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Factors Affecting Vascular Clogging, Wound Status and Bacterial Culture in Diabetic Foot Ulcers

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Abstract

Background: We assumed that vascular clogging, wound status, and bacterial culture tests are significant characteristics in developing a treatment plan for diabetic patients with foot ulcers. The goal of our study was to find out the potential factors affecting vascular clogging, wound status, and bacterial culture tests in diabetic patients with foot ulcers.
Methods: The medical records of diabetic patients with foot ulcers presenting at our institution between January 2016 and December 2017 were reviewed. A total of 58 diabetic patients with foot ulcers were included. Patient baseline characteristics including co-morbidities, as well as foot and ulcer characteristics, were analyzed.
Results: By using the univariate and multivariate binary logistic regression analysis, we have found significant factors affecting vascular clogging, wound status and bacterial culture test. Age, glomerular filtration rate (GFR), and glycosylated hemoglobin (HbA1c) were significant factors influencing vascular status. Age, dialysis, GFR, white blood cell (WBC), and osteomyelitis were significant factors influencing wound status. Also, sex (male), dialysis and high WBC affect the possibility of Gram-negative aerobic bacterial infection more so than other bacterial infection.
Conclusion: This study has confirmed significant factors affecting vascular status, wound status, and bacterial culture tests, including co-morbidities, as well as foot and ulcer characteristics at baseline. Physicians should consider these factors in developing treatment plans or predicting outcomes of diabetic foot ulcers.

Keywords: Diabetic foot ulcer; Wound classification; Vascular clogging; Bacterial culture

Introduction

Diabetes is one of the leading causes of chronic disease and limb loss worldwide. It affected about 451 million (age of 18–99 years) people in 2017. By 2045, the number of reported diabetes cases are expected to increase to 693 million. Diabetic foot problems are common around the world, resulting in major difficulties in daily lives and also economic consequences for patients, their families, and society in general.

It is very important for clinicians to know which of the available clinical and laboratory findings are associated with poor outcomes in diabetic patients with foot ulcers. Prediction of outcomes in diabetic patients with foot ulcers might be helpful for clinicians to optimize and individualize management strategies.

In previous studies, peripheral artery diseases (PAD), presence of infection, and diabetic foot ulcer classification according to staging systems such as perfusion, extent, depth, infection and sensation (PEDIS)/Infectious Diseases Society of America (IDSA) classification grades or diabetic foot infection (DFI) wound scores have all been shown to be prognostic factors for diabetic foot disease [1-6]. Therefore, we assumed that vascular clogging, wound status, and bacterial culture tests are relevant in developing a treatment plan or predicting the prognosis of diabetic patients with foot ulcers.

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The goal of our study was to find out the potential factors affecting vascular clogging, wound status, and bacterial culture tests in diabetic patients with foot ulcers.

Methods

Patients and data collection

The study was approved by the Institutional Review Board of Chonbuk National University Hospital (IRB No. 2016-10-020-002) and performed in accordance with the principles of the Declaration of Helsinki. The informed consent was waived. The medical records of diabetic patients with foot ulcers presenting at our institution between January 2016 and December 2017 were reviewed. Data on age, sex, history of smoking, duration of diabetes mellitus, diabetic foot ulcer classification (PEDIS/IDSA grade, and DFI wound score), wound angiosome, underlying disease (hypertension, congestive heart failure, dyslipidemia, chronic kidney disease [CKD] stage), major vascular disease, history of dialysis, bacterial culture results, and blood test results (including glycosylated hemoglobin [HbA1c], white blood cell [WBC], creatinine [Cr], glomerular filtration rate [GFR]) had been obtained on admission to the hospital.

The results of all tests and epidemiological characteristics were checked via the medical records. All patients were examined for wound status, vascular clogging, and pathologic culture on admission.

Each ulcer was classified using both the PEDIS/IDSA grade and the DFI wound score. The IDSA and the International Working Group on the Diabetic Foot developed a system for classifying diabetic foot wounds. The system is referred to by the acronym PEDIS, which is comprised of the first letters of perfusion, extent (size), depth (tissue loss), infection, and sensation (neuropathy). PEDIS grades are from 1 to 4 (uninfected, mild, moderate, and severe).

Lipsky et al. [7] developed a 10-item scoring system to measure outcomes in studies of various antimicrobial treatments for DFIs. The scoring system consists of a semi-quantitative assessment of the signs of inflammation, combined with measurements of both wound size and depth. Explicit definitions, which allow numerical scoring of wound parameters, are provided. Wound measurements consist of size, depth, and the amount of undermining. The10-item wound score ranges from 3 to 49.

The ulcer site was documented and categorized as angiosome, including anterior tibial artery, posterior tibial artery, and multiple vessels.

Computed tomographic angiography of lower extremities is usually performed to check the vascular status. However, because of the risk of contrast-induced nephropathy, patients with severely decreased kidney function were tested by radiologists using color Doppler ultrasonography on lower extremity vessels. The results were classified as either normal or abnormal, noting whether there were clogs in single or multiple vessels. For patients with abnormal vascular status, percutaneous transluminal angiography was performed, and if needed, the clogged vessels were revascularized [8-11]. Diabetic foot ulcer was classified using the two systems, the PEDIS/IDSA grade and the DFI wound score, at the time of admission. Debridement was performed by removing all necrotic tissues. Bacterial culture was performed using the necrotic tissue obtained.

Three-phase bone scan was performed for suspected osteomyelitis (e.g., ulcer depth >3 mm, purulence in exposed bone of foot, or erythrocyte sedimentation rate >60 mm/hr, etc.) at the time of admission. Bone biopsy culture, a quite invasive method to evaluate osteomyelitis, was sometimes used in the operating room, but we have not adopted bone biopsy culture as a routine method.

Statistical analysis

R language version 3.3.3 (R Foundation for Statistical Computing, Vienna, Austria) and T&F program version 2.5 (YooJin BioSoft, Seoul, Korea) were used for all statistical analyses.

Binary logistic regression analysis was performed, and for analyzing the combined effect of more than two variables, multivariable logistic regression analysis was performed using the backward stepwise procedure as the variable selection method to minimize Akaike information criterion.

Univariate and multivariate binary logistic regressions were performed for PEDIS/IDSA grades (3, 4 vs. 1, 2), 10-item DFI wound score (cutoff, 20), vascular clogging (yes vs. no), and bacterial culture (others vs. Gram-aerobic).

Results

A total of 58 diabetic patients with foot ulcers were included. The characteristics of these patients are shown in Tables 1-3. The mean age of participants with ulcers was 60.5±13.4 years. There were 45 men and 13 women. The mean duration of diabetes was 16.26±7.29 years. The PEDIS/IDSA grade was 1 in 11 cases (19%), 2 in 18 cases (31%), 3 in 28 cases (48.3%), and

Table 1. Baseline characteristics (n=58)

Variable	Value
Sex	
Female	13 (22.4)
Male	45 (77.6)
Age (yr)	60.48±13.43
PEDIS/IDSA grade	
1	11 (19)
2	18 (31)
3	28 (48.3)
4	1 (1.7)
Wound parameters (10-item)	7.0±4.5
Wound parameters (8-item)	4.7±3.1
Wound measurements (subtotal)	9.5±4.6
DFI wound score (10-item)	16.6±7.6
DFI wound score (8-item)	14.2±6.2
Wound angiosome	
ATA	11 (18.9)
PTA	19 (32.8)
Multi	28 (48.3)
Wound angiosome	
Single	30 (51.7)
Multi	28 (48.3)
Vascular clogging	
No	38 (65.5)
SFA	4 (6.9)
PTA	1 (1.7)
Multifocal artery	15 (25.9)
Bacterial culture	
Gram-positive aerobic	22 (37.9)
Gram-positive & -negative aerobic	4 (6.9)
Gram-negative aerobic	25 (43.1)
Others	7 (12.1)

Values are presented as number (%) or mean±SD.
PEDIS, perfusion, extent, depth, infection and sensation; IDSA, Infectious Diseases Society of America; DFI, diabetic foot infection; ATA, anterior tibial artery; PTA, posterior tibial artery; SFA, superior femoral artery.

4 in one case (1.7%). The mean DFI wound score (10-item) was 16.6±7.6. With respect to vascular clogging, 38 (65.5%) showed no clogging, five (8.6%) showed single vessel blockage, and 15 (25.9%) showed multi vessels blockage. Regarding bacterial cultures, 22 (37.9%) showed Gram-positive aerobic, 25 (43.1%) showed Gram-negative aerobic, four (6.9%) showed Gram-positive & negative aerobic, and seven (12.1%) showed

others.

Tables 4 and 5 show the univariate and multivariate binary logistic regression analyses for significant factors affecting vascular clogging in diabetic foot ulcer infection. According to the univariate analysis, age, Cr, and GFR were determined as significant factors affecting vascular status; whereas age, GFR, and HbA1c were found out to be significant factors influencing vascular status via multivariate analysis.

Tables 6-9 show the univariate and multivariate binary logistic regression analyses for the significant factors affecting diabetic foot ulcer classification in diabetic foot ulcer infection. In univariate analysis, WBC was the significant factor affecting diabetic foot ulcer classification (PEDIS/IDSA grade); whereas age, dialysis, GFR, WBC, and osteomyelitis were significant factors influencing wound status with diabetic foot ulcer classification in multivariate analysis (PEDIS/IDSA grade). Based on univariate analysis, osteomyelitis was the significant factor affecting diabetic foot ulcer classification (10-item DFI score); whereas in multivariate analysis, age, dialysis, GFR, WBC, and osteomyelitis were found out to be significant factors influencing wound status based on the wound classifications (10-item DFI score).

Tables 10 and 11 show the univariate and multivariate binary logistic regression analyses for the significant factors affecting bacterial culture (Gram-aerobic vs. others) in diabetic foot ulcer infection. According to the univariate analysis, low HbA1c affects the possibility of Gram-negative aerobic bacterial infection more than other bacterial infections, whereas in multivariate analysis, sex (male), dialysis, and high WBC affect the possibility of Gram-negative aerobic bacterial infection more than other bacterial infections.

Discussion

One of the most common complications of diabetes is the development of foot ulcers. Diabetic foot ulcers are related with serious morbidity as well as increased mortality. Therefore, understanding the factors that may contribute to the cure of diabetic foot ulcer would be helpful in both prevention and management strategies.

We have considered the factors affecting the treatment and prognosis of patients with diabetic foot ulcers as vascular clogging, current wound status, and the bacterial culture test, affecting antibiotic therapy. The severity and prediction of these three aspects can be helpful in predicting the prognosis of patients with diabetic foot ulcers and in determining the treat-

Table 2. Baseline characteristics of patient history (n=58)

Variable	Value
Final status	
No infection	56 (96.6)
Hopeless	1 (1.7)
Major amputation	1 (1.7)
DM duration (yr)	16.26±7.29
Ulcer (history, n=25)	
No	2 (8)
Yes	23 (92)
Antibiotic treatment (history, n=24)	
No	2 (8.3)
Yes	22 (91.7)
Smoking	
No	34 (58.6)
Yes	24 (41.4)
Hypertension	
No	24 (41.4)
Yes	34 (58.6)
Retinopathy (n=33)	
No	7 (21.2)
Yes	26 (78.8)
Nephropathy	
No	31 (53.4)
Yes	27 (46.6)
Neuropathy	
No	47 (81)
Yes	11 (19)
IHD (coronary insufficiency)	
No	51 (87.9)
Yes	7 (12.1)
Coronary artery angioplasty	
No	53 (91.4)
Yes	5 (8.6)
Coronary artery bypass surgery	
No	57 (98.3)
Yes	1 (1.7)
Congestive heart failure	
No	51 (87.9)
Yes	7 (12.1)
Cerebrovascular disease	
No	57 (98.3)
Yes	1 (1.7)

(*Continued to the next*)

Table 2. Continued

Variable	Value
Previous revascularization surgery (including PTA)	9 (100)
Dyslipidemia (n=2)	
No	1 (50)
Yes	1 (50)
Dialysis	
No	42 (72.4)
Yes	16 (27.6)
CKD stage	
Not related	31 (53.4)
DDKT	1 (1.7)
Stage 2	1 (1.7)
Stage 3	6 (10.3)
Stage 3–4	1 (1.7)
Stage 4	1 (1.7)
Stage 4–5	1 (1.7)
Stage 5	16 (27.6)

Values are presented as number (%) or mean±SD.
DM, diabetes mellitus; IHD, ischemic heart disease; PTA, posterior tibial artery; CKD, chronic kidney disease; DDKT, death donor kidney transplantation.

Table 3. Baseline characteristics of blood test and osteomyelitis (n=58)

Variable	Value
Creatinine (mg/dL)	2.5±2.6
GFR (mL/min)	57.9±37.2
Hb1Ac (%)	8.4±2.4
WBC (×10³/μL)	10.7±5.1
Osteomyelitis	
No	36 (62.1)
Yes	22 (37.9)

Values are presented as mean±SD or number (%).
GFR, glomerular filtration rate; HbA1c, glycosylated hemoglobi; WBC, white blood cell.

ment strategy.

Recently, a number of larger studies reported the possible determinants of outcome in diabetic foot disease, such as the UK multicenter study initiated by Ince et al. [1] and the EU-RODIALE study [3]. These studies found that the presence of PAD was an independent predictor of the outcome. However, to the best of our knowledge, no previous studies had systematically–via multivariable analysis–assessed the potential factors that contribute to PAD, including co-morbidities, as well

Table 4. Univariate binary logistic regression analysis using vascular clogging (yes vs. no)

Predictor	Subgroup	OR (95% CI)	P-value
Sex	Male vs. female	2.024 (0.487–8.407)	0.332
Age		1.062 (1.012–1.114)	0.013[a]
PEDIS/IDSA grade (1–4)	2 vs.1	2.667 (0.529–13.433)	0.234
	3 vs.1	1.067 (0.224–5.075)	0.935
	4 vs.1	0 (0–infinity)	0.992
	(3 or 4) vs. (1 or 2)	0.540 (0.180–1.621)	0.272
Wound parameters (10-item)		0.958 (0.848–1.083)	0.497
DFI wound score (10-item)		0.976 (0.907–1.049)	0.506
10-Item DFI score (cutoff=20)	≥20 vs. <20	0.458 (0.138–1.521)	0.202
Wound angiosome (ATA, PTA, multi)	PTA vs. ATA	2.625 (0.437–15.777)	0.464
	Multi vs. ATA	2.191 (0.527–16.093)	0.45
Wound angiosome	Multi vs. single	1.510 (0.508–4.484)	0.458
Bacterial culture	Others vs. Gram-positive aerobic	1.211 (0.393–3.734)	0.505
	Others vs. Gram-negative aerobic	1.214 (0.404–3.647)	0.729
DM duration		1.025 (0.951–1.104)	0.522
Ulcer (history)	Yes vs. no	0.150 (0.007–3.092)	0.219
Antibiotic treatment (history)	Yes vs. no	0.222 (0.011–4.358)	0.322
Smoking	Yes vs. no	0.917 (0.304–2.760)	0.877
Hypertension	Yes vs. no	3.000 (0.908–9.914)	0.072
Retinopathy	Yes vs. no	1.111 (0.177–6.991)	0.911
Nephropathy	Yes vs. no	2.300 (0.761–6.952)	0.14
Neuropathy	Yes vs. no	0.662 (0.155–2.833)	0.578
IHD (coronary insufficiency)	Yes vs. no	1.500 (0.301–7.476)	0.621
Coronary artery angioplasty	Yes vs. no	1.296 (0.198–8.473)	0.786
Congestive heart failure	Yes vs. no	2.917 (0.583–14.586)	0.192
Cerebrovascular disease	Yes vs. no	0 (0–infinity)	0.992
Dyslipidemia	Yes vs. no	1.000 (0–infinity)	1
Dialysis	Yes vs. no	2.500 (0.763–8.192)	0.13
Creatinine		1.254 (1.010–1.558)	0.041[a]
GFR		0.973 (0.956–0.991)	0.003[b]
Hb1Ac		0.935 (0.723–1.209)	0.608
WBC		0.969 (0.866–1.084)	0.576
Osteomyelitis	Yes vs. no	0.826 (0.268–2.545)	0.739

OR, odds ratio; CI, confidence interval; PEDIS, perfusion, extent, depth, infection and sensation; IDSA, Infectious Diseases Society of America; DFI, diabetic foot infection; ATA, anterior tibial artery; PTA, posterior tibial artery; DM, diabetes mellitus; IHD, ischemic heart disease; GFR, glomerular filtration rate; HbA1c, glycosylated hemoglobin; WBC, white blood cell.
[a]$P<0.05$; [b]$P<0.01$.

as foot and ulcer characteristics at baseline. In our analysis, older age, decreased GFR, and HbA1c were significant factors contributing to the presence of PAD in diabetic foot ulcers patients (Fig. 1).

Aging is a significant predictor of the risk of limb amputa- tion and hospital mortality in diabetic patients with foot ulcers [12]. Aged patients usually have progressive atherosclerosis with peripheral neuropathy, CKD, as well as PAD [13-15]. Our study has revealed that older agepatients have greater PAD among diabetic foot ulcers patients.

Table 5. Multivariate binary logistic regression analysis using vascular clogging (yes vs. no)

Predictor	OR (95% CI)	P-value
Age	1.063 (1.001–1.129)	0.048[a]
GFR	0.962 (0.937–0.988)	0.004[b]
Hb1Ac	1.356 (0.887–2.074)	0.16

OR, odds ratio; CI, confidence interval; GFR, glomerular filtration rate; HbA1c, glycosylated hemoglobin.
[a]$P < 0.05$; [b]$P < 0.01$.

The etiologies of CKD and diabetic foot ulcers have many common pathogenetic pathways, with both etiologies involving vasculopathy and microangiopathy. In the kidney, the pathogenic pathways lead to endothelial dysfunction, activation of Ras and transforming growth factor-β, inflammation with podocytes, and mesangial cell death. All paths ultimately lead to renal vasoconstriction, fibrosis that leads to glomerulosclerosis, and tubular degeneration, resulting in a decrease in renal blood flow, GFR, and renal function. Several studies have

Table 6. Univariate binary logistic regression analysis using PEDIS/IDSA grade (3, 4 vs. 1, 2)

Predictor	Subgroup	OR (95% CI)	P-value
Sex	Male vs. female	0.820 (0.238–2.825)	0.753
Age		1.020 (0.981–1.061)	0.318
Vascular clogging	SFA vs. no	0.270 (0.026–2.835)	0.275
	PTA vs. no	0 (0–infinity)	0.991
	SFA or PTA vs. no	0.202 (0.021–1.984)	0.17
	Multifocal artery vs. no	0.708 (0.214–2.35)	0.573
	Yes vs. no	0.540 (0.180–1.621)	0.272
Bacterial culture	Others vs. Gram-positive aerobic	1.341 (0.463–3.887)	0.589
	Others vs. Gram-negative aerobic	0.869 (0.307–2.458)	0.791
DM duration (yr)		1.003 (0.934–1.077)	0.928
Ulcer (history)	Yes vs. no	1.300 (0.072–23.435)	0.859
Antibiotic treatment (history)	Yes vs. no	1.444 (0.080–26.231)	0.804
Smoking	Yes vs. no	0.752 (0.264–2.145)	0.594
Hypertension	Yes vs. no	1.773 (0.616–5.102)	0.288
Retinopathy	Yes vs. no	0.469 (0.086–2.547)	0.38
Nephropathy	Yes vs. no	1.149 (0.409–3.226)	0.792
Neuropathy	Yes vs. no	0.503 (0.130–1.951)	0.32
IHD (coronary insufficiency)	Yes vs. no	0.721 (0.146–3.552)	0.688
Coronary artery angioplasty	Yes vs. no	4.480 (0.469–42.792)	0.193
Coronary artery bypass surgery	Yes vs. no	0 (0–infinity)	0.991
Congestive heart failure	Yes vs. no	0 (0–infinity)	0.99
Previous revascularization surgery (including PTA)	Yes vs. no	0 (0–infinity)	0.996
Dyslipidemia	Yes vs. no	0 (0–infinity)	1
Dialysis	Yes vs. no	2.018 (0.620–6.569)	0.244
Creatinine		1.064 (0.867–1.305)	0.553
GFR		1.005 (0.990–1.019)	0.533
Hb1Ac		1.097 (0.862–1.395)	0.451
WBC		1.198 (1.041–1.378)	0.011[a]
Osteomyelitis	Yes vs. no	0.746 (0.257–2.161)	0.589

PEDIS, perfusion, extent, depth, infection and sensation; IDSA, Infectious Diseases Society of America; OR, odds ratio; CI, confidence interval; SFA, superior femoral artery; PTA, posterior tibial artery; DM, diabetes mellitus; IHD, ischemic heart disease; GFR, glomerular filtration rate; HbA1c, glycosylated hemoglobin; WBC, white blood cell.
[a]$P < 0.05$.

Table 7. Multivariate binary logistic regression analysis using PE-DIS/IDSA grade (3, 4 vs. 1, 2)

Predictor	OR (95% CI)	P-value
Age	1.061 (1.004–1.122)	0.037[a]
Dialysis (yes vs. no)	21.370 (1.961–232.917)	0.012[a]
GFR	1.031 (1.001–1.062)	0.042[a]
WBC	1.245 (1.054–1.470)	0.01[b]
Osteomyelitis (yes vs. no)	2.397 (0.582–9.867)	0.226

PEDIS, perfusion, extent, depth, infection and sensation; IDAS, Infectious Diseases Society of America; OR, odds ratio; CI, confidence interval; GFR, glomerular filtration rate; WBC, white blood cell.
[a]$P<0.05$; [b]$P<0.01$.

found that coexistence of CKD is associated with a greater severity of PAD [16-20]. In line with previous studies, our study has shown that decreased GFR was a significant factor predicting the presence of PAD in diabetic foot ulcers patients.

The measurement of the long-term blood glucose control index, HbA1c, is used to evaluate and plan the treatment of diabetic patients. Chronic hyperglycemia is a well-known risk factor for diabetes-related vascular complications. Several studies have suggested that HbA1c may also be related with the incidence of large-vessel disease in patients with diabetes [21-24]. Our study was also consistent with the results of previous studies. In our study, HbA1c was a significant factor pre-

Table 8. Univariate binary logistic regression analysis using 10-item DFI score (cutoff=20)

Predictor	Subgroup	OR (95% CI)	P-value
Sex	Male vs. female	0.883 (0.247–3.154)	0.848
Age		1.025 (0.984–1.069)	0.24
Vascular clogging	SFA vs. no	0 (0–infinity)	0.993
	PTA vs. no	0 (0–infinity)	0.997
	SFA or PTA vs. no	0 (0–infinity)	0.992
	Multifocal artery vs. no	0.687 (0.197–2.404)	0.557
	Yes vs. no	0.458 (0.138–1.521)	0.202
Culture	Others vs. Gram-positive aerobic	0.528 (0.176–1.584)	0.255
	Others vs. Gram-negative aerobic	1.381 (0.463–4.119)	0.562
DM duration		0.984 (0.912–1.060)	0.666
Ulcer (history)	Yes vs. no	0.643 (0.036–11.632)	0.765
Antibiotic treatment (history)	Yes vs. no	0.692 (0.038–12.572)	0.804
Smoking	Yes vs. no	1.494 (0.505–4.415)	0.468
Hypertension	Yes vs. no	0.909 (0.307–2.691)	0.863
Retinopathy	Yes vs. no	0.400 (0.069–2.309)	0.306
Nephropathy	Yes vs. no	1.070 (0.366–3.128)	0.902
Neuropathy	Yes vs. no	0.604 (0.142–2.579)	0.496
IHD (coronary insufficiency)	Yes vs. no	1.375 (0.277–6.833)	0.697
Coronary artery angioplasty	Yes vs. no	1.193 (0.183–7.781)	0.854
Coronary artery bypass surgery	Yes vs. no	0 (0–infinity)	0.992
Congestive heart failure	Yes vs. no	0 (0–infinity)	0.991
Dyslipidemia	Yes vs. no	0 (0–infinity)	1
Dialysis	Yes vs. no	1.556 (0.479–5.051)	0.462
Creatinine		1.027 (0.833–1.265)	0.805
GFR		1.000 (0.985–1.015)	0.992
Hb1Ac		1.147 (0.895–1.469)	0.278
WBC		1.097 (0.984–1.223)	0.095
Osteomyelitis	Yes vs. no	3.600 (1.165–11.127)	0.026[a]

DFI, diabetic foot infection; OR, odds ratio; CI, confidence interval; SFA, superior femoral artery; PTA, posterior tibial artery; DM, diabetes mellitus; IHD, ischemic heart disease; GFR, glomerular filtration rate; HbA1c, glycosylated hemoglobin; WBC, white blood cell.
[a]$P<0.05$.

Table 9. Multivariate binary logistic regression analysis using 10-item DFI score (cutoff = 20)

Predictor	OR (95% CI)	P-value
Age	1.067 (1.002–1.136)	0.044[a]
Dialysis (yes vs. no)	18.288 (1.323–252.735)	0.03[a]
GFR	1.030 (0.994–1.068)	0.106
WBC	1.203 (1.033–1.401)	0.017[a]
Osteomyelitis (yes vs. no)	18.093 (3.150–113.750)	0.001[b]

DFI, diabetic foot infection; OR, odds ratio; CI, confidence interval; GFR, glomerular filtration rate; WBC, white blood cell.
[a]$P < 0.05$; [b]$P < 0.01$.

dicting the presence of PAD in diabetic foot ulcers patients.

Wound classification systems help physicians to evaluate the prognosis of diabetic foot patients or to plan treatment plans. In our study, older age, dialysis patients, decreased GFR, high WBC, and concomitance of osteomyelitis were significant factors for determining the severity of foot disease (Fig. 2).

As mentioned earlier, elderly patients may have various complications, including PAD, which can have a negative impact on wound status. Decreased GFR or the need of dialysis may indicate severe progression of diabetes in patients, contributing to worsened wound status. Diabetic foot ulcers pa-

Table 10. Univariate binary logistic regression analysis using bacterial culture (Gram-aerobic vs. others)

Predictor	Subgroup	OR (95% CI)	P-value
Sex	Male vs. female	3.188 (0.773–13.144)	0.109
Age		0.995 (0.957–1.034)	0.795
PEDIS/IDSA grade (1–4)	2 vs. 1	1.400 (0.300–6.535)	0.669
	3 vs. 1	1.517 (0.361–6.373)	0.57
	4 vs. 1	0 (0–infinity)	0.992
	3 or 4 vs. 1 or 2	1.151 (0.407–3.257)	0.791
Wound parameters (10-item)		1.051 (0.934–1.183)	0.407
DFI wound score (10-item)		1.023 (0.954–1.097)	0.527
10-Item DFI score (cutoff = 20)	≥20 vs. <20	0.724 (0.243–2.159)	0.562
Wound angiosome (ATA, PTA, multi)	PTA vs. ATA	0.750 (0.169–3.327)	0.705
Wound angiosome (ATA, PTA, multi)	Multi vs. ATA	0.463 (0.112–1.908)	0.287
Wound angiosome	Multi vs. single	0.556 (0.194–1.593)	0.274
Vascular clogging	SFA vs. no	0 (0–infinity)	0.993
	PTA vs. no	0 (0–infinity)	0.996
	SFA or PTA vs. no	0 (0–infinity)	0.992
	Multifocal artery vs. no	1.412 (0.426–4.683)	0.573
	Yes vs. no	0.824 (0.274–2.473)	0.729
DM duration (yr)		1.021 (0.950–1.097)	0.57
Smoking	Yes vs. no	0.500 (0.169–1.476)	0.21
Hypertension	Yes vs. no	1.105 (0.384–3.180)	0.853
Retinopathy	Yes vs. no	6.000 (0.631–57.058)	0.119
Nephropathy	Yes vs. no	1.470 (0.517–4.181)	0.47
Neuropathy	Yes vs. no	1.125 (0.300–4.212)	0.861
IHD (coronary insufficiency)	Yes vs. no	0.989 (0.200–4.878)	0.989
Coronary artery angioplasty	Yes vs. no	0.302 (0.032–2.887)	0.299
Congestive heart failure	Yes vs. no	1.905 (0.386–9.411)	0.429
Dyslipidemia	Yes vs. no	1.000 (0–infinity)	1
Dialysis	Yes vs. no	3.000 (0.910–9.889)	0.071

(Continued to the next page)

Table 10. Continued

Predictor	Subgroup	OR (95% CI)	P-value
CKD stage	DDKT vs. not related	0 (0–infinity)	0.997
	Stage 2 vs. not related	0 (0–infinity)	0.997
	Stage 3 vs. not related	0.792 (0.125–5.009)	0.804
	Stage 3–4 vs. not related	0 (0–infinity)	0.997
	Stage 4–5 vs. not related	0 (0–infinity)	0.997
	Stage 5 vs. not related	2.639 (0.761–9.153)	0.126
Creatinine		1.163 (0.942–1.436)	0.161
GFR		0.995 (0.981–1.010)	0.526
Hb1Ac		0.695 (0.510–0.947)	0.021[a]
WBC		1.065 (0.958–1.184)	0.24
Osteomyelitis	Yes vs. no	0.336 (0.107–1.053)	0.061

OR, odds ratio; CI, confidence interval; PEDIS, perfusion, extent, depth, infection and sensation; IDSA, Infectious Diseases Society of America; DFI, diabetic foot infection; ATA, anterior tibial artery; PTA, posterior tibial artery; SFA, superior femoral artery; DM, diabetes mellitus; IHD, ischemic heart disease; CKD, chronic kidney disease; DDKT, death donor kidney transplantation; GFR, glomerular filtration rate; HbA1c, glycosylated hemoglobin; WBC, white blood cell.
[a]P<0.05.

Table 11. Multivariate binary logistic regression analysis using bacterial culture (Gram-aerobic vs. others)

Predictor	OR (95% CI)	P-value
Sex (male vs. female)	13.680 (1.294–144.622)	0.03[a]
Smoking (yes vs. no)	0.071 (0.011–0.464)	0.006[b]
Dialysis (yes vs. no)	3.868 (0.644–23.234)	0.139
Hb1Ac	0.525 (0.320–0.862)	0.011[a]
WBC	1.271 (1.062–1.522)	0.009[b]

OR, odds ratio; CI, confidence interval; HbA1c, glycosylated hemoglobi; WBC, white blood cell.
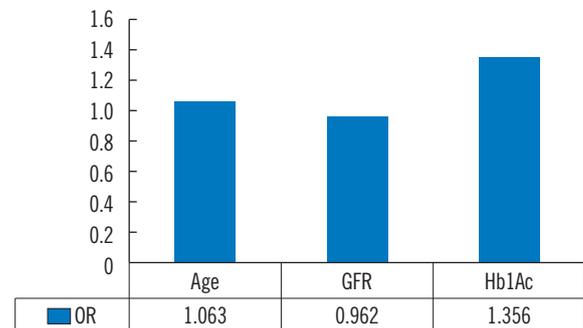[a]P<0.05; [b]P<0.01.

Fig. 1. Vascular clogging (yes vs. no) in DFU. Multivariate binary logistic regressing. DFU, diabetic foot ulcers; GFR, glomerular filtration rate; HbA1c, glycosylated hemoglobin; OR, odds ratio.
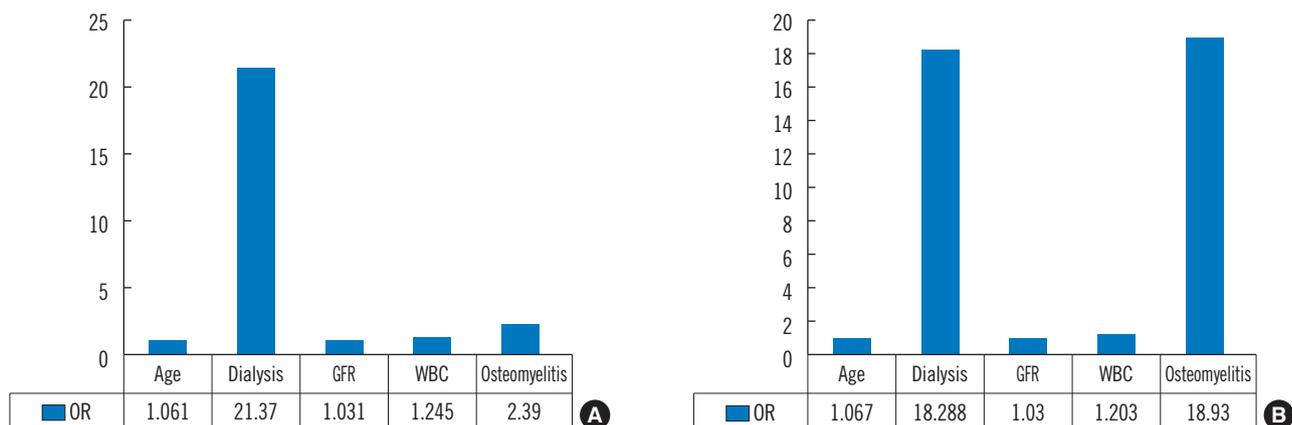

Fig. 2. Multivariate binary logistic regressing. (A) Significant factors for PEDIS/IDAS grade (3, 4 vs. 1, 2) in diabetic foot ulcers (DFU). (B) Significant factors for 10-item DFI score (cutoff=20) in DFU. PEDIS, perfusion, extent, depth, infection and sensation; IDAS, Infectious Diseases Society of America; DFI, diabetic foot infection; GFR, glomerular filtration rate; WBC, white blood cell; OR, odds ratio.
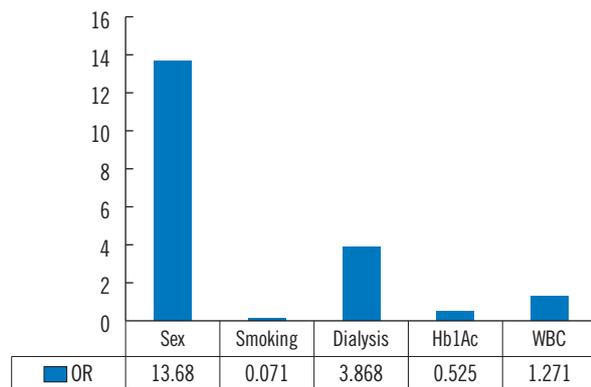
Fig. 3. Bacterial culture (Gram-aerobic vs. others) in DFU. Multivariate binary logistic regressing. DFU, diabetic foot ulcers; HbA1c, glycosylated hemoglobin; WBC, white blood cell; OR, odds ratio.

tients with high WBC and osteomyelitis might already have an infection, which is likely to lead to greater severity of disease.

To the best of our knowledge, there have been no studies analyzing the factors that influence bacterial culture test results, which are the basis of antibiotic therapy in diabetic foot ulcers patients. In our study, sex (male), dialysis, and high WBC seem to affect the possibility of Gram-negative aerobic bacterial infection more than other bacterial infections (Fig. 3).

Polymicrobial organisms have been isolated from infected diabetic foot ulcer patients since the 1980s. Gram-positive cocci bacteria are the most commonly isolated microorganisms, Gram-negative being the second, and anaerobic bacteria being the third. These results do not reflect the various clinical features and can vary from region to region, which may affect the treatment strategy.

Despite the previous results, according to the literature from Asia and the Middle East, the most predominant bacteria isolated from diabetic foot ulcers patients are said to be Gram-negative organisms [25]. Similar to previous studies, our study has shown that Gram-negative aerobic bacteria was the most predominant isolated organism. The possibility of a variance in results of bacterial culture tests in diabetic foot ulcers patients across different regions and races must be noted, and custom treatment plans for each region and/or race should be considered.

Our study had some limitations. First, this study was based on a retrospective chart review. Second, the iagnosis of osteomyelitis was made only in a limited number of patients, using three-phase bone scan. Though three-phase bone scans are less sensitive and specific than magnetic resonance imaging, they were used for rapid screening at an appropriate point in time.

In conclusion, the present study has confirmed significant factors affecting vascular status, wound status, and bacterial culture tests, including co-morbidities, as well as foot and ulcer characteristics at baseline. Older age and decreased GFR have affected both the vascular status and wound status. In addition, sex (male), dialysis, and high WBC seem to affect the possibility of Gram-negative aerobic bacterial infection more than other bacterial infections. However, recent studies have shown that empirical treatments may vary from region to region and that further research is needed, as the predominant bacteria tends to differ accross different ethnic groups and regions.

Conflict of interest

No potential conflicts of interest relevant to this article are reported.

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