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Wagner Evre III ve IV Diyabetik Ayak Ülserlerinin Mikrobiyolojik Değerlendirmesi

Microbiological Evaluation of Wagner Grade III and IV Diabetic Foot Ulcers

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Özet

GİRİŞ ve AMAÇ: Bu çalışmanın amacı patojen mikroorganizma popülasyonları ve antibiyotik dirençleri açısından kritik klinik ve cerrahi müdahale ihtiyacı duyulan Wagner evre III ve IV diyabetik ayak ülserli olguları incelemektir.

YÖNTEM ve GEREÇLER: Gereç ve Yöntemler: Wagner sınıflamasına göre evre 48 evre III ve 42 evre IV olmak üzere toplam 90 diyabetik ayak ülserli olgu çalışmaya dahil edildi. Hasta kayıtları gözden geçirildi ve hasta bilgileri ile mikrobiyoloji kültür sonuçları analiz için kaydedildi.

BULGULAR: Gram (-) mikroorganizmalar 124 örnekte ve Gram (+) mikroorganizmalar 37 örnekte izole edildi. E. Coli, P. aeruginosa, Staphylococcus aureus ve Proteus mirabilis en sık görülen enfeksiyon ajanları olarak tespit edildi. Kültür antibiyogram sonuçlarında Enterococcus türleri % 80, Staphylococcus % 42,8, pseudomonas aeruginosa % 38,4, E.coli ise % 64,5 antibiyotik direnci gösterdi. İzole edilen bakterilerin % 31,6'sı ise birden çok ilaca direnç gösterdi.

TARTIŞMA ve SONUÇ: Cerrahlar, cerrahi müdahale gerektiren Wagner Evre III ve IV ülserli diyabetik ayak olgularında yaygın antibiyotik direncinin farkında olmalıdırlar.

Anahtar Kelimeler: Diyabetik ayak enfeksiyonu, Wagner, Antibiyotik direnci

Abstract

INTRODUCTION: The aim of the study to investigate the cases of Wagner grade III and IV diabetic foot ulcers which need critical clinical and surgical intervention, regarding to the pathogen microorganism populations and their antibiotic resistances.

METHODS: A total 90 cases consist of 48 grade III and 42 grade IV ulcers according to the Wagner classification were included to the study. The carts are reviewed retrospectively, and patients' demographics and microbiological culture results were recorded for analysis.

RESULTS: Results: Gram (-) microorganisms were isolated in 124 samples, Gram (+) microorganisms were isolated in 37 samples. E. Coli, P. aeruginosa, Staphylococcus aureus and Proteus mirabilis were the most frequently detected infectious agents. Enterococcus species showed 80%, staphylococcus showed as much as 42.8%, pseudomonas aeruginosa showed as much as 38.4%, E.coli showed as much as 64.5% drug resistance in the culture antibiogram studies. 31.6% of the isolated bacteria showed a multidrug resistance.

DISCUSSION AND CONCLUSION: Surgeons should be aware of common antibiotic resistance in diabetic foot cases had Wangner grade III and IV ulcer which require surgical intervention.

Keywords: Diabetic foot infection, Wagner, Antibiotic resistant

GİRİŞ

Foot infections are one of the most common causes of mortality and morbidity in patients with diabetes mellitus (DM). Diabetic foot infections constitute a major burden not only for the patient but also for governments due to the overall health cost. It was reported that in the USA almost 20% of hospital admissions of diabetic patients are due to diabetic foot ulcers (1). It was also reported that diabetic foot ulcers are the main cause of lower extremity amputations in the USA, responsible for 50%-70% of amputations (1,2). Almost 25% of diabetic patients suffer from diabetic foot ulcers throughout their life and nearly 15%-20% eventually undergo limb amputation (3).

As a result of the increase in the total number of diabetic patients, the total number of diabetic foot ulcer patients and diabetic foot ulcer related complications have significantly increased. In a study conducted in Turkey in patients over the

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age of 20, diabetes mellitus was shown to have a prevalence of 7.2% (4). When we consider the rate of diabetic foot ulcers among diabetic patients (5%-7%), it is estimated that at least 200,000-300,000 people suffer from diabetic foot and related complications (5).

Despite the diabetes training programs, wound care, protective measures against the formation of diabetic ulcers, proper treatment of the diabetic foot ulcers should involve the classification of the wound and isolation of the responsible microorganism (6,7). There are several classifications regarding the diabetic foot ulcers in the literature (8). Wagner classification, which includes every stage of the diabetic ulcers from a vulnerable foot with intact skin to the gangrenous foot (9) (Table 1)(10) . The Wagner grade III and IV ulcers requires more critical multidisciplinary approach in order to prevent inevitable possibility of amputation than the lower grades. So cases with Wagner grade III and IV diabetic ulcers are chosen as the target for investigating the both microbial population and drug resistance.

Table 1. Wagner Classification

Intact skin with a bony prominence and/or callus formation (risk factor for ulcer formation)	Stage 0
Superficial ulcer without involvement of the subcutaneous tissues	Stage 1
Deep ulcer with the involvement of tendon, bone, ligaments or joint	Stage 2
Deep ulcer associated with abscess and/or osteomyelitis	Stage 3
Gangrene of the fingers and/or metatarsal bones	Stage 4
Gangrene of the heel and/or the whole foot which is unsalvageable and prompts amputation	Stage 5

Diabetic foot infections may range from uncomplicated cellulitis to purulent ulceration and gangrenous necrosis. The decrease in resistance to infections, phagocytic activity, deficiency in cellular and humoral immunity, impairments in macro- and micro-circulation leading to peripheral vascular insufficiency play an important role in the formation of diabetic foot ulcers (11). Pathogens are generally originated from the patient's own bacterial or fungal flora. One of the most typical properties of diabetic foot infections is its polymicrobial nature. This finding is particularly more prominent in hospitalized patients with bone involvement and/or tissue necrosis. Gram (+) cocci, Gram (-) bacilli, and anaerobes can be isolated from these lesions. The most commonly isolated microorganisms from diabetic foot infections are (12- 14):

- **Gram (+) cocci:** Staphylococcus aureus, group B streptococci, enterococci, Staphylococcus epidermidis.
- **Gram (-) bacilli:** Escherichia coli, Proteus vulgaris, Proteus mirabilis, Pseudomonas aeruginosa, Klebsiella pneumoniae, Enterobacter and Citrobacter species.
- **Anaerobic bacteria:** Bacteroides fragilis, other Bacteroid species, Peptococci, Clostiridium species, Prevotella melaninogenicia.

The bacteria that are reproduced from the swab specimens taken from the surface of the wound or the regions adjacent to the wound reflect only the surface colonization so they may be insufficient for the determination of the

infectious agent. For this reason, the ideal microbiological method is to obtain deep tissue cultures (DTC). However, if it is not possible to acquire DTC, specimens can be taken by curettage from the base of the wound after a careful clean-up process. These specimens give more reliable results than swab specimens (12,13). In diabetic foot infections, antibiotic treatments are recommended. In severe diabetic foot infections, antibiotics that are used include wide-spectrum penicillins, cephalosporins, carbapenems, quinolones, and aminoglycosides. However, for foot ulcers, there exist no treatment regimen on which a consensus has been reached because of the clinical studies about this issue include also the non-diabetic patients besides diabetic patients and the number of the patients in these studies are not high enough to indicate any significant differences between different treatment regimens.

In this study, the microorganisms reproducing in the wound site, and the antimicrobials to which they are resistant were determined retrospectively in patients with diabetic foot infections, so avoidance of such antimicrobials is intended. The aim the present study was to present the pathogen microorganism populations in Wagner grade 3 and 4 diabetic foots and their antibiotic resistances.

MATERIAL AND METHODS

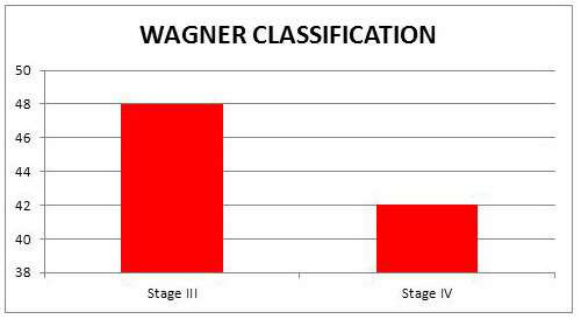
Ninety patients with type 2 DM who were followed with the diagnosis of diabetic foot infection were retrospectively scanned for the results of their wound culture and antibiograms. Patients' data regarding age, sex, type and duration of diabetes, presence of diabetic complications, and physical examinations were accessed from patient records. Localization of diabetic ulcers were documented and classified according to the Wagner classification. Leukocyte counts, erythrocyte sedimentations rates (ESR), C-reactive protein (CRP) levels, X-ray films of the involved extremity for osteomyelitis findings were collected and documented. The microbiological samples were analysed both conventionally and by automatized systems. Antibiograms were evaluated according to the National Committee for Clinical and Laboratory Standards (NCCLS).

RESULT

Among the total 90 diabetic patients with foot ulcers, 67 were male (74.4%) and 23 were female (25.6%). Mean age was 52 (37-78) years. All patients were diagnosed with type 2 DM and in follow up by endocrinologists. Eight patients had a diabetic foot infection with acute onset, 82 patients were presented with subacute or chronic ulcers. Sixty three patients were being treated with insulin and 23 patients were being treated with oral antidiabetics. Four patients were not on any antidiabetic drugs. Twenty three patients had diabetic nephropathy and 56 of the patients had diabetic retinopathy. Sixteen

patients had triphasic lower extremity arterial blood flow, 48 had biphasic and 36 had monophasic blood flow. Radiological findings of 42 patients with infected diabetic foot ulcers suggested osteomyelitis. According to the Wagner classification, 48 had stage III and 42 patients had stage IV ulcers (Figure 1).

Figure 1:



When the localization of ulcers was evaluated, 63 patients had ulcers at the level of and distal to the metatarsophalangeal joint, whereas 27 patients had lesions proximal to this level. All patients underwent debridement, and superficial and deep wound cultures were taken with tissue biopsies and culture swabs. Seventeen patients underwent a distal amputation in the first debridement and 25 more patients eventually underwent distal amputation after several debridements. In 48 patients, wound healing was achieved by serial debridements together with appropriate antibiotic use according to antibiograms and wound reconstruction. When the results of the cultures were analysed, Gram (-) microorganism were isolated in 124 samples, and Gram(+) microorganisms were isolated in 37 samples (Figure 2). Isolated microorganisms were then seeded on antibiogram disks, which had 17 different antibiotics. Isolated microorganisms, in descending order, were 19.2% E.coli (31/161), 16.1% P.aeruginosa (26/161), 8.7% Staphylococcus aureus (14/161), 8.1% Proteus mirabilis (12/161), 7.4% Serratia mercecens (12/161), 6.2% Enterococcus species (10/161), 4.9% Proteus vulgaris (8/161) (Figure 3). Thirty two percent (29/90) of the infections were polymicrobial.

Figure 2:

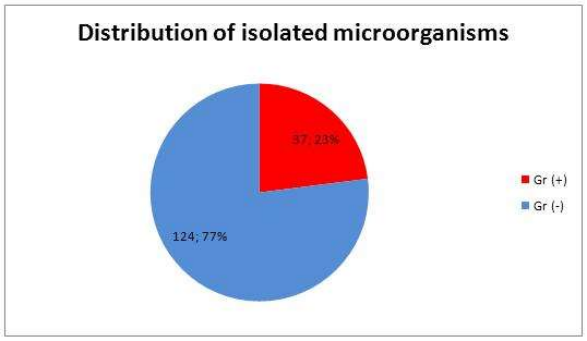
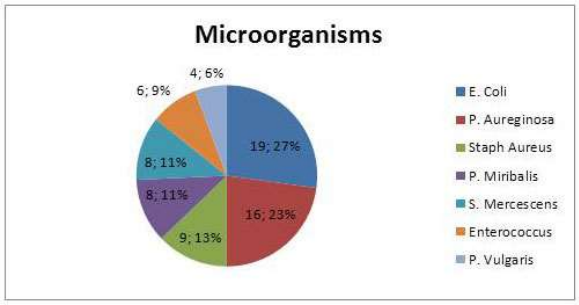


Figure 3:



Levofloxacin and ciprofloxacin resistances were observed in 8 of 10 (80%) patients who had Enterococcus species in their culture. Among 14 patients who had Staphylococcus 4 (28.5%) had oxacillin and 6 (42.8%) had ampicillin resistance. Pseudomonas aeruginosa was isolated in 26 patients, 10 (38.4%) were resistant to piperacillin-tazobactam and 10 were resistant to amikacin. E.coli was isolated in 31 patients, 20 (64.5%) of which were resistant to piperacillin-tazobactam and 17 (54.8%) were resistant to

ciprofloxacin. Multi-drug resistance was detected in 31.6% of the isolated bacteria.

Because of technical restrictions anaerobic cultures were not used in the study. Isolated Gram (+) microorganisms are shown in Table 2 and Gram (-) microorganisms are shown in Table 3.

Table 2. Distribution of antibiotic resistant strains of Gram positive bacteria isolated

ANTIBIOTIC	Enterococcus Spp.	Streptococcus Spp.	S. Aureus
Levofloxacin	8	2	4
Cefoxitin	0	0	3
ciprofloxacin	8	-	1
Oxacillin	2	1	4
Penicillin	6	0	6
Ampicillin	7	0	6
Vancomycin	0	0	0
TMP/SMX	4	1	3
Rifampicin	-	1	2
Erythromycin	-	1	2
Tetracycline	7	1	2

Table 3. Distribution of antibiotic resistant strains of Gram negative bacteria isolated

ANTIBIOTIC	S. Marcescens	P. Vulgaris	P. Mirabilis	P. Aeruginosa	E. Coli
Ampicillin	6	5	8	-	17
PTZ	1	2	4	10	20
Cefuroxime	6	4	3	1	11
Ceftriaxone	-	3	2	-	11
Ceftazidime	-	-	2	6	11
Cefepime	-	-	2	6	11
Meropenem		-	2	3	0
Ciprofloxacin	-	-	2	9	17
Amikacin	-	1	-	10	0
Gentamicin	-	-	-	11	14
TMP-SMX	-	3	8	1	17

(TMP – SMX : Trimethoprim / Sulfamethoxazole) (PTZ: Piperacillin-Tazobactam)

DISCUSSION

Diabetic patients are prone to develop chronic wound infections because of the immune deficiency, macro- and micro-vascular angiopathy, and diabetic neuropathy. Diabetic foot ulcers are one of the most unwanted complications of diabetes, which causes chronic infections, causing excess antibiotic usage, prolonged hospitalization, and increased overall cost of treatment. Microbiological analysis of diabetic foot infections is essential since a timely treatment directed against the specific microorganism is particularly important in diabetic patients. Difficulties in specimen transport, presence of anaerobic microorganisms, and polymicrobial nature of the infections can cause problems and difficulties in isolation of the responsible pathogen (14-18). In most cases it is hard to determine the presence and extent of infection. It is particularly important to diagnose deep abscesses that may end up with the loss of the extremity or even the patient's death. The local and systemic signs of infection such as erythema, pain, warmth and tenderness may not be evident in some diabetic patients with abscesses and osteomyelitis. Clinical, hematologic and bacteriologic tests may yield false negative results in patients with diabetic foot infections. Systemic findings, such as fever, are not evident in almost ⅔ of patients even though the extent of infection of the involved extremity is serious enough to end up with the loss of the extremity (13). Also some hematologic findings, like leucocytosis, may not be present in some patients. Defining the extent of tissue injury and isolating the causative microorganism are the main steps in the treatment of diabetic foot ulcers. For this purpose, several classifications are suggested. Wagner classification is generally preferred because of the ease of its implementation (12,13). There are several methods for the isolation of the causative microorganism. These are swab culture, curettage, DTC/bone culture,

and needle puncture. Swab and curettage cultures have a high risk of contamination since there is a contact with the skin flora, while DTC/bone cultures and needle punctures are considered as gold standards with a minimal contamination risk.

The non-limb-threatening infections in the form of mild cellulitis are also usually polymicrobial. However, in these mild cases, monomicrobial infections are usually caused by Staphylocci species. In these infections Gram (-) bacilli and anaerobes are not isolated as a monomicrobial etiologic factor. Pseudomonas aeruginosa is more commonly isolated in hospitalized patients. Lipsky et al. reported that among non-hospitalized patients, diabetic foot infections are 46% monomicrobial and 47% polymicrobial. Anaerobes are isolated in about 13% of the infections. In our study, diabetic foot infections are 68% monomicrobial and 32% polymicrobial. The results of this study is summarized in Table 4 (19).

Table 4. Commonly isolated microorganisms in serious diabetic foot infections (%)

	Grayson 1994***	Bamberger 1987**	Wheat 1986*
Monomicrobial	16	16	29
Polymicrobial	80	84	69
No growth	4	-	2
Aerobe only	58	19	59
Aerobe+Anaerobe	40	78	41
Only anaerobe	2	-	-

*(19) ; **(20) ; ***(12)

According to the literature anaerobic bacteria take part in most of the diabetic foot infections especially in deeply located ones (20). Gram (-) anaerobic bacilli are found in lesser amounts. Clostridium species are the least encountered ones (21,22). Gram (+) anaerobes being sensitive to most of the antimicrobials including penicillin are not determinants of the treatment decision. Including the species of bacteroides which can have multi-resistant properties, Gram (+) anaerobic bacilli are detected less frequently, but due to their antimicrobial resistance, they deserve consideration (20,23). In cases of diabetic foot infections, previous use of antibiotics in patient's history can give an idea about the infectious agent(s). In these patients, the bacteria that are not included in the spectrum of the currently given antibiotic give rise to the infection. Also the pattern of resistance may differ according to the given antibiotic. Similarly, when compared with community acquired infections, more resistant bacteria strains are detected in nosocomial infections (13,20,24).

In different studies among Wagner Stage ≤2 patients the swab cultures had predicted with 82%-90% accuracy the microorganisms grown in the deep tissue biopsies. This percentage was found to be 30%-78% among stage ≥3 patients (25,26). There have been an increase in the true prediction rates of swab cultures in the recent years due to improvements in swab techniques and, as a result, a decrease in the contamination risks has been observed (27).

Severe diabetic foot infections are generally of polymicrobial etiology whereas mild to moderate infections have a single pathogen. Even though the causative microorganisms are usually Gram

(+), severe and life threatening infections are commonly caused by Gram (-) and anaerobic bacteria (28,29).

Patients without a prior antibiotic treatment are generally present with Gram(+) cocci monobacterial infection. Severe and extremity threatening infections are almost always mixed type of infections and responsible microorganisms are, in addition to Gram+ bacteria, E.coli, Proteus spp., Klebsiella spp., Morganella morganii, Enterobacteriacea spp., Pseudomonas aeruginosa, Bacteroides spp., Clostridium spp. (14,16,30,31).

In different studies, different pathogens have been observed to be the most common cause. For example, Sert et al. reported the most common isolated microorganisms as S. aureus, KNS, Enterococcus spp., P.aeruginosa, and E.coli, in descending order (32). In our study the most commonly isolated microorganisms are E.coli, P.aeruginosa, and S.aureus. These microorganisms were usually found in the stool. In this study, because of the low socio-economic status of our patients who mostly live in unhygienic environment, these factors were more frequently isolated.

Motta et al. reported in their study involving 138 patients with diabetic foot infections that Enterobacteriaceae spp. were commonly isolated with K.pneumonia (21.2%), Morganella morganii (19.9%), and E.coli (15.4%). In the same study, it was reported that 6% of the isolated microorganisms produce extended-spectrum beta lactamases, which suggests that antibiotic resistance in Gram (-) microorganisms become more prevalent even in community acquired infections (33).

In 2006, Tentolouris et al. reported in their study involving 84 infected and non-infected diabetic foot ulcer patients that the most commonly isolated microorganism was S.aureus, 50% of which was methicillin-resistant S.aureus (34). In our study, the most commonly isolated microorganisms were E.coli, P.aeruginosa, and S.aureus.

Arterial perfusion of the involved extremity is the primary determinant of the effective antibiotic concentration in the tissue in addition to pharmacodynamic properties of the antibiotic. In the literature, studies are not considered to be reliable enough to compare the effects of different antibiotic regimens, mainly because of the heterogeneous properties of the diabetic patients with foot ulcers and the insufficient standardization of the compared groups. These studies also confer the need for more detailed classification systems for diabetic foot ulcer infections. In the recent treatment guidelines, ampicillin-sulbactam, clindamycin-ciprofloxacin, piperacillin-tazobactam, and imipenem-cilastatin are the most commonly suggested antibiotic regimens in patients with moderate to severe diabetic foot infections (2,15).

In a study evaluating the efficacy of piperacillin-tazobactam in 23 patients with Wagner stage 1 to 4 diabetic foot infections, 97% recovery is

reported (35). In another study, in which piperacillin-tazobactam is compared with another antibiotic -ertapenem- the recovery rate from infection was reported to be 71% (14). Grayson et al. reported that, when imipenem-cilastatin is compared with ampicillin-sulbactam, clinical cure rate was 81% and microbiological cure rate was 75% for imipenem-cilastatin (36).

A multidisciplinary approach is needed in the treatment of diabetic foot infections. Antibiotic treatment is a major component of the treatment protocol of these infections. Surgical treatment combined with empirical antibiotic treatment with broad antibiotics directed against the most possible microorganisms was shown to decrease morbidity and mortality (37). In mild superficial infections, which are usually caused by Gram (+) cocci, more specific-narrow spectrum antibiotics should be preferred, whereas in more severe infections, broad-spectrum antibiotics, which are effective against Gram (+), Gram(-), and anaerobic microorganisms, should be preferred (38,39).

There is no consensus about the duration of antimicrobial treatment in patients with diabetic foot ulcers. The duration of treatment should be determined according to the stage of the infected wound. Recently, it has been accepted that a 14-day treatment is considered to be sufficient for low risk infections. For severe and high risk infections, the duration of treatment varies according to the general status of the patient and the severity of the infection. In patients with infected ulcers extending deep into the underlying bones and causing osteomyelitis, if an amputation is performed, a two-week antibiotic therapy may suffice, whereas if amputation is not performed, an antibiotic treatment of at least 6 weeks will be needed (40,41).

Technical restrictions that preclude the use of anaerobic cultures, heterogeneity of patient groups, lack of correlation between severity of infection and infection markers were the major limitations of this study.

Diabetic foot infections are a common heath problem since they decrease life quality, increase treatment and hospitalization costs, and increase morbidity and mortality of the diabetic patients. Uncontrolled antibiotic use is known to increase bacterial resistance, hence complicate the infections and make it difficult to treat these infections with frontline antibiotics (10). For these reasons, the prevention of diabetic foot infections is important for the community as well as for the economy. As a result, the classification of diabetic foot wounds, isolation of the pathogenic bacteria is the main steps to determine the appropriate treatment protocol, which may change from patient to patient. Since common pathogens are different in our study from the literature, surgeons should be aware of common antibiotic resistance in diabetic foot patients.

REFERENCES

1. Sapico FL, Bessman AN. Foot infections in the diabetic patient. In: Gorbach SL, Bartlett JG, Blacklow NR, Eds. Infectious Diseases. Second ed. Philadelphia: WB Saunders, 1998:1270 – 2.
2. Lipsky BA. Osteomyelitis of the foot in diabetic patients. Clin Infect Dis, 1997; 25:1318 – 26.
3. Albrant DH. Management of foot ulcers in patients with Diabetes. J Am Pharm Assoc, 2000; 40: 467 – 74.
4. Satman İ, Şengül AM, Uygur S, Salman F, Baştar İ, Sargın M, Tütüncü Y, Karşıdağ K, Dinççağ N, Özcan C et al. The TURdep Group, Diyabetes Div. İstanbul Univ. State Inst. Statistics and Min. Health – Turkey 36th EASD Jarusalem,17-21 September 2000. Provisional Programme p: 49. Diabetologia, 2000; Suppl 1.
5. Ertuğrul MB, Baktıroğlu S, Aksoy M, Çalangu S. Diyabetik Ayak ve Enfeksiyonu. Klimik Dergisi, 2004; 17 : 3 -12.
6. Levin M.E. Foot Lesions in Patient with Diabetes Mellitus. Endocrinol Metab Clin North Am 1996; 25: 447-462.
7. Boulton AJM: The importance of abnormal foot pressure and gait in the causation of foot ulcers. In Connor H, Boulton AJM, wards JD (edt). The Foot in Diabetes. John Wiley & Sons. 1987; 11-21.
8. Wagner W F. The Dysvascular Foot: A System for diagnosis and treatment. Foot Ankle 1981; 2: 62-122.
9. Ulusoy S. Diyabetik ayak enfeksiyonları. Modern Tıp Seminerleri: 33: 2006; 40-45.
10. Bozkurt F, Tekin R, Çelen M.K, Ayaz C. Wagner classification and culture analysis of diabetic foot infection. Dicle Tıp Derg / Dicle Med J Cilt / Vol 38, No 1, 31-34.
11. Gough A, Clapperton M, Rolando N, Foster AV, Philpott-Howard J, Edmonds ME: Randomised placebo-controlled trial of granulocyte-colony stimulating factor in diabetic foot infection. Lancet 1997; 350: 855-859.
12. Ulusoy S, Arda B, Bayraktar F, Sesli E, Özinel MA, Yamazhan T, Ünal İ, Kısakol G, Tüzün M. Diyabetik ayak infeksiyonları: 179 olgunun değerlendirilmesi. Flora 2000; 5: 220-228.
13. Grayson ML: Diabetic foot infections-antimicrobial therapy. Infect Dis Clin North Am l995;9: 143-161. 81
14. Armstrong DG, Lipsky BA. Advances in the treatment of diabetic foot infections. Diyabetes Technol Ther 2004; 6(2):167-177
15. Lipsky BA, Berendt AR, Deery HG, Embil JM, Joseph WS, Karchmer AW, LeFrock JL, Lew DP, Mader JT, Norden C et al. Diagnosis and treatment of diabetic foot infections. Guidelines for Diabetic Foot Infections. CID, 2004; 39 (1 October): 885 – 910.
16. Ansari MA, Shukla VK. Foot Infections. Int J Low Extremy Wounds, 2005; 4(2): 74 – 87.
17. Brodsky J, Schneidler C. Diabetic foot infections. Orthop Clin North Am 1991; 22(3):473-489.
18. Shea KW. Antimicrobial therapy for diabetic foot infections. A practical approach. Postgrad Med 1999; 106(1):1-10.
19. Lipsky BA. Pecorato RE, Wheat LJ. The diabetic foot. Soft tissue and bone infection, Infect Dis Clin North Am 1990; 4: 409-432
20. Wheat LJ, Ailen SD, Henry M, Kerek CB, Siders Ja, Kuebler T, Fineberg N,Norton J. Diabetic foot infections - Bacteriologic analysis. Arch Intern Med 1986;146: 1935-38.
21. Bamberger DM, Daus GP, Gerding DN. Osteomyelitis in the feet of diabetic patients. Am J Med 1987; 83:653-55.
22. Hollinworth H. Managing a patient with an infected foot ulcer. J wound Care 1993; 2: 22-26.
23. Joseph WS; Treatment of lower extremity infections in diabetics. Drugs 1991; 42: 984-86
24. West NJ. Systemic antimicrobial treatment of foot infections in diabetic patients. Am J Health Syst Pharm 1995;52: 1199-1202.
25. Slater R, Lazarovich T, Boldur I, Ramot Y, Buchs A, Weiss M, Hindi A, Rapoport MJ. Swab culteres accurately identify bacterial pathogens in diabetic foot wounds not involving bone. Diabet Med 2004; 21: 705-709
26. Pellizzer G, Strazzabosco M, Presi S, Furlan F, Lora L, Benedetti P, Bonato M, Erle G, de Lalla F. Deep tissue biopsy vs. superficial swab culture monitoring in the microbiological assessment of limb-threatening diabetic foot infection. Diabet Med 2001; 18:822-827.
27. Ertuğrul MB. Diyabetik Ayak Enfeksiyonlarında Kemik Doku ve Yumuşak Dokudan İzole Edilen İnfeksiyon Etkeni Mikroorganizmaların Karşılaştırılması Uzmanlık Tezi. İstanbul. İstanbul Üniversitesi İstanbul Tıp Fakültesi, 2003.
28. Sapico FL, Bessman AN. Quantitative aerobic and anaerobic bacteriology of infected diabetic feet. J Clin Microbiol 1980; 4:413-420.

29. Pathare NA, Bal A, Talvalkar GV, Antani DU. Diabetic foot infections: a study of microorganisms associated with the different wagner grades. Indian J Pathol Microbiol 1998; 41: 437-441.
30. Frykberg RG, Armstrong DG, Giurini J, Edwards A, Krawette M, Kravitz S, Ross C, Stavosky J, Stuck R, Vanore J. Diabetic foot disorders. A Clinical Practice Guideline. J Foot Ankle Surg 2000; 39: Supplement.
31. Ge Y, MacDonald D, Hait H, Lipsky BA. Microbiological profile of infected diabetic foot ulcers. Diabet Med, 2002; 19:1032 – 1035.
32. Sert M, Tetiker T, Koçak M, Aksu HSZ. Diyabetik ayak enfeksiyonlarında ampirik antibiyotik kullanılması. Endokrinolojide Yönelişler, 2000; 9 : 47- 49.
33. Motta RN, Oliveira MM, Megahaes PS, Dias AM, Araqao LP, Forti AC, Carvalho CB. Plasmid mediated extended spectrum beta lactamase producing strains of Enterobacteriacea isolated from diabetic foot infections in a Brasilian diabetic center. Braz J Infect Dis, 2003; 7: 129 – 34.
34. Tentolouris N, Petrikkos G, Vallianou N, Zachos G, Daikos GL, Tsapogas P, Markou G, Katsilambros N. Prevalance of methicillin-resistant *Staphylococcus aureus* in infected and uninfected diabetic foot ulcers. Clin Microbiol Infect, 2006; 12: 178 – 196.
35. Zeillemaker AM, Veldkamp KE. Piperacillin – Tazobactam therapy for diabetic foot infection. Foot Ankle Int, 1998; 19(3):169-172.
36. Grayson ML, Gibbons GW, Habershaw GM, Freeman DV, Pomposelli FB, Rosenblum BI, Levin E, Karchmer AW. Use of ampicillin/sulbactam versus imipenem/cilastatin in the treatment of limb-threatining foot infections in diabetic patients. Clin Infect Dis, 1994; 18: 683 – 693.
37. Bridges RM, Deitch EA. Diabetic foot infections. Surg Clin North Am. 1994; 74 : 537-555.
38. Abdulrazak A, Bitar ZI, Al-Shamali AA, Mobasher LA Bacteriological study of diabetic foot infections. J Diabetes Complications 2005; 19:138-141.
39. Özkan, Y., R. Çolak, K. Demirdağ, M. A. Yıldırım, G. Özalp, S. S. Koca. Diyabetik ayak sendromlu 142 olgunun retrospektif değerlendirilmesi. Türkiye Klinikleri J Endocrin 2004; 2:191-195.
40. Boutoille D, Leautez S, Maulaz D, Krempf M, Raffi F. Skin and osteoarticular bacterial infections of the diabetic foot. Treatment. Presse Med 2000; 29: 396-400.
41. Örmen B, Türker N, Vardar İ. Diyabetik ayak infeksiyonlarının klinik ve bakteriyolojik değerlendirilmesi. İnfeksiyon Derg 2007; 21: 65-69