probability ratio (0.67)[107]. Within a multidisciplinary approach to managing DF infections in which the key issue was to eliminate superficial collections with swabs, to avoid as far as possible deep collections with swabs, and mainly to use tissue samples, significant changes were observed in the period from 2003 to 2007 from a microbiological, therapeutic and economic perspective[60]. There was a significant reduction in the number of species of bacteria per sample (a change from a mean of 4.1 to 1.6). The prevalence of microorganisms considered colonizers fell drastically from 23.1% to 5.8% ($p < 0.001$). More gram-positive organisms were isolated, particularly *S. aureus*, fewer gram-negative pathogens, both enterobacteria and *P. aeruginosa*, and anaerobic isolates remained at low levels (5%). Likewise there was a decreased prevalence of multiresistant microorganisms (35.2% versus 16.3%) and MRSA (52.2% versus 18.9%) ($p < 0.001$). The impact of these measures represented a saving of € 14,914 related to microbiological work and € 109,305 due to the reduction in prescription of broad-spectrum antibiotics.

After specimens are obtained, they should be placed in a transport system for tissue, curettages, or swabs that permits survival of anaerobic bacteria. It is essential that specimens are identified appropriately and shipped quickly to the laboratory keeping them at room temperature.

Recommendations:

1. In diabetic foot infections, the sample for ideal microbiological study is deep tissue biopsy.

2. Other options include curettage and percutaneous aspiration.

3. Swab samples, if done, should be taken from the deep portion of the ulcer, by pressing the swab over on tissues to extract them.

4. Microbiological diagnosis

Microbiological studies should identify or detect antigenically or genomically the microorganisms present in the samples, check their sensitivity to antimicrobials in a reliable manner and provide data, if possible, to guide on the clinical implications of these microorganisms. All should be done as rapidly as possible so it has an effective impact to adjust an empirical treatment.

Samples will be processed in the laboratory with the greatest possible speed. Gram staining has little correlation with the culture but may allow for assessing quality of the sample by the presence of PMNs. Anaerobic bacteria will always be sought. Processing will be based on traditional microbiological methods. The standard test is seeding on enriched, selective and differential (including chromogenic) media that allows not only to recover the species involved but even to make a presumptive identification, and in some cases to assess sensitivity to antimicrobials (chromogenic media for MRSA). A qualitative, quantitative, or semiquantitative technique may be used[108]. The media for anaerobes will be incubated in a jar or a chamber. After identification, sensitivity to antimicrobials is determined in the safest and fastest manner possible. This point is crucial because it allows for adjusting empirical treatments and modifying them when they fail.

On the near horizon is the introduction of molecular techniques [real time PCR, microarrays, fluorescent in situ hybridization (FISH), DNA (rRNA) sequencing] for the microbiological study [etiology, pathogenicity and sensitivity to antimicrobials] of DF infections. These techniques will allow to establish routine detection of the causal agents, their virulence determinants, and their sensitivity to antibiotics in hours and not in days as occurs with traditional microbiological tests. In addition, another of its benefits is that it is more sensitive[59,62,109].

It is not easy specify the clinical significance of the bacteria isolated[110]. It is clear for highly virulent microorganisms, such as *Streptococcus pyogenes*, but not for most other recovered species that are usually opportunistic or commensal pathogens. To address this issue, various solutions have been proposed. A bacterial count above 10^5 cfu per cm^2 or gram marks difference between colonization and infection[111], so quantitative methods to determine the bacterial burden are very useful, but since they are very complicated and take a long time they are not routinely performed in microbiology laboratories. A semiquantitative method has also been shown to be useful[104,112]. They are simpler, but not so much as to enjoy generalized acceptance. This is why it is necessary to have microbiological criteria that help assessment of qualitative results which are those usually received by the microbiology clinician. Bacteria such as *S aureus*, β-hemolyt-

ic streptococci, enterobacteria or anaerobes should be given importance from the start[113], while the rest should only be considered when they are found in a pure culture or isolated repeatedly[110,113]. The development of molecular procedures can contribute to this task. A PCR method has been developed demonstrating the presence of 5 virulence genes (*sea, sei, lukE, hlgv* and *cap8*) present in a significantly greater percentage in *S. aureus* isolates from grade 2-4 ulcers than grade 1[62]. However, no species should be disregarded[48] given the polymicrobial nature of the biofilm of chronic diabetic foot ulcers[72]. Microbiological studies should be repeated in the case of an unfavorable course.

5. Role of biomarkers in diagnosis, prognosis, and treatment monitoring

Serological markers of inflammation, such as increased erythrocyte sedimentation rate and C-reactive protein (CRP) in the past or procalcitonin (PCT) of more recent use, may be of value to distinguish between colonization and infection, to suspect the presence of more severe infection and/or osteomyelitis, to determine the prognosis in severe forms and particularly to assess response to treatment[114-116]. Although there is no detailed information on their use in the DF infection, the information may be extrapolated from other areas of the infectious diseases particularly severe bacterial infections whatever the infectious site causing the disease.

CRP is an acute phase protein released by the liver cells following their stimulation by mediators of inflammation such as interleukins (IL-6, IL-8). Its peak level is reached in plasma about 48 hours from onset of the disease. Its plasma levels may remain elevated for days even after elimination of the infectious focus and are increased in different infectious diseases. The mean plasma concentration in healthy adults is 0.08 mg/dL. Levels >20 mg/dL in patients with consistent clinical signs would point to the bacterial origin of the condition, and levels <0.5 mg/mL are associated with a probability of bacteremia/sepsis below 1-2% (except in patients with liver disease)[117]. CRP showed no correlations with the severity of host response or difference between survivors and nonsurvivors in sepsis conditions. It also has poor predictive value and has not demonstrated potency as a severity marker. Serial CRP measurement may be useful for early diagnosis of nosocomial infections in the ICU (increase in levels >5 mg/dL or >25% of previous value) as it is less expensive, more accessible, simpler and more rapid than other markers[118]. CRP plays a significant role in orientation of antibiotic therapy in localized diseases, has a greater diagnostic value than temperature increases in diagnosis of infection, and therefore is proposed in some texts as a routine test to be performed at initial evaluation in patients with suspected sepsis[119]. However, its efficacy appears to be better established in the follow-up and monitoring of response to antibiotic therapy, so some authors recommend performance of serial measurements, where it has been shown that its use is also sensitive as an indicator of resolution of sepsis in microbiologically proven cases[117,120,121]. CRP is a 116 amino acid peptide, prohormone of calcitonin, whose blood levels are increased in septic patients, selectively in bacterial infections, by inhibiting the cytokines and endotoxins released by them, the final step in the synthesis of calcitonin. It has a half-life of 24 hours. CRP elevations are detected in the 2 hours after endotoxinemia or bacteremia.

Detection of PCT has been confirmed as a sepsis marker in severe infection in multiple trials. Compared to other diagnostic tests, PCT has the advantage that it increases earlier and is more specific in significant bacterial infections (as compared to CRP and leukocytosis), and low blood levels of PCT rule out bacteremia. As for cut-off points, patients with levels lower than 0.5 ng/mL are unlikely to have sepsis or septic shock, while measurements above 2 ng /mL identify high-risk patients and concentrations above 10 ng/mL are associated with patients with organ failure. The foci of localized infection without systemic inflammation do not show an increase in PCT levels. With recent PCT measurement techniques, bacterial infection can be excluded with a high negative predictive value. Consequently, it appears that PCT is a promising diagnostic test for monitoring progression and prognosis of bacterial diseases[122,123]. In addition, it can be concluded that guidelines for use of antibiotics using PCT values lead to shorter exposure to antibiotics with no harmful effects on the patient. This has advantages both in terms of the ecological cost (selection of multiresistant bacteria when using broad-spectrum antibiotics) and the economic cost[124].

6. Osteomyelitis

The spread of infection by contiguity from soft tissue may affect the underlying bone. Although bone infection may be due to penetrating wound or an ischemic ulcer, it usually occurs in a neuropathic ulcer that becomes increasingly deeper and eventually exposing bone. Thus, most commonly involved sites are the toes, the metatarsal heads, and the calcaneus. Ulceration or infection of the overlying soft tissue reaches the periostium and cause its destruction, which devitalizes the superficial cortex (osteitis). When infection reaches the Haversian system, invasion of the medullary bone and bone marrow occurs, where it spreads rapidly (osteomyelitis). Lesion of the periostium induces necrosis of underlying bone (sequesters) and periosteal reaction forming new bone (involucrum). Host response limits the infection in an area of the bone facilitating separation of the sequesters, which may be found at the base of the ulcer or be eliminated as small fragments to the skin surface, sometimes stopping the infectious process with the appearance of healthy granulation tissue, with then possible cure. In contrast, if bone infection persists new areas of bone necrosis occur with spreading infection to the surrounding soft tissue. Persistence of infection in bone is often associated with adherence of microorganisms to sequester in mono- or polymicrobial communities (biofilms) containing phenotypes that are resistant to host defenses and most antibiotic agents[125]. Approximately 10-20% of all diabetic foot soft tissue infections classified as mild are associated with osteomyelitis, while this may be occur in up to 50-60% of moderate/severe infections[126].

A wide consensus on the best diagnostic strategy is not available for DF osteomyelitis (DFO). An issue complicating the diagnosis of DFO is the difficult differential diagnosis with Charcot neuroosteoarthropathy, very common in DF, as it may cause noninfectious bone changes difficult to distinguish from those attributable to osteomyelitis. Clinically, it may occur with acute signs of inflammation in the adjacent soft tissues, either limited to appearance of a red, hot and swollen toe ("sausage toe") or affecting the entire foot. In the latter case, it will be necessary to differentiate it from acute Charcot neuroarthropathy. It may also present subacutely or chronically, forming fistulas, from bone to the skin, or preventing cure of the overlying ulcer. In this case, typical clinical signs include an ulcer over a long-standing deep bone prominence, which does not cure after 6 weeks of treatment, in the absence of ischemia and despite adequate antibiotic therapy, adequate local care and pressure unloading over the area. In a group of patients with infection threatening the limb, where the probability of DFO was calculated at approximately 66%, it was seen when bone was palpated at the base of the ulcer on examining it gently with a metallic lancet, a positive predictive value of DFO was 89%[126]. However, in a later study, including a group of patients in which the incidence of DFO was 20%, the positive predictive value of the bone probing test was only 53%[127]. Recently, this test has been assessed in a group of 199 patients with DF infection, obtaining a positive predictive value of 57%, and negative predictive value of 98%[128]. In other words, a positive test has poor diagnostic capacity but a negative test makes the diagnosis of DFO unlikely.

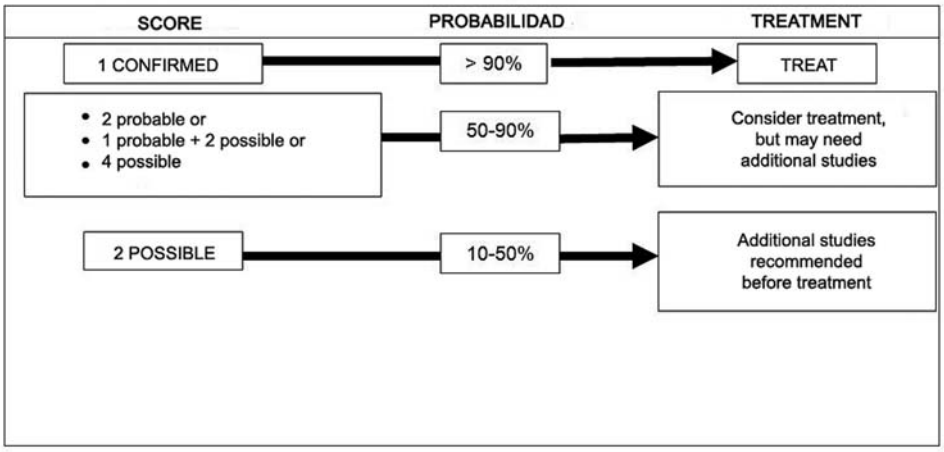

Figure 5 | **Osteomyelitis diagnosis score[125].**

Table 4 | **Criteria for diagnosis of osteomyelitis[125].**

CONFIRMED DIAGNOSIS ("Beyond a reasonable doubt")
- Histology + and bone culture +
- Purulence in bone on surgical examination
- Atraumatic release of bone fragments eliminated from an ulcer.
- Intraosseous abscesses on MRI.

PROBABLE DIAGNOSIS ("More likely than not")
- Visible cancellous bone in an ulcer.
- MRI: bone edema with other signs of OM.
- Bone sample with positive culture but histology is negative or absent.
- Bone sample with positive culture but histology is negative or absent.

POSSIBLE DIAGNOSIS ("Low probability")
- Plain X-ray: Cortical destruction.
- MRI shows bone edema or cloaca.
- Positive probe-to-bone test.
- Visible cortical bone.
- ESR >70 mm with no other plausible explanation.
- Ulcer that does not cure despite adequate unloading and perfusion after 6 weeks or ulcer lasting more than 2 weeks with clinical evidence of infection.

Plain X-ray may be normal in the early stages, though those performed a few weeks later may show periosteal thickening and bone destruction. Any foci of bone destruction adjacent to an ulcer should be considered potentially DFO, while not proven otherwise. Neither the bone probing test nor plain X-ray can safely exclude the diagnosis of DFO[129]. A Tc^{99} bone scan is considered poorly specific and is not recommended and scintigraphy with labeled leukocytes may be used when MRI is contraindicated. Magnetic resonance is most useful imaging technique for diagnosis of DFO, both to assess the extent and involvement of associated soft tissues, and for planning surgery[130,131]. However, it does not allow to distinguish between infection and Charcot neuroosteoarthropathy.

The gold standard to diagnose DFO is isolated bacteria in a bone sample, adequately obtained to avoid contamination, together with histological findings of inflammatory cells and osteonecrosis. Whenever possible, antibiotic therapy should be discontinued 2 to 4 weeks previously, but the clinical condition may prevent this discontinuation, so it culture should be interpreted in this clinical context. When the clinician suspects or wishes to rule out the diagnosis of osteomyelitis, the first step is to obtain a plain X-ray. If on the initial X-ray there are signs suggesting of osteomyelitis, specimens should be obtained for microbiological culture and then antibiotic treatment should be started. If the X-ray is not diagnostic, it should be treated

for 1-2 weeks as a soft tissue infection and if suspicion persists, repeat the X-ray at 2-4 weeks. If the X-ray is consistent with but not diagnostic of osteomyelitis, it can be decided to perform MRI to make or rule out the diagnosis or start an empirical treatment for an additional 2-4 weeks.

Bone biopsy is recommended when the diagnosis of osteomyelitis remains doubtful after imaging tests, and if it demonstrates osteomyelitis but the etiological agent and/or its sensitivity to antibiotics is not known. When the affected bone is in the midfoot or hindfoot, performing bone biopsy is especially recommended before performing treatment, because if it fails, the probability of major amputation is much higher than in forefoot lesions. Cultures of fistula trajectories show little correlation with bone cultures of, and are therefore not recommended. It is important to identify the causal agent of the infection, and also its sensitivity to antimicrobials, as it has been shown that antibiotic treatment, based on the culture of a bone sample is associated with better clinical results (80% remission) than antibiotic therapy based on the culture of a sample obtained with swab of adjacent soft tissues (50% remission). In bone biopsy, at least two specimens should be obtained if possible, for pathological and microbiological study. Complications of this technique have not been published so it is considered a quite safe procedure. Unfortunately, bone biopsy is not widely used. In most cases, clinicians base diagnosis more on symptoms and signs, combined with imaging techniques and laboratory data (increased erythrocyte sedimentation rate and CRP).

The International Working Group on the Diabetic foot (IWGDF) has recently proposed a scheme that groups different criteria according to 4 categories of probability of establishing the diagnosis of DFO125 (table 4)(figure5). This diagnostic approach has not been validated in practice, but represents a leap forward in terms of standardization of diagnosis and the treatment decision[132].

Recommendations:

1. FD osteomyelitis (FDO) is usually by contiguity.

2. Persistence of FDO is associated with biofilms.

3. Magnetic resonance imaging is the most useful technique for diagnosing DFO.

4. The gold standard for diagnosing DFO is bacteria isolated from a bone tissue sample, with histological findings of inflammatory cells and osteonecrosis.

5. Bone biopsy should be performed if diagnostic imaging is doubtful.

B) ISCHEMIA

Evaluation of the arterial perfusion is an essential component of diagnosis of diabetic patients with an infected trophic lesion in the foot. Diagnosis of critical ischemia associated with an infected diabetic foot requires confirmation by objective methods[133].

1. Clinical Examination

The history must be directed to the main characteristic symptoms of chronic lower limb ischemia, particularly a history of intermittent claudication and pain at rest. The physical examination should include palpation of pulses (femoral, popliteal, tibial and pedal) as well as perception of any murmurs or thrills at the femoral level.

2. Supplemental tests

The ankle/brachial index (ABI) should be routinely performed in all diabetic patients, for which a sphygmomanometer and a continuous Doppler device is usually used. The ABI is the ratio of systolic pressure ratio between the ankle and the arm, and is normal for any value in the range from 0.9 to 1.4. An ABI below 0.9 indicates obliterating arterial disease of the lower extremities and its hemodynamic impact is directly proportional to the reduction in the index. Thus, for instance, ischemic ulcers usually occur at ankle systolic pressures of 50-70 mmHg, and pain at rest at 30-50 mmHg. ABIs higher than 1.4, in contrast, suggest arterial incompressibility, usually by arterial calcification, and do not allow to evaluate the presence of underlying occlusive arterial lesions. It is not known whether arterial calcification plays a role in obtaining falsely elevated ABI values but within the normal range (0.9-1.4) or pathological (<0.9).

Digital pressure on the first toe (critical level <50 mmHg), pulse volume recordings and transcutaneous partial oxygen pressure (critical level <30 mmHg) are alternative diagnostic methods to the ABI when it is suspected that the result of this is artifacted by arterial calcification.

Echo-Doppler is a non-invasive examination providing morphological and hemodynamic information about the different stenotic and occlusive lesions located in the affected limb of an infected diabetic foot. Unlike the ABI, echo-Doppler does not provide any information on the overall hemodynamic impact to which the foot or the trophic lesion are subject, and is indicated, as well as other morphological examinations, when the possibility of revascularization is already being considered.

Angio-MRI and angio-CT represent morphological examinations (anatomical) to report lesion topography and allow to establish the most appropriate endovascular or surgical revascularization strategy. Angio-MRI and angio-CT are minimally invasive (vein puncture) but their result is unreliable in small-caliber vessels (MRI and CT), for example, below the knee, and in the presence of arterial calcifications (CT) or intraluminal stents (MRI); also their use is limited in cases of allergy to contrast media or renal failure.

Digital intravenous subtraction angiography (DIVAS) is the gold standard in morphological diagnosis of obliterating arterial disease associated with an infected diabetic foot. Like other morphological examinations, it does not provide hemodynamic information and, therefore, cannot be used to establish the diagnosis of critical ischemia in patients with an infected diabetic foot. It requires arterial puncture and administration of iodinated contrast, so it is usually reserved as a prior or simultaneous examination to endovascular or surgical revascularisation.

Table 5 Empirical antibiotic treatment of diabetic foot infections.

Infection	First Choice	Alternative
Mild	Oral amoxicillin-clavulanic acid	Oral levofloxacin or moxifloxacin
Mild-Moderate		Oral clindamycin
		Oral cotrimoxazole
		Oral linezolid
Moderate-Severe	IV ertapenem	IV amoxicillin-clavulanic acid
	±	or
	IV daptomycin or IV linezolid or IV glycopeptide[1]	IV 3rd generation cephalosporin + IV metronidazole
		or
		IV fluoroquinolone[2] + IV metronidazole
		or
		IV piperacilln-tazobactam[3]
		or
		IV imipenem or IV meropenem[3]
		±
		IV daptomycin or IV linezolid or IV glycopeptide[1]
Severe	IV imipenem or meropenem	IV tigecycline
	or	+
	IV piperacillin-tazobactam	IV fluoroquinolone[2] or IV amikacin
	+	
	IV daptomycin or IV linezolid or IV glycopeptide[1]	

[1] Suspected MRSA
[2] Ciprofloxacin or levofloxacin
[3] Suspected *P. aeruginosa*

Recommendations:

1. In all diabetic foot ulcers, palpation of pulses should be performed to detect the presence of ischemia.

2. Hemodynamic methods (ankle/brachial index, digital pressure) are generally used to quantify the degree of ischemia.

3. Morphological methods (echo-Doppler, angio-CT, angio-MRI, arteriography) should be used for planning the surgical strategy if revascularization is to be performed.

TREATMENT

ANTIBIOTICS

There are no data supporting antibiotic treatment of chronic ulcers, even with a positive culture. Antibiotic treatment will be indicated if there are clinical criteria of local or systemic infection[47]. Laboratory data are of limited value for diagnosis of infection, except in the case of osteomyelitis[61].

Antibiotic treatment of diabetic foot infections is determined by ischemia restricting antibiotic arrival to septic focus, impairment of leukocyte function, and potential renal failure in these patients[90]. Ischemia and leukocyte abnormalities make response of infection to treatment poorer in diabetics and they may worsen rapidly in hours or a few days[90,134]. Functional neutrophil defects in diabetics make it advisable to use bactericidal antibiotics and for an extended period, ischemia determines the use of high doses and the prevalence of renal failure leads to avoidance of nephrotoxic drugs, such as aminoglycosides, vancomycin and amphotericin B[47,90,134-136].

The severity of the infection, the duration of the lesions, and the risk factors related to the occurrence of bacterial resistance, together with local sensitivity patterns, determine the

selection of empirical antimicrobial treatment, the site where it is performed and the administration route. However, gram-positive cocci are the predominant pathogens in any circumstance, so they should always be covered. MRSA should be considered in the following circumstances: 1) colonization or previous infection of the patient by this microorganism, 2) prevalence of MRSA infection at the site or hospitalization unit over 10%, and 3) if two or more of the following criteria are met: a) hospital admission in the past year or patient is from a nursing home or healthcare center with endemic MRSA disease, b) treatment with a fluoroquinolone in the previous 6 months, c) patient over 65 years of age, and d) patient in dialysis program for chronic renal failure[137]. The same measures should be taken for ESBL- producing *E. coli* if: age over 65 years, patient is a woman, hospitalization in the previous year, recurrent urinary tract infection, and prior use of fluoroquinolones. Diabetes itself is an established risk factor for this microorganism[138]. Chronic ulcers receiving multiple treatments is also a risk factor for *Enterococcus* spp, CNS, *P. aeruginosa*, particularly in humid and macerated ulcers, and other non-fermenting gram-negative bacilli[49].

Based on these criteria, home oral treatment with amoxicillin-clavulanic is recommended for minor infections, and with cotrimoxazole or linezolid if there is risk of MRSA or CNS. In patients allergic to beta-lactams, levofloxacin, moxifloxacin, clindamycin or cotrimoxazole may be used, except when there is suspected streptococci. Duration of treatment in these cases can be sufficient with 7 to 14 days[47,91]. This same regimen is valid for moderate-mild infections, though monitoring should be closer due to the increased risk of a poor disease course.

For moderate-severe infections with risk of losing the limb, hospitalization and broad-spectrum intravenous treatment is advised for two to four weeks. Because of their high prevalence, antibiotics must be active against the following organisms: aerobic gram-positive cocci (*Staphylococcus* spp and *Streptococcus* spp), aerobic gram-negative bacilli (Enterobacteriaceae) and anaerobes (*Streptococcus* spp, *Peptostreptococcus* spp and *Bacteroides* spp)[47,91,136]. In this situation, ertapenem, a third-generation cephalosporin[71] plus metronidazole[139] or amoxicillin-clavulanate may be used[140]. If there is a high suspicion of the participation of *P. aeruginosa*, piperacillin-tazobactam may be administered or an antipseudomonal carbapenem[141]. Ertapenem, due to the high risk of ESBL-producing *E. coli* in these patients, its ease of use (monotherapy and single daily intravenous and intramuscular dose), soft tissue penetration and favorable clinical experience, appears to be the first choice for parenteral administration[71,142]. To this should be added daptomycin, linezolid or rarely vancomycin (if patient has no renal failure) if there is a high probability of MRSA.

Finally, in severe infections with systemic impact and life-threatening[143,144], all possibilities should be covered with beta-lactams with antipseudomonal activity (carbapenem or piperacillin-tazobactam) combined also with daptomycin, linezolid or vancomycin if there is a risk of MRSA. In the case of allergies to beta-lactams, the alternative requires the use of tigecycline combined with a fluoroquinolone (ciprofloxacin or levofloxacin) or amikacin (table 5).

After determining the causative agent and its sensitivity, adjustment of antimicrobial treatment will then be done[145]. In moderate-severe and severe MRSA skin and soft tissue infections, daptomycin is one of the treatments of choice, at doses of 8-10 mg/kg in the case of ischemia, due to the risk of bacteremia, its rapid bactericidal action, activity in biofilms of chronic ulcers and vegetative bacteria and lack of toxic effects on the kidney[143,146-150]. Linezolid is another excellent choice because of its tissue penetration, although it is bacteriostatic and in the event of renal failure long-term treatment accentuates thrombocytopenia[151-153], and it also has the advantage that it can be administered both parenterally and orally. Vancomycin has a slower bactericidal action than daptomycin and requires use of high doses when the MIC for MRSA is $\geq$1, which is not very recommended in diabetic patients with potential or manifest renal failure[68,154-156].

In infections by ESBL-producing *E. coli*, the treatment of choice are carbapenems, including ertapenem, particularly if *P. aeruginosa* is not present, to avoid exerting selective pressure on this microorganism. Because of its activity against the anaerobes involved in this type of infections, its rapidly bactericidal action little influenced by the inoculum effect[157,158], and its confirmed clinical efficacy and safety in the DF[71,159], its use should be considered in these infections.

Although the pathogenetic role of *P. aeruginosa* is not clear, since in mixed infections in which it was isolated in the SIDESTEP study, a similar clinical response was reported between ertapenem and piperacillin-tazobactam[71], we should consider both its empirical and targeted treatment in those infections that are life-threatening and/or where there are risk factors for its presence such as long-term chronic ulcers[49], exudative ulcers or those treated with wet bandages or hydrotherapy[4,160-163], in warm climates in people with feet that sweat due to inadequate shoes and no use of socks[164], and after having received antibiotic treatment in the past month[50].

Monotherapy with beta-lactams at high doses, namely, a carbapenem, piperacillin-tazobactam or fourth-generation cephalosporin's[165] or quinolones (particularly in patients allergic to penicillin) is as effective as combined treatment with aminoglycosides and safer, according to data from nonrandomized clinical series[47,166], so that there are no reasons to maintain the traditional recommendation of combined treatment with beta-lactams and aminoglycosides. The clinical efficacy of other combinations, beta-lactams with fluoroquinolones, or macrolides, or rifampin has not been evaluated. However, this should be guided by the results of the antibiotic susceptibility testing and local epidemiology[166]. Administration of piperacillin-tazobactam[167] or carbapenem in continuous or prolonged infusion[168,169] improves clinical results versus the standard administration in regular doses, reducing mortality and hospital stay in patients with severe infections by *P. aeruginosa*. The basis of this benefit is the favorable modifica-

tion of pharmacodynamic and pharmacokinetic parameters[4]. Intravenous and/or topical colistin, alone or in combination with rifampin or imipenem, is the only option available for panresistant *P. aeruginosa* infections, also reported in these infections, although clinical experience is limited[170,171].

The antimicrobial drugs having a greater *in vitro* activity against *A. baumannii* are carbapenems, sulbactam, aminoglycosides, rifampin and tigecycline. The carbapenems, except for ertapenem, and sulbactam are considered antimicrobials of choice against severe infections by *A. baumannii*[166]. The sensitivity of *A. baumannii* to antimicrobials is different between countries, between centers, and between areas of the same hospital. In our country, 41% of the clinical isolates of *A. baumannii* are resistant to carbapenems and stay in an ICU is an independent risk factor for this[166]. Intravenous colistin, combined or not with rifampin, is the only alternative to infections caused by *A. baumannii* by strains resistant to the previous drugs[172]. Tigecycline is active *in vitro* against strains resistant to carbapenems, although clinical experience is limited[172]. Finally, the development of multiple resistance has rescued medications that were lost in oblivion, such as fosfomycin[173], with results similar to other drugs, so in these cases they are again an appropriate therapeutic option[174].

Failure of a correct antibiotic treatment may be due to the development of resistance, overinfection or extension to bone. We should remember that hospitalized patients and those previously treated with broad-spectrum antibiotics over a long period usually have resistant bacteria.

SURGICAL MANAGEMENT OF INFECTION

When infection affects superficial layers, the local treatment with debridement and cleaning is usually sufficient. However, in the presence of a severe infection, a more aggressive surgical debridement is required. This type of surgical debridement should be done in the presence of abscesses in deep locations, necrotizing fascitis, gas gangrene, extensive involvement of soft tissues or compartment syndrome[175].

The objectives of this treatment would be to prevent progression of infection, to preserve life of the patient, to preserve the limb and to preserve limb function. Therefore, before debridement, we must assess perfusion of the foot. In absence of ischemia, debridement will be extensive to remove all necrotic tissue present at a single time, since healing occurs rapidly in the absence of ischemia. In contrast, in the presence of ischemia, abscess drainage and debridement of necrotic tissue will be performed, leaving doubtful areas to be removed later, if necessary, after performing revascularization.

Before performing any incision, we should consider the compartments of the foot that may be affected and that may be involved in the debridement. Due to the possibility that it will be necessary to perform partial amputations if osteomyelitis is found, initial incision will coincide us with the incision required for this amputation. Thus, we will plan the incision with a view to the subsequent treatment we will perform to preserve function foot as far as possible[176]. Therefore, before entering the operating room, the patient should be informed of the treatment he/she is to undergo and sign consent including the need for possible minor amputations or expansion of the initially planned level of amputation.

In the operating room, an incision will be made to reach healthy tissue proximal and distal to the wound. The depth of the incision should reach a plane of fascia or muscle free from infection. The presence of fistulas and/or cavities will be explored, abscesses will be drained and debridement will be performed, taking this opportunity to collect samples for microbiology and a bone biopsy for histological study. After debridement, washing with saline or an antibiotic solution will be preformed (although there are no conclusive studies in diabetic foot)[176]. Washing may be done with saline-filled syringe or a pulse lavage system, isolating the limb with a wash bag to prevent aerosolization of microorganisms to the exterior[177]. After completing washing, closure will be planned with clean instruments which have not been used in debridement phase. Before closure, some authors use antibiotic impregnated beads (vancomycin, tobramycin or gentamycin)[178] to fill in the dead space that may remain and allow sustained antibiotic release in bone resection areas with osteomyelitis. Closure may be performed directly, by second intention or by deferred direct closure. Direct closure is not advisable in ischemic patients or in severe infections, so as be able to continue monitoring the wound bed during healing. Closure by second intention is the most frequently used; in the absence of ischemia, granulation tissue can cover the defect rapidly, if there are no other reasons that prevent it (hyperpressure areas or deformities). Deferred direct closure is performed using skin grafts or muscle flaps. It is the most commonly used in cases with great substance loss to enhance discharge.

The last alternative is amputation[179]. Amputations of a toe or transmetatarsals closed after revascularization allow for good function of the foot. Conversely, if there is great substance loss that prevents foot function, or if there are ulcers which have not healed despite patency of the graft or in patients with life threatening sepsis, infracondylar amputation should be considered. Supracondylar amputation is reserved for very frail patients unable to walk[160].

Recommendation:

Any infected ulcer should be debrided. If the ulcer is superficial, it will be sufficient a small curettage that removes necrotic residues and help to stimulate growth of margins. In cases of infections affecting deeper fields, debridement should be more aggressive and include resection of all necrotic and infected tissues, restore skin level of abscesses and minor amputations.

TREATMENT OF ISCHEMIA

The primary objectives of treatment of neuroischemic ulcer in DF are to relieve ischemic pain if present, to heal ulcers, to prevent loss of the limb, to improve patient function and

quality of life, and to prolong survival, without amputations. In some patients with severe comorbidities or with a very low chance of satisfactory revascularization, primary amputation may be more appropriate treatment.

Control of cardiovascular risk factors is essential in all diabetic patients with ulcer as in all patients with lower extremity obstructive arterial disease (LEOAD). Therefore, a multidisciplinary approach is appropriate for pain control, cardiovascular risk factors and other comorbid illnesses.

1. Medical Treatment of Ischemia

Pharmacological therapy or any other treatment for ischemia has greater chances of yielding results in patients who were asymptomatic before the appearance of foot lesions, and in patients with superficial lesions with higher perfusion pressures.

Cilostazol, a phosphodiesterase III inhibitor, may be used if there is associated intermittent claudication, provided the patient can walk, which depends on the location of ulcer and whether it has good pressure unloading[180].

Acetilsalycilic acid (ASA) and other platelet antiaggregant drugs (clopidogrel) are important for long-term treatment of LEOAD to reduce risk of atherothrombotic events[181]. Although they have been shown to have a beneficial effect on permeability of revascularization surgery and on progression of the femoral atherosclerosis, there is no evidence that these drugs improve the course of critical ischemia.

Neither heparin nor vitamin K antagonists have demonstrated efficacy in the treatment of critical ischemia and ulcers.

The prostanoids, drugs for parenteral use that prevent platelet and leukocyte activation and protect the vascular endothelium, improve healing of ischemic ulcers and reduce the number of amputations without increasing amputation free survival[182].

2. Analgesia

Pain control is essential to improve function and quality of life. Ideally, this relief is obtained through revascularization of the limb, but while this is being carried out and in cases where it is not possible, use of narcotics is often necessary.

Based on pain intensity, analgesia will be administered regularly instead of as needed and reinforcement of hygiene and postural measures such as placement of the extremity in gravity-assisted position.

Drugs to be used include acetaminophen, methamizole, or non-steroidal anti-inflammatory drugs, taking care in hypertensive patients and those with renal failure. Often these are insufficient and it is necessary to use weak opioids (tramadol, codeine) or major opioids (fentanyl, oxycodone or bupremorphne)[133].

3. Revascularization

The natural course towards amputation of a neuroischemic ulcer in DF makes revascularization indicated for saving the limb. Determination of the best revascularization method is based on the balance between the risk of a specific intervention and the degree and durability of clinical improvement expected from it. The results of revascularization depend on the extent of the disease in the arterial tract (inflow, outflow and diameter and length of diseased segment), the degree of systemic disease (comorbidities affecting life expectancy and influencing permeability of the revascularization technique) and type of procedure performed. In conclusion, the results of large clinical trials should be considered in the context of individual status of each patient and not forgetting that the results of revascularization procedures depend on both anatomical and clinical factors.

Table 6 TASC morphological stratification of infrapopliteal lesions.

Type A lesions
- Single lesions less than 1 cm in tibial or peroneal vessels.

Type B lesions
- Multiple focal stenoses of tibial or peroneal vessels, each less than 1 cm in length.
- One or two focal stenoses, each less than 1 cm in length, in tibial trifurcation.
- Short tibial or peroneal artery stenosis and femoropopliteal PTA.

Type C lesions
- Stenosis 1-4 cm in length.
- Occlusions measuring 1-2 cm in length of tibial or peroneal vessels.
- Extensive stenoses of tibial trifurcation.

Type D lesions
- Tibial or peroneal occlusions greater than 2 cm.
- Diffuse disease in tibial or peroneal vessels.

Atherosclerotic disease of distal arteries, associated with diabetes, can be found in combination with other proximal areas or as the predominant infrapopliteal disease. These patients usually remain asymptomatic thanks to an excellent collateral network; if they have clinical signs of critical ischemia is it because they have severe and extensive three-vessel disease and only 20-30% have a focal lesion with good run-off.

Morphologically, these lesions are characterized by a diffuse segmental involvement particularly of tibial vessels, where only 50% have a patent vessel to the foot, usually the peroneal artery. There is a high prevalence of long occlusions (>10 cm long) and proximal involvement is usually minimal at the iliac level (1%) and approximately 10% at the level of the femoropopliteal segment[183].

Revascularization by open surgery of occlusive disease of the distal arteries is carried out mainly by bypass with autologous material (preferably saphenous vein). In turn, endovascular surgery techniques mainly include percutaneous transluminal angioplasty (PTA), which may be combined with stenting, laser and plaque volume reduction techniques. The exponential increase of the use of these endovascular procedures, compared with open surgical revascularisation, is primarily due to the greater benefit with respect to the secondary risk of low percentages of morbidity and mortality associated with the percutaneous techniques. Mixed techniques (open + endovascular surgery) may be used.

Although specific stratified lesions are being modified, lesion morphology is not sufficiently specified in the TASC consensus classification of lesions. Type A lesions represent lesions with excellent results in endovascular treatment; type B lesions are those that provide sufficiently good results with endovascular methods to make this the approach of choice, unless open revascularization is required for another associated lesion in same anatomical area; type C lesions obtain superior long-term results with open revascularization and endovascular methods should only be used in patients at high surgical risk for open repair; type D lesions, in turn, have not achieved optimum treatment results with endovascular procedures to consider them a primary treatment (table 6).

The management guidelines established in the TASC II[133] recommend endovascular surgery in infrapoplíteal disease for cases with limb salvage, which is what occurs in patients with infection of a neuroischemic ulcer. Technical success will depend on the length of the lesion to treat and the number of vessels treated, among others.

As regards use of primary stenting in distal trunks, we do not have an adequate level of evidence to justify it, and angioplasty is the first endovascular strategy, with a limb salvage rate at 3 years of 91%, a low cost and a greater lesion length than stent series[184]. Infrapopliteal stenting would be indicated in cases with suboptimal PTA results (residual stenosis >50%) and flow limiting dissections do not improve after prolonged dilatations.

In comparison with conventional surgical techniques, the treatment of choice is the bypass or primary amputation. Femorodistal revascularization surgery with the internal saphenous vein is characterized by its technical complexity and associated morbidity and mortality (up to 18% in some series), with a primary permeability rate at 5 years of 60-70%, secondary of 70-80% and limb salvage of 74-85%[185,186].

Medium-term results obtained in these patients, when endovascular treatment and conventional surgery is feasible, are similar in both groups in terms of amputation free survival time, quality of life and mortality, so many groups have chosen the endovascular procedures. In the short term, however, surgery is associated with increased morbidity and a higher health cost, as a consequence of long hospital stays and resource utilization.

Thus, in patients with a life expectation shorter than 2 and with a significant added comorbidity or with no useful vein, angioplasty should be offered as the first treatment option; and in patients with expectation of life over 2 years and good living conditions, surgery provides improved long-term results with a smaller number of repeat surgeries[187].

Recommedations:

1. Early revascularization is optimum treatment of ischemia in diabetic foot ulcer.

2. The main treatment goals of revascularization in diabetic foot ulcers are to relieve ischemic pain, heal ulcers, prevent loss of the limb, and improve both quality of life and function of the patient.

3. If endovascular and open revascularization may be chosen, endovascular procedures should be used.

PRESSURE OFF-LOADING

The two basic components of diabetic foot ulcers are neuropathy and increased local pressure. Therefore, once infection is eliminated from the ulcer, we must try to minimize pressure in the area to obtain healing and avoid relapses.

Pressure off-loading can be done with foot inserts (full contact foot inserts or custom molded shoes), shoes (shoe changes, foot inserts, orthosis, socks), or by surgery (Achilles tendon lengthening, silicone injections, elimination of skin callosities, bone surgery -metatarsal head resection, osteotomies, arthroplasties, ostectomies, exostectomy, external fixations-)[188-190].

Wheel chairs, walkers, felt plantar unloading pads, cerclages at the knee or ankle joint level may also be used and even bed rest is a method of pressure unloading.

The simplest and most inexpensive way to remove pressure in a given area is elimination of skin callosities though no study has been designed to assess this[188].

Full contact foot inserts have been shown to be the best method to reduce pressure. The NICE guide concludes that there is no significant difference between use of full contact foot inserts, non-fixed foot inserts, and shoes with rear support[191]. What is noteworthy about this type of off-loading versus other non-fixed devices, is the adherence to therapy, which largely depends on the patient having to change the off-loading device (the more it can be changed by the patient, the less it will be used), the capacity to perform their daily activities and the stability perceived in walking when using the device[192]. Therefore, any device that the patient uses, if it is used continuously, will be effective in relieving the pressure.

Reduction of plantar pressure in the forefoot is obtained by shoes with rear support, insoles specifically designed for patients with off-loading in the area of the ulcer or changes to the outer shoe (rocking chair sole). The materials and the design of the insoles are highly variable, so it is not possible to compare results in the reviews made on the subject. Any device used for off-loading must be accompanied by a biomechanical study of the foot. Thus, we must no transfer the load we relieve to another area, where overload may cause a new ulcer[193].

Therefore, a professional should perform a clinical assessment of the patient and adjust the clinical needs on the technical possibilities, evaluating in each case different aspects (neuropathy, joint mobility range, deformities, partial amputations). The type of footwear should be selected with the necessary changes and materials that best suit individual need of each patient. Therefore, it may be the vascular surgeon, nurse, podologist or family doctor who detects the foot at risk, but it should be a rehabilitator, podologist or orthopedic technician who designs and adapts the most appropriate treatment[194].

Recommendation:

Off-loading is necessary to achieve closure of the ulcer. The device applied should be the one that best adapts to the patient and allows to continue performing the required dressings, always involving the patient in strict compliance with use of the off-loading device.

LOCAL THERAPEUTIC APPROACHES

1. Debridement

Debridement is an essential part of local therapeutic approaches (LTA) in an ulcer. In cases of acute ulcers, to remove is remnants of necrotic and infected tissue, and in the case of chronic infections, to stimulate and promote healing.

2. Dressings

In the case of ischemic or infected ulcers, a dry dressing is recommended to reduce the chances of progression of infection and necrosis, although this also delays healing. Once infection and ischemia are treated and the ulcer is clean, a wet dressing is recommended. It is recommended to use dressings to cover the wound and avoid overinfection, though there is no evidence on the use of one over another, so it is recommended to use the dressing with the lower cost based on the clinical indications for its use, the experience of the professional using it and the preferences of the patient.

3. Growth Factors

Only granulocyte colony-stimulating factor (G-CSF) has shown positive effects with no clinical evidence of effectiveness versus platelet-derived growth factor PDGF, epidermal growth factor (EGF) and transforming growth factor beta (TGF-beta). In any case, the NICE review panel recommends that G-CSF should be applied exclusively to wounds that are stabilized and with no signs of moderate or severe infection, so it should not be used in hospitalized patients, but in patients already sent to primary care. Consequently, their hospital use would be limited to patients participating in clinical trials.

4. Hyperbaric Oxygen Therapy

Several retrospective studies and case descriptions recommend the use of hyperbaric oxygen therapy[165,195-197], but it was not until the last decade that prospective studies were conducted[198], showing benefits both individually and in a meta-analysis that concluded that it showed a clear improvement in reducing major amputations. Due to methodological problems in some of the studies, the difficulty in accessing the treatment in most of our centers, and its cost, together with the unproven efficacy in the treatment of necrotizing fascitis[199,200] and the lack of cost-effectiveness studies, consideration of this therapy should be an individual decision in each case until a prospective randomized trial is conducted that can validate this intervention in these patients[165,201], so its routine use is not recommended.

5. Skin Replacements

Table 7 Diabetic foot referral criteria.

1- Normal Referral
- Uninfected Neuropathic Ulcer
- Neuropathic Ulcer with Mild Infection

2.- Preferential Referral
- Neuroischemic ulcer (or suspected) with no pain at rest or pain that subsides with minor analgesics
- Suspicion or evidence of osteomyelitis.
- Unhealed ulcer after two months of appropriate measures (treatments, off-loading, debridement)
- Mild infection that does not improve after 7 days of adequate treatment

3.- Urgent Referral
- Neuroischemic ulcer (or suspected) with pain at rest that failed to subside with minor analgesics
- Suspicion or evidence of mild-moderate, moderate-severe, or severe infection

Both Dermagraft and Graftskin have shown positive effects on ulcer closure (50%) and in decreasing healing time, but without reducing the risk of amputation. However, given the poor quality of the evidence, the lack of evidence regarding prevention of amputations or other surgical interventions, and their high cost, they do not recommend to offer this treatment to hospitalized patients except when they are part of a clinical trial.

6. Negative Pressure Therapy

Although with low quality evidence, studies have shown positive effects in reducing the number of amputations. Long-term economic studies need to be conducted to assess treatment cost-effectiveness.

7. Other Local Therapies

There is no evidence on effectiveness and therefore it is not recommended to offer as adjuvant therapy to hospitalized patients, unless they are enrolled into a clinical trial, the following treatments: electric stimulation, autologous derived-platelet rich plasma, regenerative tissue matrix and dalteparin[191,202].

8. Topical Antibiotic Therapy

Although classically agents such as neomycin, polymyxin, gentamicin, and mupirocin have been used topically, there are no quality studies supporting use of these antibiotics topically in terms of wound healing or reduction in the number of amputations[203,204]. However, in ischemic feet without the possibility of revascularization and in the case of multiresistant bacteria, they may be used since they achieve a higher concentration in the ulcer than systemically[205].

Recommendation:

No clear benefit has been shown in terms of the use of a given dressing. Vacuum therapy and hyperbaric oxygen therapy appear to be associated with a lower number of amputations, but should not be provided routinely.

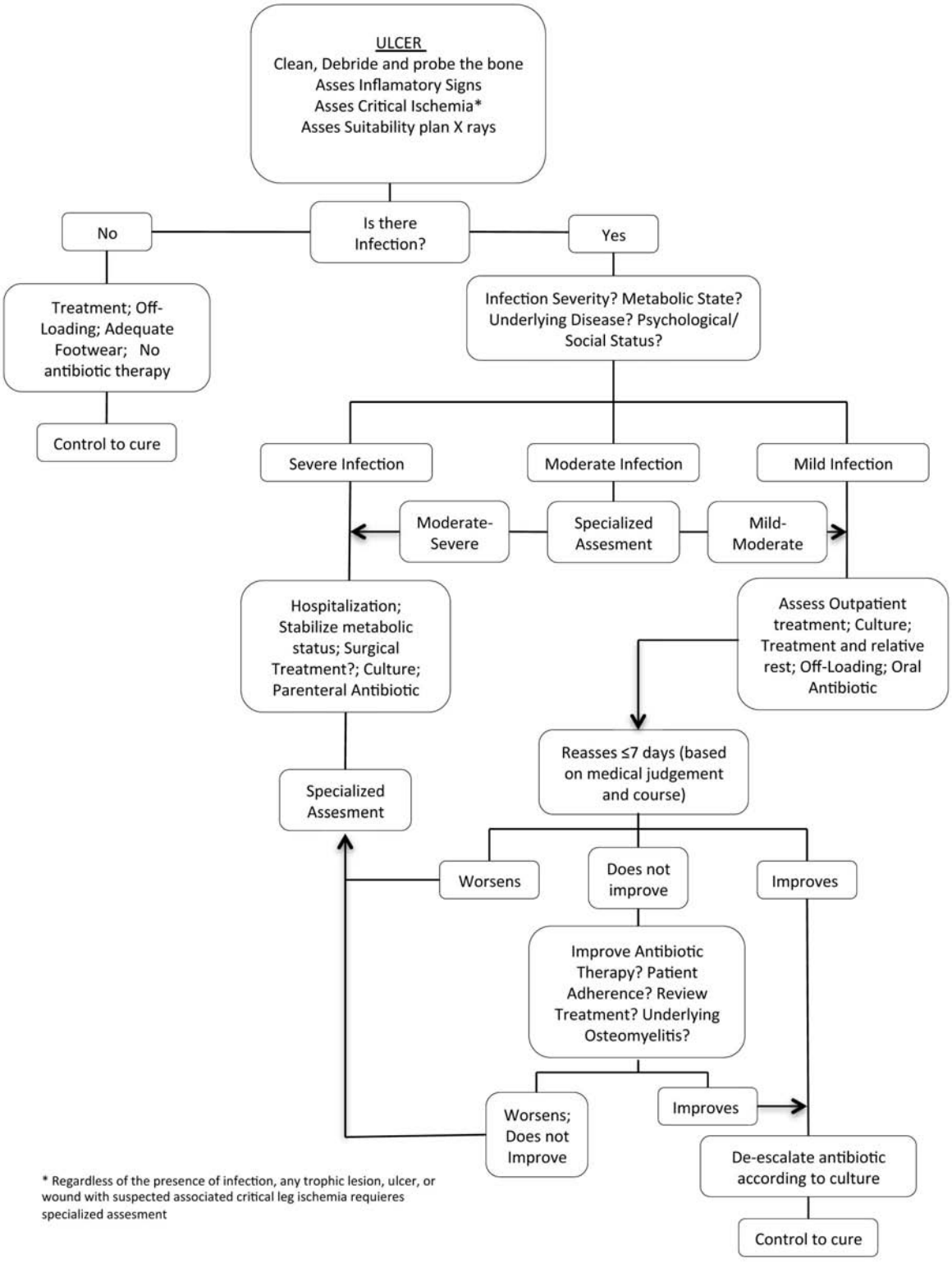

Figure 6 | **Diabetic foot management algorithm.**

NONANTIBIOTIC TREATMENT IN SEVERE INFECTIONS

Notable among nonantibiotic treatments proposed in the literature because of their importance in the clinical course and impact on prognosis, particularly in the most severely affected patients, are the use of colony stimulating factors (G-CSF)[165,206-212], treatment with immunoglobulins and finally packages of measures used to treat severe sepsis and septic shock promoted by the Surviving Sepsis Campaign.

Based on the documented improvement in neutrophil function *in vitro*[213], 5 clinical trials have been made to assess the efficacy of use of G-CSF in these patients[206-211], but no specific benefits were shown in each of them in their different endpoints. However, a meta-analysis performed with the group of studies, although it did not confirm their influence on shortening infection resolution time, did show a lower incidence of amputations and other surgical procedures[165,212].

After the results reported by a European group from a double-blind clinical trial on the use of **immunoglobulins** in streptococcal toxic shock, where a clear statistically significant decrease was shown in mortality based on blockade of superantigens, their use should always be considered in life-threatening cases[214,215].

Finally, we should not forget the importance of compliance with the packages of therapeutic measures based on obtaining a series of objectives in the first six hours in patients who are in a state of severe sepsis and septic shock devised by the Surviving sepsis Campaign[216], which, though they exceed the purpose of this consensus, should be considered in the management of patients with severe DF infections.

MANAGEMENT GUIDELINES

Optimal management of DF infections requires both rapid assessment of the patient and selection of the most suitable doctor in the healthcare system who has sufficient availability and expertise. The design of a management algorithm is particular relevance in the practical management of these patients, including referral specialists (internist, specialist in infectious diseases, endocrinologist, orthopedist, general surgeon, vascular surgeon, microbiologist, rehabilitator), in close cooperation with podology and primary care professionals, both medical and nursing staff (figure 6).

The first step in suspected DF infection is its diagnostic confirmation, established by clinical criteria of the clinical classification and which distinguishes between the different degrees of infection and absence of infection. Next, that is, if clinically documented infection is present, its severity should be established, again using the clinical criteria. Of particular relevance at this point is to identify patients who require immediate referral to a hospital to receive broad-spectrum parenteral antibiotic therapy and/or possible surgical evaluation.

The classification distinguishes between mild, mild-moderate, moderate-severe, and severe infections, while the first two can be managed on an outpatient basis, the latter two require immediate patient care at the hospital. Of all infections, those that raise more questions about their management and the possibility of being treated on an outpatient basis are moderate infections. Moderate infections include a broad range of clinical presentations, from those clearly close to a mild infection to others that may jeopardize the limb of the patient, so moderate infections have been subdivided in mild-moderate and moderate-severe, and should be evaluated by specialists.

Generally, hospitalization should be considered when there are signs systemic toxicity (fever, leukocytosis), metabolic instability (severe hypoglycemia, acidosis), deep or rapidly progressive infection, extensive necrosis, critical ischemia, need for an emergency diagnostic or therapeutic approach, or in cases where the patient cannot self care or lacks adequate social support. The classification criteria, therefore, combine local factors of the foot, systemic impact in the patient and other medical, psychological and social aspects.

REFERRAL CRITERIA

Patients with diabetic foot ulcers in whom there is suspicion or certainty about the presence of situations that could affect the limb that cannot be resolved in primary care should be assessed and, if appropriate, ideally treated in diabetic foot units or if unavailable, in the specialized care setting. In general, in cases of mild-moderate, moderate-severe, or severe infection, or if there is ischemia (table 7).

DIABETIC FOOT UNITS

Diabetes mellitus, with a prevalence 10-15% of the population[217], is the most common cause of lower limb amputation. By identification and education of patients at risk and early detection and adequate treatment of complications, the rate of amputations can be reduced by 80%[218] and ulcer healing increased by 70-85%[219].

For application of a good prevention and treatment plan, understanding of the pathophysiology of DF ulcer is necessary, i.e., the "path to amputation." Ulcer is the precursor in more than 85% of amputations[18]. The longer the ulcer remains open, the more likely it is to be infected, and infection is "coup de grâce" leading to amputation[220].

An effective organization requires systems and guides for education, screening, risk reduction, treatment, and evaluation of results. Local variations in resources and staff will determine the way it is applied. Ideally, it should include:

1.-Education of patients, caregivers, and medical staff at hospitals and primary care centers.

2.-System for detection of patients at risk, with regular examination of patients.

3.-Rapid and effective treatment.

4.-Structure to cover the needs of chronic patients.

DF units should cover the whole diabetic foot process from its diagnosis. It includes screening of every diabetic and particularly those with a high-risk foot, focusing especially on

education and adequate footwear, treatment of trivial foot lesions, such as removal of callosities, nail disorders and cleaning of blisters, and finally the action to be taken in the event of occurrence of an ulcer.

However, screening and management of trivial diabetic foot lesions is outside the scope of these guidelines, therefore we will focus on units to prevent amputation from the occurrence of an ulcer.

There are many models of units, the priority is treatment of infection and revascularisation to obtain ulcer healing, without mentioning ulcer dressing, foot off-loading and metabolic control and cardiovascular prevention in patients.

The skills required by the team according to the DRAFT (Diabetic Accelerated Response Acute Foot Team) guidelines are: - Assessment of ulcer and grading of infection and/or ischemia in it; - Adequate sample collection and microbiological cultures; - Hemodynamic and anatomic vascular assessment and open and endovascular revascularization as appropriate; - Neurological assessment; - Debridement and amputations; - Initiate and modify depending on the lesion, patient, and culture appropriate antibiotic treatment; - Appropriate postoperative monitoring to reduce the risk of reulceration and reinfection[221].

The general recommendations for a unit for treatment of complicated diabetic foot, that is, those with ulcer or skin break in the foot, inflammation or edema in any part of the foot, or any sign of infection, foot fracture or dislocation with no history of significant injury, inexplicable foot pain, or gangrene in part or all of the foot, are[129]:

- Each health area should have a care guide for patients with complicated diabetic foot.

- A multidisciplinary team should treat patients with complicated diabetic foot.

- The multidisciplinary team should include health professionals with specialized skills and competences required to attend patients with complicated diabetic foot.

- The multidisciplinary team may vary depending on the characteristics of each area, but should include a vascular surgeon with diagnostic and therapeutic skills in open and endovascular revascularization and a general surgeon, an internist or a specialist in infectious diseases, an endocrinologist, a podologist and/or nursing staff with knowledge of care of diabetic foot lesions. The team should have access to services and specialists that allow them to carry out the necessary care of these patients.

- The multidisciplinary foot care team should:

· Assess and treat the diabetes of the patient, which should include interventions to minimize cardiovascular risk, and treat renal failure or anemia that may occur.

· Assess, review, and evaluate the patient's initial response to medical, surgical and diabetes treatment.

· Assess the foot and determine the need for special care wound, debridement, off-loading, and other surgical interventions.

· Assess patient pain and determine the need for its treatment and even referral to the pain unit.

· Perform a vascular assessment and revascularization if necessary.

· Review treatment of the infection.

· Determine the need for interventions to prevent development of Achilles tendon contractures and other foot deformities.

· Assessment and orthopedic treatment to facilitate healing and prevent recurrences.

· Access to physiotherapy.

· Plan discharge, which should include to ensure assessment and care of the patient in primary care and follow-up by specialists.

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