

Risk Factors of Methicillin-Resistant *Staphylococcus Aureus* and *Pseudomonas* Infection in Diabetic Foot Ulcers in Korea

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Abstract

Background: In diabetic foot infection (DFI), risk of methicillin-resistant *Staphylococcus aureus* (MRSA) and *Pseudomonas* infection should be considered in selecting empirical antibiotics. The aim of this study is to analyze the risk factors of MRSA and *Pseudomonas* infection with respect to DFI in Korea to help to choose adequate empirical antibiotics.

Methods: This study included 737 patients with diabetic foot ulcers, who were admitted to diabetic wound center of the Korea University Guro Hospital between January 2012 and December 2016. Deep tissue or bone biopsy cultures were taken during surgical debridement in the operating room. Using eight categories of patients' clinical characteristics (age, gender, smoking history, dialysis, HbA1c, wound duration, TcPO₂, previous use of antibiotics), we evaluated the risk factors for MRSA and *Pseudomonas aeruginosa* infection.

Results: Among the 832 microbial isolates, 114 were MRSA (13.7%) and 78 were *Pseudomonas* (9.4%), respectively. Wound duration (>4 weeks) was the only statistically significant factor associated with the MRSA infection in univariate and multivariate logistic analysis. For *Pseudomonas aeruginosa* infection, three factors – gender, smoking history, and HbA1c level (<7.0%) – were found to be statistically significant in univariate analysis. In multivariate analysis, smoking history and previous treatment history emerged as strong predictors for the *Pseudomonas aeruginosa* infection.

Conclusion: Wound duration can be the risk factor of MRSA infection in diabetic foot ulcers. Regarding *Pseudomonas* infection, smoking history and previous treatment history can be risk factors.

Keywords: Diabetic foot, Methicillin-resistant staphylococcus aureus, Pseudomonas, Risk factors

Introduction

In the treatment of patients with DFI, major treatment plans are followings; surgical debridement and/or antibiotic use. According to current clinical guideline from Infectious Disease Society of America (IDSA), if there is an infection, surgical debridement should be performed to excise the infected tissue with deep tissue culture obtained. Empirical antibiotics should also be administered until wound cultures define the causative organisms and their antibiotic susceptibility patterns, allowing targeted antibiotics regimens [1,2]. 3–5 days waiting period for results is the critical period for initial infection management, during which most of the clinicians use a broad-spectrum empirical antimicrobial therapy, which covers most of the causative pathogens. In particular, risk factors of MRSA and *Pseudomonas* infection should be considered for choice of empirical antibiotics [1,2]. Henceforth, it is imperative that primary surgeons know the risk factors of MRSA and *Pseudomonas* infection in diabetic foot ulcer specific to their own country.

Several studies have attempted to identify risk factors of MRSA infection in western population (North American and European). Previous use of antibiotics, previ-



Original Article

Received: July 31, 2017
 Revised: September 25, 2017
 Accepted: September 26, 2017

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No potential conflict of interest relevant to this article was reported.

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ous hospitalization, and extended duration of the wound in diabetic foot ulcer were noted as risk factors for MRSA infection [3-6]. However, it is difficult to apply the results from the western population to Korean population due to intrinsic limitations such as the difference of prevalence of causative pathogens and characteristics of patients. At the same time, to the best of our knowledge, study on the risk factors of *Pseudomonas* infection in diabetic foot ulcer with large sample is very limited. To bridge the gaps, this study aims to find risk factors for MRSA and *Pseudomonas* infection in diabetic foot ulcer using the data of Korean population.

Methods

Materials

Medical records of 737 patients with diabetic foot ulcers, who were admitted and treated at the diabetic wound center of Korea University Guro Hospital between January 2012 and December 2016, were reviewed. A complete medical history was obtained at the first visit. Based on previous studies, eight risk factors were reviewed for each patient: age, gender, smoking history, dialysis, HbA1c value, wound duration, value of transcutaneous partial oxygen tension (TcPO₂) and previous use of antibiotics (Table 1). Furthermore, the aforementioned factors were included in the comparative study for

Table 1. Patient demographics (n=737)

Variable	Total (n=737)
Age (year)	64 ± 12.1
Gender	
Male	530 (71.9%)
Female	207 (28.1%)
Smoking history	
Yes	224 (30.3%)
No	513 (69.7%)
Dialysis	
Yes	167 (22.6%)
No	570 (77.4%)
HbA1C (%)	7.8 ± 1.6
Wound duration (week)	23.2 ± 6.3
TcPO ₂ (mmHg)	30.8 ± 23.7
Previous use of antibiotics	
Yes	556 (75.4%)
No	181 (24.6%)

TcpO₂, Transcutaneous partial oxygen tension.

determining infection risk factor of MRSA and *Pseudomonas aeruginosa*. This study protocol was approved by the Institutional Review Board of Korea University Guro Hospital.

Microbiological Methods

Only intraoperative samples collected from surgical procedures were included in the study. Surgical debridement was performed to remove all non-viable tissue from the wounds. Deep tissue and/or bone specimens were obtained and immediately stored in aseptic tube for culture. The specimens were sent to microbiologic laboratory in the same hospital and incubated at 35°C for 24–48 hours in Sheep blood agar plate. Furthermore, MacConkey II agar plate was used for aerobes and Chocolate agar plate for anaerobes.

Statistical analysis

Qualitative variables were expressed as percentages, and quantitative variables were expressed as means ± standard deviation (SD). To assess the association between the potential risk factors and infection of MRSA and *Pseudomonas aeruginosa*, univariate analysis was performed (SPSS 12.0 version, SPSS Inc.). Screening for potential risk factors was done using Chi-square test and logistic regression analysis. The P-values <0.20 were considered significant for inclusion in the multivariate analysis. Multivariate linear logistic regression models were formulated and tested to adjust for covariates. Adjusted odds ratios (OR) and 95% confidence intervals (CI) were calculated, and P-values <0.05 were considered as statistically significant.

Results

Out of the 737 patients included in this study, 623 patients (84.5%) had isolated causative organisms and 832 microbial isolates were identified. Among 832 isolates, 114 organisms (13.7%) were MRSA as the most common isolated organisms. *Pseudomonas aeruginosa* was identified by 78 organisms (9.4%).

Analysis of eight potential risk factors for MRSA and *Pseudomonas aeruginosa* infection is shown in Table 2. As per univariate analysis, wound duration (>4 weeks) was the only statistically significant factor associated with the MRSA infection (P=0.009).

In case of *Pseudomonas aeruginosa* infection, three factors including gender, smoking history, and HbA1c level (<7.0%) were identified as statistically significant factors. Male patients

Table 2. Analysis of risk factors for MRSA and *Pseudomonas* infection

Risk factor	MRSA infection			<i>Pseudomonas</i> infection		
	Yes	No	P-value	Yes	No	P-value
Age			0.702			0.717
≥ 65	50 (14.7%)	289 (85.3%)		37 (10.9%)	302 (89.1%)	
< 65	64 (15.8%)	342 (84.2%)		41 (10.1%)	365 (89.9%)	
Gender			0.710			0.005
Male	81 (15.0%)	459 (85.0%)		67 (12.4%)	473 (87.6%)	
Female	33 (16.1%)	172 (83.9%)		11 (5.4%)	194 (94.6%)	
Smoking history			0.160			0.008
Yes	31 (12.7%)	214 (87.3%)		36 (14.7%)	209 (85.3%)	
No	83 (16.6%)	417 (83.4%)		42 (8.4%)	458 (91.6%)	
Dialysis			0.971			0.071
Yes	27 (15.3%)	149 (84.7%)		25 (14.2%)	151 (85.8%)	
No	87 (15.5%)	476 (84.5%)		53 (9.4%)	510 (90.6%)	
HbA1c			0.189			0.005
≥ 7.0% (Uncontrolled DM)	66 (14.0%)	406 (86.0%)		38 (8.1%)	434 (91.9%)	
< 7.0% (Controlled DM)	48 (17.6%)	225 (82.4%)		40 (14.7%)	233 (85.3%)	
Wound duration			0.009			0.340
≥ 4 weeks	94 (17.4%)	445 (82.6%)		60 (11.1%)	479 (88.9%)	
< 4 weeks	20 (9.7%)	186 (90.3%)		18 (8.7%)	188 (91.3%)	
TcPO ₂			0.844			0.865
≥ 40 mmHg	31 (15.7%)	166 (84.3%)		20 (10.2%)	177 (89.8%)	
< 40 mmHg	83 (15.1%)	465 (84.9%)		58 (10.6%)	490 (89.4%)	
Previous use of antibiotics			0.699			0.060
Yes	80 (15.0%)	454 (85.0%)		63 (11.8%)	471 (88.2%)	
No	34 (16.1%)	177 (83.9%)		15 (7.1%)	196 (92.9%)	

TcpO₂, Transcutaneous partial oxygen tension.

had a 2.5-fold increased odds for *Pseudomonas aeruginosa* infection and 1.9-fold for patients with smoking history and 1.8-fold for patients with controlled DM (HbA1c <7.0%) respectively.

Smoking history, HbA1c level, and wound duration were further evaluated in a multiple logistic regression analysis for MRSA infection. In the same way, gender, smoking history, dialysis, HbA1c level, and previous use of antibiotics were evaluated for *Pseudomonas aeruginosa* infection (Table 3). Only wound duration (>4 weeks) was a significant predictor for the MRSA infection (OR, 2.21; P=0.001). Smoking history and previous treatment emerged as strong predictors for the *Pseudomonas aeruginosa* infection, whereas gender and HbA1c were no longer found as predictive factors. Patients with a smoking history had a 1.7-fold increased odds for

Table 3. Multivariate logistic analysis for the MRSA and *Pseudomonas* infection

Risk factor	OR (95% CI)	P-value
MRSA infection		
Wound duration (≥ 4 weeks)	2.21 (1.39-3.52)	0.001
HbA1c (≥ 7.0%, Uncontrolled DM)	0.68 (0.47-0.98)	0.061
<i>Pseudomonas</i> infection		
Smoking history	1.74 (1.03-2.93)	0.037
Treatment history	1.94 (1.06-3.54)	0.030

Pseudomonas aeruginosa infection. Similarly, patients previously treated in other hospital had 1.9-fold increased odds for infection.

Discussion

The prevalence of diabetes worldwide was estimated to be 2.8% in 2000 and is expected to increase to 4.4% in 2030 [2,7]. Epidemiological studies suggest that 2.5% of diabetic patients develop diabetic foot ulcers each year and 15–25% during their lifetime [2,8]. Diabetic foot ulcer can be caused by various risk factors such as peripheral neuropathy, peripheral vascular disease, trauma, and impaired resistant to infection [2,9,10]. DFI is the most frequent and disastrous complication of the diabetic foot [2,11]. It is considered as a serious health and socioeconomic problem than any other diabetes-related complications and is the major cause of non-traumatic lower extremity amputations [2,12].

Staphylococcus aureus is one of the most common pathogens in DFI, as much as 46% of which is MRSA [1]. The prevalence of MRSA in DFI continues to rise, which ranges from 5% to 30% [13,14]. MRSA in DFI is associated with slower healing, more osteomyelitis, and more surgical procedures [15,16]. Maybe for these reasons, it is commonly believed to be an important cause of poor outcomes, increased healthcare cost, and mortality [3,13,17,18]. *Pseudomonas aeruginosa* is associated with immune-compromised hosts and is relatively uncommon as a cause of DFI in non-tropical, developed countries [19,20]. However, it causes severe tissue damage with its tendency to resist phagocytosis and antibiotics [21]. Furthermore, it can cause sepsis or amputation, so it should always be considered as a possible pathogen [22]. Accordingly, prevention of MRSA and/or *Pseudomonas* in DFI should be warranted.

This large, single-center study examined the risk factors of MRSA and *Pseudomonas* infection in diabetic foot ulcer patients. In this study, only deep tissue or bone culture obtained in operating room during surgical debridement were employed, thus ensuring reliable results on true pathogen of DFI.

Previous studies have noted the following risk factors for MRSA DFI: presence of multidrug-resistant organisms, history of MRSA DFI, previous use of antibiotics, previous hospitalization, and extended duration of the foot wound [3-6,23]. Other studies done in the United States found that males were at a higher risk of developing MRSA infection [14]. Kallen et al found out that patients undergoing chronic dialysis showed the higher incidence of invasive MRSA than the general population. Ding et al showed that HbA1c was a risk factor for the methicillin-resistant *Staphylococci* infection. In our study, univariate analysis revealed that wound duration (>4

weeks) was the only statistically significant factor associated with the MRSA infection ($P=0.009$). Interestingly, previous use of antibiotics, which is a well-known factor of MRSA infection, was not found as a statistically significant factor ($P=0.699$). Multiple logistic regression analysis also revealed wound duration (>4 weeks) as a strong predictor for MRSA infection ($P=0.001$). Recently, in Korea, not only DFI, but also other MRSA infection have been prevailing due to community-acquired MRSA infection [24,25]. It is probable that health care-associated factors, such as previous use of antibiotics, are less likely the cause of MRSA infection than in the past.

In case of *Pseudomonas* infection, there exist many studies on its microbiological epidemics and treatment, but there has been no large study that revealed the risk factors for *Pseudomonas* infection in diabetic foot ulcer. In our study, smoking history and previous use of antibiotics were identified as strong predictors for the *Pseudomonas aeruginosa* infection. Similarly, in respiratory disease, cigarette smoking has been verified as a risk factor for infection by the opportunistic pathogens like *Pseudomonas aeruginosa* [26,27]. The underlying mechanism and causes of *Pseudomonas aeruginosa* infection in diabetic foot ulcers need to be evaluated in future study.

Our previous research has shown that the various phenomena related to diabetic foot ulcers in Korea are different from those in the western. For example, microbiology in DFI was different. The incidence of Gram-negative organisms in Korean population was higher than in western population, among which *Pseudomonas* was the most common and important organism [2]. Other prior study that our center conducted regarding the risk factors of major amputations in diabetic foot ulcer patients in Korean population also showed the difference. For instance, foot infection was an important risk factor in other countries, but not in Korea. We postulated that the difference was due to the different treatment protocols [28]. Moreover, even in the same geographic area, there was difference by race. A study conducted in the UK examined the difference in the prevalence of diabetic foot ulcers between the Asian and Caucasian population with possible contributing factors. According to the study, prevalence of the ulcers among the Asian population was about three times lower than that of the European population, which is attributed to lower peripheral vascular disease, neuropathy, and foot deformity [29]. It is likely that genetic factors contribute to the racial difference. Future research considering the geographi-

cal and racial difference should be conducted, and ultimately it is necessary to establish a specific treatment protocol that is appropriate for Korean population.

Conclusion

In our single-center based, large population study, wound duration (>4 weeks) was identified as the only risk factor for MRSA infection in DFI. In the case of *Pseudomonas aeruginosa* infection, patient's history of smoking and previous use of antibiotics were identified as the risk factors by multivariate logistic regression analysis. Our study provides an evidence for the need of empirical antibiotic therapy regimens for DFI specific to local clinical practices. We suggest the need for our own guidelines based on microbiology of Korean population for designing an effective empirical antibiotic treatment.

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