JIDC | THE JOURNAL OF INFECTION IN DEVELOPING COUNTRIES

Original Article

Risk factors affecting the development of adverse outcomes in patients with diabetic foot infection

Rukiye İnan Sarikaya[1], Ömer Karaşahin[1]

[1] Department of Infectious Diseases and Clinical Microbiology, Health Sciences University, Erzurum Regional Training and Research Hospital, Erzurum, Turkey

Abstract

Introduction: Diabetic foot infection (DFI) can result in lower extremity amputation and death in patients with diabetes and is an important cause of morbidity and mortality. The purpose of this study was to identify predictive factors for adverse outcomes consisting of major lower extremity amputation and mortality in patients with DFI.

Methodology: One hundred and two patients diagnosed with DFI and followed up in a tertiary hospital between November 2022 and April 2023 were included in this prospective study. Demographic and diabetic foot characteristics at the time of presentation, degrees of DFI, and clinical and laboratory findings of all patients were recorded. Major amputation and/or mortality were regarded as adverse outcomes. The patients were followed up throughout hospitalization until discharge or mortality. Risk factors for adverse outcomes were identified using univariate and multivariate logistic regression analyses.

Results: The median age of the patients was 60.0 years, and the majority (72.5%) were men. Adverse outcomes developed in 11 patients during follow-up. The factors linked to adverse outcomes included fever; wound necrosis; isolation of Enterobacteriaceae species in wound culture; perfusion, extent, depth, infection, and sensation (PEDIS) grade 4; and blood urea nitrogen (BUN) elevation. Following multivariate logistic regression analysis, only BUN > 31 mg/dL and presence of necrosis emerged as significant independent predictive factors for adverse outcomes.

Conclusions: The findings show that the above factors may be useful in predicting adverse outcomes in patients with DFI. Early detection of these factors may be useful in preventing morbidity and mortality in these patients.

Key words: adverse outcome; amputation; diabetic foot; infection; DFI; mortality.

J Infect Dev Ctries 2025; 19(4):520-527. doi:10.3855/jidc.19974

(Received 08 February 2024 – Accepted 19 September 2024)

Introduction

Diabetes mellitus (DM) is a chronic public health issue capable of progressing with various micro- and macrovascular complications. Approximately 537 million adults worldwide are affected by DM, and this figure is expected to reach 783 million by 2045. The number of patients affected by the chronic complications of diabetes such as diabetic foot ulcer (DFU) is therefore expected to rise in line with the prevalence of DM and longevity [1]. Many DFUs become complicated by infection. Diabetic foot infection (DFI) resulting from the addition of infection to a background of ischemia, ulcer, and neuropathy, leads to increased lengths of hospital stay, increased health service costs, lower extremity amputation, and even mortality [2].

It is estimated that among diabetic patients, at least one extremity is lost due to DFU every 30 seconds. In addition, DFU represents approximately half of all non-traumatic amputations [3]. These amputations are important surgical procedures performed for the purpose of preventing severe complications, such as widespread infection or sepsis, in patients with DFU. Amputation above the ankle, known as major lower extremity amputation, is a feared consequence of DFU. Major amputations associated with DFU result in severe physical impairment [4]. These amputations are a life-saving procedure, and the last resort in the treatment of DFI. The amputations can cause severe economic, social, and psychological effects due to immobilization of the patients [5]. In addition, major amputations in patients with DFU increase the risk of mortality compared to minor amputations, have been linked to decrease in survival rates, and have been identified as independent risk factors for mortality [6,7]. This may be associated with the increased cardiovascular risk in the patient subgroup that already has high prevalence of cardiovascular disease [8]. The most important risk factors for mortality in patients with DFU are age, wound ischemia, impaired renal function, and male gender [9]. Previous studies have focused on diabetic foot, independently of infection. However,

infection is the most widespread cause of amputation [10]. In contrast to DFU, long-term survival rates in DFI are unknown. Moreover, very few studies have focused on patients with DFI. A knowledge of potential risk factors capable of predicting adverse outcomes consisting of major amputation and mortality in patients with DFI will therefore be useful in their prevention.

The purpose of this study was to investigate risk factors associated with major amputation and mortality, regarded as adverse outcomes, in patients with DFI requiring hospitalization for treatment.

Methodology

Study design and population

This single-center study investigated risk factors associated with major amputation and mortality, regarded as adverse outcomes, in patients with DFI. Approval for the study was granted by the Erzurum Regional Training and Research Hospital ethical committee (decision no: Erzurum BEAH KAEK 2022/18-169 dated 24 November 2022). This prospective study was performed over a 6-month period between November 2022 and April 2023 at the Erzurum Regional Training and Research Hospital, Infectious Diseases and Clinical Microbiology Department, Turkey. The research was conducted in conformity with the Declaration of Helsinki.

One hundred and two patients, aged 34–87 years, and diagnosed with DFI were included in the study. Written informed consent was obtained from all participants. Major lower extremity amputation and/or death were regarded as adverse events.

Inclusion and exclusion criteria

Only patients diagnosed with DFI and aged over 18 years were included.

Pregnant women; individuals aged under 18 years; and patients with type 1 DM, chronic liver disease, chronic kidney failure and receiving hemodialysis, cancer patients under treatment, patients with active infection other than DFI, and those with acute or chronic inflammatory disease were excluded.

Data collection

A detailed medical history was obtained from all participants. The patients' demographic characteristics (age, gender, place of residence, education level, and occupation) at the time of admission to the clinic, duration of DM, antidiabetic drugs used, comorbidities, vital findings, smoking status, diabetic foot characteristics, and clinical and laboratory findings were recorded. Anthropometric data such as height, body weight, and body mass index (BMI) were obtained using standard methods. Detailed descriptions regarding DFI were recorded. Infection was defined as at least two of local swelling, erythema, pain, local increased temperature, and purulent discharge. The wounds of the patients with DFI were recorded based on the perfusion, extent, depth, infection, and sensation (PEDIS) classification published by the International Diabetic Foot Working Group. Absence of signs and findings of infection was classified as PEDIS grade 1. Patients with any two of such signs as local swelling or induration, erythema of 0.5–2 cm around the ulcer, local tenderness or pain, local increased temperature, or purulent discharge were classified as PEDIS grade 2. Patients with erythema exceeding 2 cm and any one of the grade 2 infection findings; or infection involving structures deeper than the skin such as abscess, osteomyelitis, septic arthritis, or fasciitis without any signs of systemic inflammatory response syndrome (SIRS) were classified as PEDIS grade 3. Any foot infection with signs of SIRS was classified as PEDIS grade 4 [11]. Peripheral sensory neuropathy was diagnosed using electroneuromyography, and peripheral artery disease (PAD) or venous insufficiency using Doppler ultrasonography. Osteomyelitis was diagnosed either clinically or by using methods such as magnetic resonance imaging or scintigraphy. Laboratory parameters including hemoglobin (g/dL); leukocyte, neutrophil, lymphocyte, and platelet counts (10^9/L); alanine aminotransferase (ALT) (U/L); aspartate aminotransferase (AST) (U/L); creatinine (mg/dL); blood urea nitrogen (BUN) (mg/dL); glucose (mg/dL); albumin (g/dL); C-reactive protein (CRP) (mg/L); erythrocyte sedimentation rate (ESR) (mm/h); HbA1c (%); total cholesterol (mg/dL); and low density lipoprotein cholesterol (LDLc) (mg/dL) were recorded. All the patients were found to be anti-HIV negative. Wound swab culture and sensitivity tests were also performed. Multi-drug resistance (MDR) was defined as resistance to more than 3 of the available antibiotics [12]. Biochemical parameters were measured on a Beckman Coulter AU5821 (Tokyo, Japan) device. The ESR values were measured using the Westergreen method on a StaRRsed device. Complete blood count was studied using a Sysmex XN-9000 (Kobe, Japan) device. HbA1c levels were calculated using the high-performance liquid chromatography method on a Premier Hb9210 device. The medical and surgical treatments that were administered were recorded.

Outcomes: The patients were followed-up until discharge or mortality during hospitalization. The patients with DFI remained enrolled in the study until

one of three pre-determined outcomes was observed during the ward follow-up period: improvement of DFI, major amputation, or mortality. Lesions that healed with no signs or findings of clinical infection and without amputation were regarded as cured.

Definitions: Amputations performed below the ankle were defined as minor, and those above the ankle as major [4]. Major amputation and mortality were regarded as adverse outcomes. The patients were divided into two groups based on the presence or absence of adverse outcomes.

Statistical analysis

The data were entered into and analyzed using IBM SPSS 25.0 (IBM SPSS Statistics for Windows, version 25.0. IBM Corp., Armonk, NY, USA). Demographic characteristics, underlying diseases, clinical findings, diabetic foot characteristics, laboratory parameters, and treatments were compared between the two groups in order to identify risk factors for adverse outcomes in patients with DFI. Categorical descriptive characteristics were expressed as frequency distributions and percentages, and continuous variables as median values (interquartile range). Categorical variables were compared between the groups using the Chi square test, and continuous variables using the non-parametric Mann-Whitney U test since parametric hypothesis test conditions were not met. Receiver operating characteristics (ROC) analysis was applied to estimate the ability of biomarkers and some continuous variables to predict mortality; cut-off points were determined, and the specificity and sensitivity of these were calculated. The Youden index (J = sensitivity + specificity – 1) was used to determine cut-off values. Risk factors for adverse outcomes were determined using univariate and multivariate logistic regression analysis. The multivariate logistic regression model was created with body temperature elevation (≥ 38.3

Table 1. Distributions of the patients' demographic characteristics and underlying diseases according to adverse events.

Characteristics	Total (n = 102)	Adverse outcomes		*p*
		No (n = 91)	Yes (n = 11)	
Gender (male), n (%)	74 (72.5)	66 (72.5)	8 (72.7)	0.648
Age, median (IQR)	60 (53–67)	60 (53–67)	61 (56–69)	0.447
Body mass index, (kg/m²)	27.4 (25.4–29.2)	27.4 (25.3–29.0)	27.7 (27.2–32.5)	0.268
Occupation, n (%)				0.194
Not working	31 (30.4)	29 (31.9)	2 (18.2)	
Self-employed	10 (9.8)	10 (11.0)	-	
Farmer	11 (10.8)	10 (11.0)	1 (9.1)	
Officer	5 (4.9)	4 (4.4)	1 (9.1)	
Retired	29 (28.4)	24 (26.4)	5 (45.5)	
Other	16 (15.7)	14 (15.4)	2 (18.2)	
Place of residence, n (%)				0.071
Village	29	28 (30.8)	1 (9.1)	
District	24	22 (24.2)	2 (18.2)	
Province	49	41 (45.1)	8 (72.7)	
Education level, n (%)				0.892
Not literate	23 (22.5)	21 (23.1)	2 (18.2)	
Literate	2 (2.0)	1 (1.1)	1 (9.1)	
Primary school graduate	58 (56.9)	52 (57.1)	6 (54.5)	
High school graduate	14 (13.7)	13 (14.3)	1 (9.1)	
Graduated from a Universty	5 (4.9)	4 (4.4)	1 (9.1)	
Underlying diseases, n (%)				
Coronary artery disease	42 (41.2)	36 (39.6)	6 (54.5)	0.340
Hypercholesterolemia	31 (30.4)	28 (30.8)	3 (27.3)	0.558
Hypertension	51 (50.0)	43 (47.3)	8 (72.7)	0.100
PAD	42 (41.2)	37 (40.7)	5 (45.59)	0.501
Peripheral venous insufficiency	24 (23.5)	19 (20.9)	5 (45.5)	0.080
Peripheral neuropathy	44 (50.6)	38 (48.7)	6 (66.7)	0.254
Duration of diabetes mellitus, (years), median (IQR)	15 (10–20)	15 (10–20)	10 (5–15)	0.083
Drug used for diabetes mellitus, n (%)				0.425
OAD	13 (12.7)	13 (14.3)	-	
Insulin	58 (56.9)	50 (54.9)	8 (72.7)	
OAD + insulin	28 (27.5)	26 (28.6)	2 (18.2)	
Not using	3 (2.9)	2 (2.2)	1 (9.1)	
Hospital admission due to the same DFI within the last 3 months	18 (17.6)	15 (16.5)	3 (27.3)	0.301
History of antibiotic use in the last 3 months	69 (67.6)	61 (67.0)	8 (72.7)	0.497
Smoking	32 (31.4)	28 (30.8)	4 (36.4)	0.473

DFI: diabetic foot infection; IQR: interquartile range; OAD: oral antidiabetic; PAD: peripheral artery disease.

°C), presence of wound necrosis or Enterobacteriaceae in wound culture, PEDIS grade 4, and BUN > 31 mg/dL, which emerged as significant in the univariate model (model: backward LR. entry: 0.05 and removal: 0.10). *p* values < 0.05 were regarded as statistically significant.

Results

The median age of the patients included in this study was 60.0 years (IQR: 53–67), and 74 (72.5%) were men. Amputation was performed on 32 patients (31.4%), 8 (7.9%) of which were major amputations. Mortality occurred in 4 (3.9%) cases during follow-up. Major amputation and/or mortality were regarded as adverse outcomes. Accordingly, 11 (10.8%) cases resulted in adverse outcomes. The patients' demographic characteristics and underlying diseases according to adverse events are presented in Table 1.

Distributions of clinical findings at the time of presentation and foot characteristics in terms of adverse events are summarized in Table 2. The foot characteristic of necrosis in the wound site was present in all the cases with adverse outcomes and was detected statistically significantly according to other features of the diabetic foot, such as fluid in the wound, and edema in the wound (Table 2; $p = 0.002$). PEDIS grade 4 was also significantly more common, and PEDIS grade 2 was significantly less common, in cases developing adverse outcomes ($p = 0.016$ and 0.012, respectively).

Distributions of patients' laboratory findings during presentation in terms of adverse events are summarized in Table 3. BUN levels were significantly higher in the cases concluding with adverse outcomes ($p = 0.001$). Ninety-five species of microorganisms were isolated from the wound cultures of 76 (74.5%) patients. Nineteen (18.6%) of these were polymicrobial. No microbial agent was identified in 26 patients (25.5%). Isolation of Enterobacteriaceae (n = 37, 36.3%) in wound culture was significantly more common in the cases resulting in adverse events ($p = 0.011$). MDR was detected in 15 (40.5%) of 37 Enterobacteriaceae isolates. The adverse outcomes were significantly higher in patients with MDR ($p = 0.004$).

No significant association was observed between the therapeutic regimen applied and adverse outcomes. The ROC curve of BUN levels for adverse outcomes during hospitalization is shown in Figure 1. The area under the curve was 0.762 (95% confidence interval [CI]: 0.641–0.884), and the cut-off value for predicting adverse outcomes was > 31 mg/dL ($p = 0.005$). Sensitivity in predicting adverse outcome at this cut-off value was 72.7%, with specificity of 71.4%.

The results of logistic regression analysis for adverse outcomes are presented in Table 4. Initial temperature elevation, presence of necrosis in the

Table 2. Distributions of clinical findings at the time of presentation and foot characteristics in terms of adverse events.

	Total (n = 102)	Adverse outcomes		*p*
		No (n = 91)	Yes (n = 11)	
Vital findings, n (%)				
Fever (≥ 38,3^{0}C)	7 (6.9)	4 (4.4)	3 (27.3)	**0.026**
Hypotension (< 90/60 mm/Hg)	2 (2.0)	1 (1.1)	1 (9.1)	0.205
Tachycardia (> 90 beats/min)	7 (6.9)	6 (6.6)	1 (9.1)	0.562
Wound formation time (days), median (IQR)	30 (15–60)	30 (15 – 60)	45 (30–90)	0.229
Affected side, n (%)				**0.270**
Left	47 (46.1)	40 (44.0)	7 (63.6)	
Right	41 (40.2)	38 (41.8)	3 (27.3)	
Both	14 (13.7)	13 (14.3)	1 (9.1)	
Localization of DFI, n (%)				**0.158**
Sole of foot	11 (10.8)	11 (12.1)	–	
Thumb	29 (28.4)	28 (30.8)	1 (9.1)	
Other fingers	31 (30.4)	27 (29.7)	4 (36.4)	
Heel	10 (9.8)	9 (9.9)	1 (9.1)	
Metatarsal	5 (4.9)	2 (2.2)	3 (27.3)	
Back of foot	6 (5.9)	5 (5.5)	1 (9.1)	
Stump place	3 (2.9)	2 (2.2)	1 (9.1)	
Lateral foot	3 (2.9)	3 (3.3)	–	
Ankle	4 (3.9)	4 (4.4)	–	
Fluid in the wound	89 (87.3)	79 (86.8)	10 (90.9)	0.576
Edema in the wound	86 (84.3)	77 (84.6)	8 (81.8)	0.545
Necrosis in the wound	47 (46.1)	37 (40.7)	10 (90.9)	**0.002**
Osteomyelitis	43 (42.2)	38 (41.8)	5 (45.5)	0.530
DFI severity score (PEDIS)				
Grade 2	34 (33.4)	34 (37.4)	–	**0.012**
Grade 3	62 (60.8)	54 (59.3)	8 (72.7)	0.390
Grade 4	6 (5.9)	3 (3.3)	3 (27.3)	**0.016**

DFI: diabetic foot infection; IQR: interquartile range; PEDIS: perfusion: extent: depth: infection: and sensation.

Table 3. Distributions of patients' laboratory findings during presentation in terms of adverse events.

	Total (n = 102)	Adverse outcomes		*p*
		No (n = 91)	Yes (n = 11)	
Microorganism isolation n(%)				
Polymicrobial	19 (18.6)	17 (18.7)	2 (18.2)	0.666
Enterobacteriaceae*	37 (36.3)	29 (31.9)	8 (72.7)	**0.011**
Multi-drug resistant**	15 (40.5)	8 (27.6)	7 (87.5)	**0.004**
Staphylococcus aureus	14 (13.7)	14 (15.4)	–	0.180
Non-fermenter Gram negative***	9 (8.8)	8 (8.8)	1 (9.1)	0.658
Other Gram positive****	35 (34.3)	32 (35.2)	3 (27.3)	0.438
No agent	26 (25.5)	25 (27.5)	1 (9.1)	0.171
Biomarker at the time of hospital admission				
Leukocyte (10^9/L)	10.18 (8.03–12.72)	10.37 (7.95–13.00)	9.72 (8.25–12.62)	0.978
Neutrophil–lymphocyte ratio	4.33 (2.73–7.42)	4.29 (2.75–7.18)	6.15 (2.53–11.71)	0.242
Hemoglobin (g/dL)	12.0 (10.9–13.8)	12.2 (10.9–13.8)	10.7 (8.8–13.6)	0.213
Platelet (10^9/L)	312 (247–374)	312 (247–375)	284 (247–367)	0.670
CRP (g/dL)	66 (20–138)	54 (15–138)	110 (50–171)	0.116
ESR (mm/hour)	61 (35–85)	61 (35–81)	64 (27–101)	0.340
BUN (mg/dL)	24 (17–39)	23 (17–34)	40 (25–58)	**0.005**
Creatinine (mg/dL)	1.1 (0.8–1.4)	1.0 (0.8–1.3)	1.1 (0.9–4.2)	0.084
Glucose (mg/dL)	235 (161–314)	234 (156–318)	250 (183–263)	0.842
AST (U/L)	18 (14–26)	18 (14–26)	20 (14–26)	0.812
ALT (U/L)	17 (12–25)	17 (12–25)	19 (11–25)	0.957
HbA1c (%)	9.4 (8.2–11.0)	9.4 (8.2–11.1)	8.9 (7.5–10.9)	0.323
Total cholesterol (mg/dL)	159 (133–190)	159 (136–186)	156 (118–225)	0.840
LDLc (mg/dL)	102 (87–123)	102 (87–123)	89 (78.5–144)	0.655
Albumin (g/dL)	2.9 (2.7–3.5)	2.9 (2.6–3.5)	2.8 (2.7–3.5)	0.530

Escherichia coli, Klebsiella pneumoniae, Enterobacter spp., Proteus spp., Citrobacter spp., Morganella morganii, Serratia marcescens.* ** *Multi-drug resistant among 37 Enterobacteriaceae spp. isolates.* * *Pseudomonas aeruginosa, Acinetobacter baumannii.* **** *Staphylococcus epidermidis, Enterococcus spp., Streptococcus spp.* ALT: alanine aminotransferase; AST: aspartate aminotransferase; BUN: blood urea nitrogen; CRP: C-reactive protein; DFI: diabetic foot infection; ESR: erythrocyte sedimentation rate; LDLc: low density lipoprotein cholesterol.

wound site, detection of Enterobacteriaceae in wound culture, PEDIS grade 4, and BUN > 31 mg/dL were found to be significant based on univariate logistic regression analysis. The multivariate logistic regression analysis of these parameters determined that the presence of necrosis in the wound increased the risk of adverse outcomes by 20.125 times, and BUN exceeding 31 mg/dL increased the risk by 9.454 times; and these were independent risk factors.

Discussion

Patients with DFI are at an increased risk of major amputation or mortality with the presence of chronic ischemia threatening the lower extremities [10]. Previous studies have described various risk factors for major amputation and mortality in patients with DM. In the present study, fever, presence of necrosis in the wound site, isolation of Enterobacteriaceae species from wound culture, PEDIS grade 4, and BUN > 31 mg/dL emerged as predictive factors for adverse outcomes in patients with DFI. Based on the model

Figure 1. Receiver operating curve (ROC) curve of blood urea nitrogen levels at hospitalization for adverse outcomes.

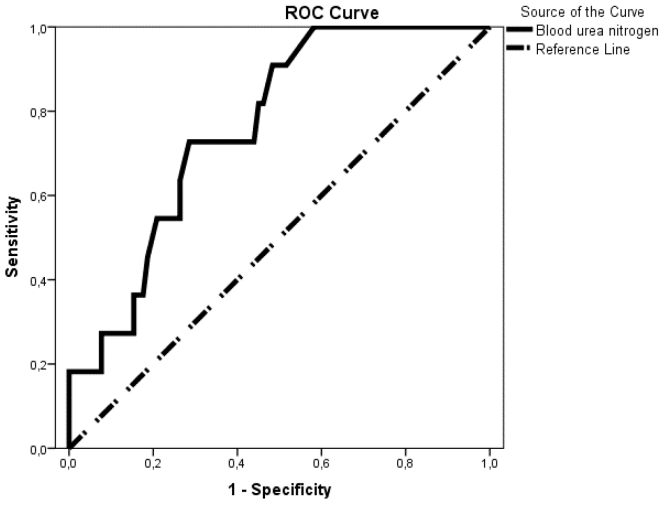

Table 4. Logistic regression analysis of adverse outcomes.

	Univariate OR (95% CI)	*p*	Multivariate OR (95% CI)	*p*
Fever (≥ 38,3 °C)	8.156 (1.546–43.020)	0.013		
Necrosis in the wound	14.595 (1.791–118.914)	0.012	20.125 (2.293–176.611)	**0.007**
Enterobacteriaceae in the wound	5.701 (1.408–23.079)	0.015		
PEDIS grade 4	11.000 (1.899–63.705)	0.007		
BUN > 31 mg/dL	6.667 (1.640–27.107)	0.008	9.454 (2.072–43.132)	**0.004**

BUN: blood urea nitrogen; CI: confidence interval; OR: odds ratio; PEDIS: perfusion: extent: depth: infection: and sensation.

constructed with these risk factors, BUN exceeding 31 mg/dL and the presence of necrosis in the wound site were identified as independent predictive factors for adverse outcome development.

Nephropathy is one of the microvascular complications of DM [13]. However, studies examining the relationship between renal function and major amputation have reported inconsistent results. While some research has described impaired renal function as a predictive factor for major amputation, other studies have found no association between kidney function, urea levels, and amputation or major amputation in patients with DM [5,13–15]. Our own findings indicate that increased BUN in patients with DFI is associated with adverse outcomes. A BUN value exceeding 31 mg/dL predicted adverse outcomes with 72.7% sensitivity and 71.4% specificity. Adeleye *et al.* reported that kidney failure was an independent predictor of mortality in patients with DFU [16]. Diabetic nephropathy is associated with an increased risk of neuropathy and PAD [17]. Studies have reported that, high levels of BUN in severe infections other than DFI occur as a result of tissue damage, increased protein breakdown, and renal urea reabsorption due to dehydration [18,19]. In addition, kidney failure can lead to mortality in patients with DFI by triggering such cardiovascular risk factors as oxidative stress and inflammation [16]. The elevation of BUN in cases concluding with mortality and major amputation in the present study was associated with the involvement of more than one of the risk factors described above.

Varying degrees of necrosis may be seen due to peripheral circulation impairment deriving from micro- or macrovascular complications in patients with DFI. Previous studies have shown that the presence of necrosis in the wound is associated with an increased risk of major amputation [16,20]. Consistent with previous research, in our study, the presence of necrosis in the wound site resulted in 20 times higher risk of adverse outcome development [16,20]. A recent study from Nigeria reported similar findings, to the effect that mortality rates were 8 times higher in patients with DFU with gangrene than in those without gangrene [16]. Shatnawi *et al*. also reported that gangrene resulted in 4.2 times increased risk of major amputation, while kidney failure raised it 3.5 times [20]. Impaired arterial perfusion can also represent a cause of amputation in diabetics. The presence of necrosis in the wound, together with the presence of PAD and atherosclerosis in other vessels can also cause an increased risk of cardiac death.

Body temperature elevation, one of the criteria for SIRS, also emerged as a potential risk factor for the development of adverse outcomes in this study. This finding is consistent with other studies of major amputation [5,14,15].

Previous studies examining the relationship between the microbial agent in diabetic foot and the development of major amputation or mortality have reported varying results. The limited available data concerning pathogens associated with mortality suggest that polymicrobial infection with *Pseudomonas* spp. and the monomicrobial isolation of other Gram-negative microorganisms may be associated with poorer outcomes in soft tissue infections in hospitalized diabetic patients [21]. According to a report by Cardosa *et al*., *Acinetobacter* spp. and *Klebsiella* spp. isolated in the wound cultures of patients with DFI were associated with a greater risk of major amputation [22]. Fejfarova *et al*. reported a statistically significant greater presence of resistant *Staphylococcus* spp. in patients with DM with lower extremity amputation compared to non-amputated patients [23]. In contrast to previous studies, no significant effect of *Staphylococcus aureus* isolation on adverse outcome development was observed in the present study. This may be attributable to the limited number of patients with *S. aureus* as a causative agent and the fact that these were all included in the group without adverse outcome development. Enterobacteriaceae species (n = 37, 36.3%) were the agents that were most frequently isolated from our patients' wound culture specimens. Zubair *et al*. found that approximately 43.5% of 278 isolates were associated with MDR, which is similar to the 40.5% of Enterobacteriaceae species isolates that were associated with MDR in our study [24]. Our study found that microorganisms belonging to Enterobacteriaceae species and MDR Enterobacteriaceae species were more frequently detected in patients with adverse outcomes. However, this did not represent an independent risk factor. Surme *et al*. determined that MDR *Pseudomonas aeruginosa* was predictive of poor prognosis [25]. The fimbriae and capsule with colonizing ability, toxin production, and tissue invasion, all play a role among the factors determining the virulence of Enterobacteriaceae species [26]. Therefore, surveillance of MDR bacteria is needed to reduce the risk of adverse outcomes.

The PEDIS classification, that is widely employed in clinical practice, exhibited a significant effect on the development of adverse outcomes in this study. PEDIS grade 4 severity was detected significantly more frequently in patients developing adverse outcomes.

Similar to our findings, Chaudhary *et al.* reported that PEDIS grades in patients with DFI had a significant impact on major amputation and mortality rates [27]. Since PEDIS grade 4 severity indicates the presence of diffuse cellulitis or deep infection SIRS, a relationship between higher PEDIS grades and major amputation or mortality is an expected finding

There were a number of limitations to this study. First, all the patients were enrolled from a single center. Various risk factors for major amputation and mortality have been reported in the literature, such as age, male gender, smoking, type 2 DM, high BMI, hypertension, dyslipidemia, weak glycemic control, peripheral neuropathy, and PAD. The patient profiles, ethnic origins, and cultural characteristics in these studies may have given rise to this variation. The number of patients is also a limitation of the present study, and may therefore be usefully increased in future multi-center research. Another limitation in this study is the use of swab cultures, since we were unable to obtain intraoperative cultures. Since microorganisms grown in swab cultures generally reflect surface colonization, they may be insufficient for the identification of infectious agents. It was not possible to obtain deep tissue culture, which is the most ideal method in terms of microbiology. Therefore, swab cultures obtained from our patients were a limitation of our study, and may have been a factor affecting microbiological results. However, the strength of this study lies in its prospective nature.

Conclusions

Among the factors analyzed in this study, BUN > 31 mg/dL and the presence of necrosis in the wound emerged as independent risk factors for adverse events in patients with DFI.

DFI is associated with high morbidity and mortality. Understanding the risk factors for adverse outcomes in patients with DFI, and developing strategies to control these will help ensure that patients with DFI receive appropriate medical care and prevent major amputation or mortality.

Corresponding author

Rukiye İnan Sarıkaya, MD.
Department of Infectious Diseases and Clinical Microbiology, Health Sciences University,
Erzurum Regional Education and Research Hospital, Erzurum, Yakutiye, 25240, Turkey.
Tel: +90 4422317428,
Fax: +90 4423160777, +90 5385017899
Email: rukiinan@hotmail.com

Conflict of interests

No conflict of interests is declared.

References

1. International Diabetes Federation (2021) IDF Diabetes Atlas, Tenth edition. Available: https://diabetesatlas.org/atlas/tenth-edition/. Accessed: 10 May 2023.
2. Lipsky BA, Senneville É, Abbas ZG, Aragón-Sánchez J, Diggle M, Embil JM, Kono S, Lavery LA, Malone M, van Asten SA (2020) Guidelines on the diagnosis and treatment of foot infection in persons with diabetes (IWGDF 2019 update). Diabetes Metab Res Rev 36: e3280. doi: 10.1002/dmrr.3280.
3. Abdissa D, Adugna T, Gerema U, Dereje D (2020) Prevalence of diabetic foot ulcer and associated factors among adult diabetic patients on follow-up clinic at Jimma Medical Center, Southwest Ethiopia, 2019: an institutional-based cross-sectional study. J Diabetes Res 2020: 4106383. doi: 10.1155/2020/4106383.
4. Thorud JC, Plemmons B, Buckley CJ, Shibuya N, Jupiter DC (2016) Mortality after nontraumatic major amputation among patients with diabetes and peripheral vascular disease: a systematic review. J Foot Ankle Surg 55: 591–599. doi: 10.1053/j.jfas.2016.01.012.
5. Yusof NM, Rahman JA, Zulkifly AH, Che-Ahmad A, Khalid KA, Sulong AF, Vijayasingham N (2015) Predictors of major lower limb amputation among type II diabetic patients admitted for diabetic foot problems. Singapore Med J 56: 626–631. doi: 10.11622/smedj.2015172.
6. Vuorlaakso M, Kiiski J, Salonen T, Karppelin M, Helminen M, Kaartinen I (2021) Major amputation profoundly increases mortality in patients with diabetic foot infection. Front Surg 8: 655902. doi: 10.3389/fsurg.2021.655902.
7. Armstrong DG, Swerdlow MA, Armstrong AA, Conte MS, Padula WV, Bus SA (2020) Five year mortality and direct costs of care for people with diabetic foot complications are comparable to cancer. J Foot Ankle Res 13: 1–4. doi: 10.1186/s13047-020-00383-2.
8. Waters RL, Perry J, Antonelli D, Hislop H (1976) Energy cost of walking of amputees: the influence of level of amputation. JBJS 58: 42–46. doi: 10.2106/00004623-197658010-00007.
9. Morbach S, Furchert H, Gröblinghoff U, Hoffmeier H, Kersten K, Klauke G-T, Klemp U, Roden T, Icks A, Haastert B (2012) Long-term prognosis of diabetic foot patients and their limbs: amputation and death over the course of a decade. Diabetes Care 35: 2021–2027. doi: 10.2337/dc12-0200.
10. Almohammadi AA, Alnashri MM, Abdulrahman THR, Alsamiri SM, Alkhatieb MT (2022) Pattern and type of amputation and mortality rate associated with diabetic foot in Jeddah, Saudi Arabia: a retrospective cohort study. Ann Med Surg (Lond) 73: 103174. doi: 10.1016/j.amsu.2021.103174.
11. Schaper N (2004) Diabetic foot ulcer classification system for research purposes: a progress report on criteria for including

patients in research studies. Diabetes Metab Res Rev 20 Suppl 1: S90–S95. doi: 10.1002/dmrr.464.
12. Falagas ME, Rafailidis PI, Matthaiou DK, Virtzili S, Nikita D, Michalopoulos A (2008) Pan-drug-resistant *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumannii* infections: characteristics and outcome in a series of 28 patients. Int J Antimicrob Agents 32: 450–454. doi: 10.1016/j.ijantimicag.2008.05.016.
13. Margolis DJ, Hofstad O, Feldman HI (2008) Association between renal failure and foot ulcer or lower-extremity amputation in patients with diabetes. Diabetes Care 31: 1331–1336. doi: 10.2337/dc07-2244.
14. Kow RY, Low CL, Ruben JK, Zaharul-Azri MZ, Lim BC (2019) Predictive factors of major lower extremity amputations in diabetic foot infections: a cross-sectional study at district hospital in Malaysia. Malays Orthop J 13: 45–52. doi: 10.5704/MOJ.1911.008.
15. Aziz Z, Lin WK, Nather A, Huak CY (2011) Predictive factors for lower extremity amputations in diabetic foot infections. Diabet Foot Ankle 2011: 2. doi: 10.3402/dfa.v2i0.7463.
16. Adeleye OO, Ugwu ET, Gezawa ID, Okpe I, Ezeani I, Enamino M (2020) Predictors of intra-hospital mortality in patients with diabetic foot ulcers in Nigeria: data from the MEDFUN study. BMC Endocr Disord 20: 134. doi: 10.1186/s12902-020-00614-4.
17. Eggers PW, Gohdes D, Pugh J (1999) Nontraumatic lower extremity amputations in the medicare end-stage renal disease population. Kidney Int 56: 1524–1533. doi: 10.1046/j.1523-1755.1999.00668.x.
18. Han T, Cheng T, Liao Y, Tang S, Liu B, He Y, Gu Z, Lei C, Cao Y, Cao Y (2022) Analysis of the value of the blood urea nitrogen to albumin ratio as a predictor of mortality in patients with sepsis. J Inflamm Res 15: 1227–1235. doi: 10.2147/JIR.S356893.
19. Feng DY, Zhou YQ, Zou XL, Zhou M, Yang HL, Chen XX, Zhang TT (2019) Elevated blood urea nitrogen-to-serum albumin ratio as a factor that negatively affects the mortality of patients with hospital-acquired pneumonia. Can J Infect Dis Med Microbiol 2019: 1547405. doi: 10.1155/2019/1547405.
20. Shatnawi NJ, Al-Zoubi NA, Hawamdeh HM, Khader YS, Garaibeh K, Heis HA (2018) Predictors of major lower limb amputation in type 2 diabetic patients referred for hospital care with diabetic foot syndrome. Diabetes Metab Syndr Obes 11: 313–319. doi: 10.2147/DMSO.S165967.
21. Lipsky BA, Tabak YP, Johannes RS, Vo L, Hyde L, Weigelt JA (2010) Skin and soft tissue infections in hospitalised patients with diabetes: culture isolates and risk factors associated with mortality, length of stay and cost. Diabetologia 53: 914–923. doi: 10.1007/s00125-010-1672-5.
22. Cardoso NA, Cisneiros LL, Machado CJ, Cenedezi JM, Procópio RJ, Navarro TP (2017) Bacterial genus is a risk factor for major amputation in patients with diabetic foot. Rev Col Bras Cir 44: 147–153. doi: 10.1590/0100-69912017002007.
23. Fejfarová V, Jirkovská A, Skibová J, Petkov V (2002) Pathogen resistance and other risk factors in the frequency of lower limb amputations in patients with the diabetic foot syndrome. Vnitr Lek 48: 302–306. [Article in Czech].
24. Zubair M, Ahmad J (2019) Potential risk factors and outcomes of infection with multidrug resistance among diabetic patients having ulcers: 7 years study. Diabetes Metab Syndr 13: 414–418. doi: 10.1016/j.dsx.2018.10.014.
25. Surme S, Saltoglu N, Kurt AF, Karaali R, Balkan, II, Baghaki S, Caglar B, Ozdemir M, Vatan A, Togluk-Yigitoglu E, Budak B, Arapi B, Seker A, Can G, Gonen MS, Cetinkale O (2022) Changing bacterial etiology and antimicrobial resistance profiles as prognostic determinants of diabetic foot infections: a ten-year retrospective cohort study. Surg Infect (Larchmt) 23: 667–674. doi: 10.1089/sur.2022.150.
26. Mahon C, Lehman DC, Manuselis G (2019) Textbook of diagnostic microbiology. 6th edition. St Louis MO: Elsevier Inc. 415 p.
27. Chaudhary N, Huda F, Roshan R, Basu S, Rajput D, Singh SK (2021) Lower limb amputation rates in patients with diabetes and an infected foot ulcer: a prospective observational study. Wound Manag Prev 67: 22–30. doi: 10.25270/wmp.2021.7.2230.