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ORIGINAL ARTICLE

Predictors of healing, ulcer recurrence and persistence, amputation and mortality in type 2 diabetic patients with diabetic foot: a 10-year retrospective cohort study

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Abstract

Purpose Predictors of outcome of diabetic foot ulcer (DFU) are important to improve the management of patients. Aim of the study was to find these predictors in type 2 diabetic patients with DFU.

Methods We recruited 583 patients. They were followed-up by a multidisciplinary team. A holistic and conservative approach was used and all risk factors and co-morbidities were aggressively treated.

Results During the follow-up period, 79.6% of patients healed in a mean time of 7.6 ± 3.8 months, 6.9% showed DFU persistence, 9.9% had minor amputations, and 3.6% experienced major amputation. Seventeen percent of the patients died. Among patients who healed, 37.1% of them showed DFU recurrence. Impairment of renal function was associated to DFU persistence, amputation, and mortality. Previous cardiovascular disease predicted DFU persistence, DFU recurrence, and mortality. Lower BMI predicted DFU persistence and mortality. Osteomyelitis was a predictor of amputation and death. Markers of peripheral artery disease (PAD) predicted minor amputation and DFU recurrence. Our study shows a relatively low incidence of complications of DFU.

Conclusions Some predictors of outcome of DFU were confirmed and new predictors, like BMI and markers of PAD, were found. Our new findings suggest future strategies for nutrition support and revascularization. In addition, a holistic and conservative approach may improve the prognosis.

Keywords Diabetic foot · Type 2 diabetes · Outcome · Diabetic ulcer · Prognosis

Introduction

Diabetic foot ulcer (DFU) is a common complication of diabetes, as its global prevalence is about 6.3% [1] and the lifetime risk for a diabetic patient to develop DFU is 25% [2]. DFUs are associated with a very high nontraumatic lower extremity amputation rate: indeed, diabetic patients have a risk for nontraumatic lower extremity amputation 15- to 40-fold higher than the general population [3] and 85% of all amputations in people with diabetes is preceded by a DFU [2]. Amputations are associated with a 5-year mortality until 80% which may be even worse than that observed in several types of cancer [4, 5]. DFUs cause disability and have a negative impact on the quality of life of patients and their families not only because of amputations but also because of prolonged tissue healing (median time is 32 weeks) [5], the high percentage of DFUs that remain active (until 15%) [4], and the high percentage of recurrence (until 65% in 5 years) [4]. All these clinical and social consequences of DFUs have very high financial costs [2].

To improve outcomes of DFU, such as healing, recurrence, persistence, amputations, and mortality, all their predictors should be in-dept known with the aim to update guidelines and recommendations for the management of diabetic foot. Several studies have evaluated the predictors of outcomes in patients with diabetic foot, even if lot of

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these studies have taken into account small populations, a single or a limited number of predictors and outcomes and the time of follow-up was often quite short.

Aim of the present study was to retrospectively evaluate the impact of several predictors of wound healing, DFU recurrence, DFU persistence, amputations, and mortality in a large group of type 2 diabetic patients with DFU during a 10-year period.

Methods

We undertook a retrospective cohort study of 583 consecutive type 2 diabetic patients who attended the outpatient department of the Diabetic Foot Unit of the Clinical Institute Beato Matteo, Vigevano, Italy, for a recent (occurred no more than 30 days before the visit) and single DFU from March 2009 to June 2019. They were followed-up until February 2020. The inclusion criteria were type 2 diabetes, age ≥45 years, at least 1-year follow-up or less when amputation or death occurred before this period. However, the follow-up continues for the patients with new amputation and only the outcome death is recorded in these patients. Exclusion criteria were reduced life expectative, missing data, patients lost at follow-up, multiple ulcers, nondiabetic ulcers, such as venous ulcers, any neurological disease other than stroke, chronic treatment with immunosuppressants or steroids. The informed consent was obtained by all the subjects enrolled in the study. The study was carried out according to the ethical standards of the institutional research committee and with the 1964 Helsinki declaration. The local Institutional Review Board has approved the protocol.

All the patients were treated and followed-up by a multidisciplinary team made up of diabetologists, expert nurses trained in advanced wound dressing, internists, and dietitians. The patients were referred to other consultants, such as surgeons, vascular surgeons, orthopedic surgeons, interventional radiologists, rehabilitation physicians, nephrologists, cardiologists, and neurologists, when needed.

The patients were managed according to established guidelines, recommendations, and protocols, in particular the 2019 guidance of the International Working Group on the Diabetic Foot [6] and previous versions, the Italian Standards for the care of Diabetes, and any specific guidelines regarding any procedure, such as revascularization or treatment of osteomyelitis. A holistic and conservative approach was always used. This means that all the patients were treated not only for diabetes and its complications but also for any type of co-morbidities. In addition, any type of nonsurgical treatment was always used before using surgical approaches. Standard outpatient care included cleaning and dressing, weekly debridement, early treatment of infection, use of specific technologies, such as negative pressure therapy, pressure offloading, indication for specific footwear, early diagnosis of neuropathy, peripheral artery disease (PAD) and osteomyelitis and their immediate treatment, strict metabolic control, nutritional support, diagnosis and treatment of any co-morbidities and risk factors, indication for surgery when needed, early rehabilitation.

After the ulcer healed, the patients returned every 1–3 months to the Diabetic Foot Unit for periodic examinations. In addition, they were followed-up by periodic telephone interviews.

Data were collected from electronic medical records in the hospital and filled in a specific structured data collection form. The following biological and clinical data obtained at the first examination were collected: age, gender, diabetes duration, body mass index (BMI), smoking habits, history of established cardiovascular disease (CVD), history of previous healed ulcer, history of previous lower extremity amputation, presence of hypertension, HbA1c, total cholesterol, HDL, triglycerides, LDL, estimated glomerular filtration rate (eGFR), micro- or macroalbuminuria, ankle brachial index (ABI), transcutaneous oxygen tension (TcPO2), presence of distal polyneuropathy, presence of PAD, presence of osteomyelitis, and revascularization procedure. BMI, smoking habits, history of established CVD, hypertension, HbA1c, total cholesterol, HDL, triglycerides, micro- and macroalbuminuria, ABI, TcPO2, and distal polyneuropathy were determined or defined as previously described [7–11]. LDL was calculated with the Friedewald's formula [12]. eGFR was estimated according to the Cockcroft–Gault formula [13]. PAD was defined as the presence of an ABI <0.9 and/or aTcPO2 <46 mmHg [11]. However, all the patients underwent an echocolordoppler to evaluate the presence of artery stenoses. In some patients PAD was confirmed by positioning the sensor for TcPO2 near and above the lesion, when TcPO2 was normal at the dorsum of the foot, but the suspect of an ischemic ulcer was strong. Therefore, DFU was defined as neuropathic DFU (NDFU), vascular DFU (VDFU), or neurovascular DFU (NVDFU) [8]. The diagnosis of osteomyelitis was determined on a clinical basis and always confirmed by imaging (X-ray and/or magnetic resonance imaging). Osteomyelitis was always managed with antibiotics and local infected debridement. Only osteomyelitis persistently nonresponsive to medical therapy were treated with local minor amputation. In patients with noninvasive diagnosis of PAD, an ABI <0.8 and/or a TcPO2 <40 mmHg were considered thresholds for possible revascularization. The decision for revascularization was reached by the multidisciplinary team together with the vascular surgeon and the interventional radiologist. In addition to ABI and TcPO2, several clinical features were taken into account,

such as a rapid, slow or absent wound healing, the clinical conditions of the patient and so on. Therefore, the procedure was also exploited in some patients with ABI > 0.8 e and TcPO2 > 40 mmHg and not exploited in subjects with parameters below the thresholds. Candidates for revascularization underwent contrast angiography and percutaneous transluminal angioplasty. In three patients, interventional radiologist gave indication for open surgery.

Primary outcomes were: (1) primary wound healing, when a complete healing was documented in two consecutive visits and no amputation occurred; (2) minor amputation, defined as an amputation below the calcaneus; (3) major amputation, defined as an amputation above the calcaneus; (4) persistence of active ulcer, defined as an ulcer not healed during the follow-up period. Additional outcomes were: (1) ulcer recurrence that was a new ulcer in patients with healed ulcer; (2) mortality in the whole study population.

Statistical analysis

To find differences among more than two groups ANOVA or Kruskal–Wallis test, when appropriate, was used. Univariate analysis was carried out by using Student *t* test to find significant differences between two groups in normally distributed parameters, while Mann–Whitney *U* test was performed in non-normally distributed variables. The Pearson Chi-squared test was used for frequency comparisons. Univariate analysis was performed to screen all the variables, and the significant variables were graded and tested as potential predictors of each outcome of DFU in a multiple logistic regression analysis with a stepwise approach. Variables were adjusted for co-variates. Goodness-of-fit was assessed on smaller random subsamples of the data using the Hosmer–Lemeshow chi square test. Variables were dichotomized before the analysis as previously reported [7, 9, 11, 14]. A p value < 0.05 was considered statistically significant.

Results

Table 1 depicts the biological and clinical features of the whole population of 583 type 2 diabetic patients with DFU at baseline and all the outcomes during the follow-up period (42.8 ± 23.3 months—range 3–115 months). Among the patients, only 11 of them had a follow-up <12 months: ten patients died and one had a major amputation, but he was followed-up for 23 months after amputation and death did not occur.

Table 2 shows the biological and clinical characteristics of the patients with DFU stratified by the four primary outcomes. Generic significant differences among the four groups were found in age, gender, BMI, diabetes duration, triglycerides, eGFR, micro/macroalbuminuria, previous CVD, amputation and DFU, presence of NDFU, VDFU and NVDFU, ABI, TcPO2, lower limb revascularization and occurrence of osteomyelitis.

Among the 583 patients, 464 of them healed in a mean time of 7.6 ± 3.8 months (range 1–25 months). Compared to subjects with healed ulcers, patients with persistence of active ulcers were significantly older, had a significantly lower BMI, eGFR, and percentage of people with NDFU; in addition, they showed a greater duration of diabetes and

Table 1 Features of the whole population of 583 type 2 diabetic patients with diabetic foot ulcer and outcomes of diabetic foot ulcer during the follow-up period

Variables	
Age (years)	71.1 ± 8.8
Males (%)	55.9
BMI	27.6 ± 4.1
Smokers (%)	32.6
Hypertension (%)	73.0
Diabetes duration (years)	14.4 ± 8.8
HbA1c (%)	7.4 ± 1.4
Cholesterol (mg/dl)	169.7 ± 37.5
HDL (mg/dl)	45.6 ± 11.9
Triglycerides (mg/dl)	118.5 ± 44.6
LDL (mg/dl)	100.4 ± 34.9
eGFR (mL/min)	63.7 ± 21.4
Micro- or macroalbuminuria (%)	46.1
Previous CVD (%)	33.8
Previous DFU (%)	27.1
Previous amputation (%)	8.7
Neuropathic ulcer (%)	41.7
Vascular ulcer (%)	20.0
Neurovascular ulcer (%)	36.7
Absence of neuropathy and PAD (%)	1.5
ABI	0.93 ± 0.27
TcPO2 (mmHg)	42.4 ± 15.2
Lower limb revascularization (%)	34.3
Osteomyelitis (%)	19.2
Primary outcomes	
Healing (%)	79.6
Persistence of active ulcers (%)	6.9
Minor amputations (%)	9.9
Major amputations (%)	3.6
Additional outcomes	
Ulcer recurrence (among healed ulcers) (%)	37.1
Death (%)	17.0

eGFR estimated glomerular filtration rate, *CVD* cardiovascular disease, *DFU* diabetic foot ulcer, *TcPO2* transcutaneous oxygen tension, *BMI* body mass index, *ABI* ankle brachial index, *PAD* peripheral artery disease, *DFU* diabetic foot ulcer

Table 2 Features of patients with diabetic foot ulcers stratified by the primary outcomes

Variables	Healing ($n = 464$)	Persistence active ulcer ($n = 40$)	Minor amputation ($n = 58$)	Major amputation ($n = 21$)
Age (years)	70.5 ± 8.4	77.0 ± 10.2$^{\$}$	70.2 ± 8.6	77.9 ± 7.3$^{\#}$
Males (%)	56.7	57.5	55.2	38.1
BMI	27.9 ± 3.9	24.8 ± 4.5$^{\$}$	28.1 ± 4.8	25.2 ± 3.2¶
Smokers (%)	32.5	25.0	37.9	33.3
Hypertension (%)	72.6	77.5	72.4	76.2
Diabetes duration (years)	13.5 ± 8.0	21.4 ± 12.6$^{\$}$	15.2 ± 9.1	20.7 ± 9.9$^{\#}$
HbA1c (%)	7.4 ± 1.3	7.1 ± 1.2	7.7 ± 1.6	7.4 ± 1.6
Cholesterol (mg/dl)	167.8 ± 36.2	178.7 ± 36.5	176.3 ± 44.8	175.7 ± 43.5
HDL (mg/dl)	45.6 ± 11.7	45.3 ± 11.5	44.8 ± 13.3	49.3 ± 13.4
Triglycerides (mg/dl)	116.9 ± 44.1	123.9 ± 48.4	127.6 ± 45.5	119.3 ± 44.0
LDL (mg/dl)	98.9 ± 33.8	108.6 ± 36.8	105.9 ± 38.6	102.5 ± 42.0
eGFR (mL/min)	66.8 ± 20.7	44.1 ± 18.9$^{\$}$	55.2 ± 17.7$^{\#}$	57.6 ± 23.5*
Micro- or macroalbuminuria (%)	40.9	57.5*	67.2$^{\#}$	80.9$^{\#}$
Previous CVD (%)	26.5	65.0$^{\$}$	60.3$^{\$}$	61.9$^{\#}$
Previous DFU (%)	20.5	32.5	67.2$^{\$}$	52.4$^{\#}$
Previous amputation (%)	4.7	10.0	24.1$^{\#}$	52.4$^{\$}$
Neuropathic ulcer (%)	47.2	22.5¶	22.4¶	9.5$^{\#}$
Vascular Ulcer (%)	19.0	32.5*	22.4	14.3
Neurovascular ulcer (%)	31.9	45.0	55.2	76.2$^{\$}$
Absence of neuropathy and PAD (%)	1.9	0	0	0
ABI	0.95 ± 0.27	0.98 ± 0.25	0.79 ± 0.29$^{\$}$	0.90 ± 0.34
TcPO2 (mmHg)	41.1 ± 15.1	40.0 ± 11.2	34.5 ± 14.8$^{\$}$	30.7 ± 13.4$^{\#}$
Lower Limb Revascularization (%)	31.9	25.0	53.4¶	52.4
Osteomyelitis (%)	13.4	17.5	44.8$^{\$}$	80.1$^{\$}$

Statistical significance versus healing: * < 0.05; ¶ < 0.01; # < 0.001; $ < 0.0001

eGFR estimated glomerular filtration rate, *CVD* cardiovascular disease, *DFU* diabetic foot ulcer, *TcPO2* transcutaneous oxygen tension, *BMI* body mass index, *ABI* ankle brachial index, *PAD* peripheral artery disease, *DFU* diabetic foot ulcer

percentage of subjects with previous CVD and VDFU. With respects patients with healed ulcers, subjects with a minor amputation had a significantly lower eGFR, ABI, and TcPO2 and percentage of patients with NDFU and a significantly higher percentage of people with micro/macroalbuminuria, previous CVD, previous DFU, previous amputation, revascularization, and osteomyelitis. At last patients who had a major amputation were older, showed a significantly lower BMI, eGFR, TcPO2 and percentage of people with NDFU and a significantly higher diabetes duration and percentage of patients with/micro/macroalbuminuria, previous CVD, previous DFU, previous amputation, NVDFU, and osteomyelitis than patients with healed ulcers.

Table 3 reports the features of patients with healed DFU subdivided into two groups according to the absence or presence of ulcer recurrence during the follow-up period. As shown, the group of patients with ulcer recurrence were older, had a significantly greater percentage of subjects with micro/macroalbuminuria, previous CVD, previous DFU, previous amputation, NDFU, VDFU, and NVDFU and showed significantly lower ABI and TcPO2 than the group of patients without ulcer recurrence. Interestingly, none of the patients without polyneuropathy and PAD had ulcer recurrence.

Table 4 shows the features of the patients who died compared to those who remained alive during the follow-up period.

Multivariate analysis

To find independent predictors for each outcome, multiple logistic regression analyses were performed. All the variables that were significant at the univariate analysis were included into the list of potential predictors. Table 5 reports the significant predictors of each outcome.

High BMI (≥30) was also tested as a specific potential predictor of DFU recurrence or death. It entered the model as a predictor neither of DFU recurrence nor of death. When high BMI was included into the list of potential predictors,

Table 3 Features of patients with healed diabetic foot ulcers stratified by absence or presence of ulcer recurrence

Variables	Healed ulcers ($n = 464$)	No ulcer recurrence ($n = 292$)	Ulcer recurrence ($n = 172$)	p value
Age (years)	70.5 ± 8.4	71.2 ± 8.3	69.4 ± 8.5	0.0301
Males (%)	56.7	55.1	59.3	0.3823
BMI	27.9 ± 3.9	27.7 ± 4.1	28.3 ± 3.7	0.1672
Smokers (%)	32.5	29.4	37.8	0.0644
Hypertension (%)	72.6	70.1	75.6	0.2124
Diabetes duration (years)	13.5 ± 8.0	13.5 ± 8.2	13.4 ± 7.6	0.9018
HbA1c (%)	7.4 ± 1.3	7.4 ± 1.4	7.5 ± 1.3	0.2494
Cholesterol (mg/dl)	167.8 ± 36.2	166.5 ± 35.0	170.1 ± 38.2	0.3037
HDL (mg/dl)	45.6 ± 11.7	45.3 ± 11.4	46.0 ± 12.2	0.5448
Triglycerides (mg/dl)	116.9 ± 44.1	117.3 ± 40.8	116.3 ± 49.4	0.5478
LDL (mg/dl)	98.9 ± 33.8	97.7 ± 32.5	101.0 ± 35.9	0.3144
eGFR (mL/min)	66.8 ± 20.7	67.0 ± 20.8	66.5 ± 20.4	0.8136
Micro- or macroalbuminuria (%)	40.9	35.3	50.6	0.0012
Previous CVD (%)	26.5	18.8	39.5	<0.0001
Previous DFU (%)	20.5	12.00	34.9	<0.0001
Previous amputation (%)	4.7	2.0	9.3	0.0004
Neuropathic ulcer (%)	47.2	56.8	30.8	<0.0001
Vascular Ulcer (%)	19.0	15.1	25.6	0.0053
Neurovascular ulcer (%)	31.9	25	43.6	<0.0001
Absence of neuropathy and PAD (%)	1.9	3	0	0.0298
ABI	0.95 ± 0.27	0.97 ± 0.28	0.92 ± 0.25	0.0427
TcPO2 (mmHg)	41.1 ± 15.1	45.6 ± 16.1	41.7 ± 12.9	0.0074
Lower Limb Revascularization (%)	31.9	34.6	27.3	0.1053
Osteomyelitis (%)	13.4	12.3	15.1	0.3945

eGFR estimated glomerular filtration rate, *CVD* cardiovascular disease, *DFU* diabetic foot ulcer, *TcPO2* transcutaneous oxygen tension, *BMI* body mass index, *ABI* ankle brachial index, *PAD* peripheral artery disease, *DFU* diabetic Foot ulcer

multivariate analysis showed that age (Regression coefficient β: 1.567; Standard error SE: 0.751; OR: 4.793; 95% CI: 1.098–20.910; $p = 0.0370$) was a predictor of DFU recurrence, in addition to eGFR and previous CVD, and that TcPO2 (Regression coefficient β: 0.630; Standard error SE: 0.299; OR: 1.878; 95% CI: 1.043–3.380; $p = 0.0355$) was a predictor of death, in addition to age, diabetes, duration, eGFR, previous amputation, DFU persistence, and major amputation.

Discussion

This large retrospective study shows both confirmatory and new findings on the predictors of outcomes in diabetic patients with DFU.

A younger age (<65 years) is a strong predictor of healing in our patients with DFU. To the best of our knowledge, this association is described for the first time, even if it is quite expected. Indeed, it is well-known that failure in wound repair is strongly linked to aging [15]. On the other hand, this explains why older age is associated to negative outcomes of DFU, in particular to amputations and death, in several studies [16–20].

Renal function plays a major role in the progression of DFU. We found that both eGFR ≥60 ml/L and absence of microalbuminuria are strongly associated with wound healing. Conversely, an impaired renal function, defined as an eGFR <60 ml/L and/or the presence of micro/macroalbuminuria, strongly predicts negative outcomes of DFU, such as minor and major amputation, persistence of active ulcer and death. Previous studies described a strong association between any impairment of kidney function, including dialysis, with DFU recurrence [21], amputations, and death [16, 19, 20]. This emphasizes not only the importance of renal function in the stratification of the risk for the occurrence of negative outcomes in patients with DFU, but also the need for any strategy to avoid the decline of kidney function, including the use of innovative drugs [22].

Among the predictors, we found that a history of CVD can have a great impact on the progression of DFU to negatives outcomes. Interestingly, the absence of a history

Table 4 Features of the of type 2 diabetic patients subdivided into two groups of non-survivors and survivors during the follow-up period

Feature	Dead ($n = 99$)	Alive ($n = 484$)	p value
Age (years)	79.6 ± 7.5	69.5 ± 8.0	<0.0001
Males (%)	48.5	57.4	0.1024
BMI	25.0 ± 3.9	28.3 ± 4.0	<0.0001
Smokers (%)	32.3	32.6	0.9505
Hypertension (%)	77.8	72.1	0.2469
Diabetes duration (years)	22.7 ± 10.5	12.8 ± 7.4	<0.0001
HbA1c (%)	7.3 ± 1.3	7.4 ± 1.4	0.3292
Cholesterol (mg/dl)	171.7 ± 39.0	169.3 ± 37.2	0.5648
HDL (mg/dl)	45.9 ± 13.4	45.6 ± 11.6	0.7862
Triglycerides (mg/dl)	119.6 ± 45.1	118.3 ± 44.5	0.7934
LDL (mg/dl)	101.8 ± 35.6	100.1 ± 34.8	0.6578
eGFR (mL/min)	49.0 ± 20.5	66.8 ± 20.3	<0.0001
Micro- or macroalbuminuria (%)	56.6	44.0	0.0225
Previous CVD (%)	52.5	30.0	<0.0001
Previous DFU (%)	44.4	23.5	<0.0001
Previous amputation (%)	26.2	5.2	<0.0001
Neuropathic ulcer (%)	22.2	45.6	<0.0001
Vascular ulcer (%)	18.2	20.4	0.6073
Neurovascular ulcer (%)	59.6	32.0	<0.0001
Absence of neuropathy and PAD (%)	0	1.9	0.1719
ABI	0.91 ± 0.30	0.94 ± 0.27	0.2706
TcPO2 (mmHg)	37.3 ± 13.2	43.4 ± 15.4	0.0003
Lower limb revascularization (%)	38.4	33.5	0.3486
Osteomyelitis (%)	36.4	15.7	<0.0001
Healing (%)	40.4	87.6	<0.0001
Persistence of active ulcers (%)	26.2	2.9	<0.0001
Minor amputations (%)	14.1	9.1	0.1264
Major amputations (%)	19.2	0.4	<0.0001
Ulcer recurrence (%)	21.2	31.6	0.0395
Follow-up duration (months)	41.4 ± 25.6	43.1 ± 22.9	0.5102

eGFR estimated glomerular filtration rate, *CVD* cardiovascular disease, *DFU* diabetic foot ulcer, *TcPO2* transcutaneous oxygen tension, *BMI* body mass index, *ABI* ankle brachial index, *PAD* peripheral artery disease, *DFU* diabetic Foot ulcer

of CVD is significantly predictive of healing, while its presence correlates very well con DFU persistence, minor amputation, and DFU recurrence. This confirms previous studies [20, 21] and highlights that subjects with advanced end-organ diseases, in particular when kidney and CVDs are both present, are at very high risk for worse prognosis, as outlined by the EURODIALE study and confirmed by a recent investigation [23, 24]. In other words, the general condition of the patient is very important in the evolution of DFU.

Our data for the first time show that low BMI is a strong predictor of DFU persistence and death. Two previous studies observed that high BMI rather than low BMI was associated with worse outcomes, in particular with delay in healing and amputation [3, 17]. These conflicting findings may be due to differences among the studies. As an association of high BMI with some outcomes of DFU was found [3, 17], the hypothesis that BMI may describe a so-called "J-curve" was taken into account and therefore we repeated the multivariate analyses regarding DFU persistence and death after inclusion of high BMI in the list of potential predictors. Nevertheless, these analyses showed that high BMI did not enter the prediction model. Low BMI is often associated to terminal stages of chronic degenerative diseases, such as kidney and heart failure. This may imply that in our study the association between low BMI and some outcomes may be the expression of the concomitant advanced end-organ diseases. Really, in our analysis low BMI was independent of both eGFR and previous CVD, as this independence was found in the multivariate analysis after adjustment for co-variates. On the other hand, it is important to remember that type 2 diabetic patients often have a high BMI. Therefore, when they have a low BMI, malnutrition may be present [25]. Malnutrition may be due not only to the general conditions of the patients but also to the presence of chronic inflammation and hormonal changes [26]. Therefore, our data seem to support the hypothesis that malnutrition may have a role in ulcer occurrence and in its progression [25, 27].

A well-known predictor of DFU outcome is osteomyelitis. We observed that the absence of osteomyelitis is a good predictor of wound healing. In addition, our study shows that osteomyelitis greatly increases the risk for amputation, in particular for major amputation, as also seen in other studies [3, 28, 29]. This implies the need for an immediate assessment for the presence of osteomyelitis and an aggressive medical treatment to reduce the occurrence of amputation.

A new finding of our study regards the intriguing potential role of two markers of PAD, such as TcPO2 and ABI, in the prevention of complications of DFU. We observed that a TcPO2 <46 mmHg correlates very well with DFU recurrence in subjects with healed lesions, while a normal TcPO2 predicts healing. In addition, we observed that an ABI <0.9 is associated with a greater proportion of minor amputations, as also found by others [19]. Ischemia has a major role in ulcer occurrence and recurrence [24, 30]. So, improvement of tissue oxygenation may play a potential role in the prevention of new DFUs. In our study revascularization was made in a large proportion of patients according to the established criteria for the procedure, but generally in patients with TcPO2 <40 mmHg and ABI <0.8, as also suggested by the Wi-Fi classification [31]. According to the WiFI classification [31] a subject with TcPO2 <46 mmHg and ABI <0.9 should be categorized as "grade 1" (TcPO2 40–59; ABI 0.6–0.79). In the context of a given category of "wound" and "foot infection", this does not usually allow to categorize this subject as a "high or very high risk" subject for estimate risk of amputation at 1 year or estimate likelihood of benefit of/requirement for revascularization. However, if our data are confirmed,

Table 5 Predictors of each outcome according to the results of each multivariate analysis with a stepwise approach

Predictors	Regression coefficient β	Standard error SE	Odds ratio	95% CI	p value
Healing					
Age	−0.934	0.341	0.392	0.201–0.766	0.0062
eGFR	−0.671	0.249	0.511	0.313–0.832	0.0070
Micro/macroalbuminuria	−0.632	0.255	0.531	0.322–0.875	0.0131
Previous CVD	−0.993	0.252	0.370	0.225–0.607	0.0001
Previous DFU	−1.088	0.253	0.336	0.204–0.554	<0.0001
TcPO2	−0.730	0.266	0.481	0.285–0.811	0.0061
Osteomyelitis	−1.372	0.267	0.253	0.150–0.428	<0.0001
Persistent active ulcer					
BMI	1.041	0.374	2.833	1.359–5.902	0.0054
Previous CVD	1.408	0.363	4.090	2.005–8.341	0.0001
eGFR	1.401	0.383	4.059	1.915–8.603	0.0003
Minor amputation					
Previous CVD	0.993	0.325	2.700	1.426–5.109	0.0023
Previous DFU	1.693	0.329	5.439	2.852–0.371	<0.0001
ABI	0.720	0.322	2.056	1.091–3.872	0.0256
Osteomyelitis	1.645	0.342	5.184	2.647–10.152	<0.0001
Micro/macroalbuminuria	0.688	0.338	1.990	1.026–3.861	0.0418
Major amputation					
Micro/macroalbuminuria	1.765	0.660	5.842	1.602–21.307	0.0075
Previous amputation	2.891	0.674	18.013	4.801–67.581	<0.0001
Osteomyelitis	3.637	0.676	37.995	10.091–143.059	<0.0001
DFU recurrence					
Previous CVD	0.746	0.231	2.109	1.340–3.319	0.0013
Previous DFU	1.292	0.252	3.640	2.221–5.966	<0.0001
TcPO2	0.851	0.211	2.342	1.548–3.543	<0.0001
Death					
Age	1.229	0.523	3.420	1.226–9.539	0.0188
BMI	1.124	0.301	3.077	1.703–5.558	0.0002
eGFR	1.014	0.287	2.758	1.570–4.847	0.0004
Previous amputation	1.424	0.420	4.156	1.824–9.472	0.0007
Persistent active ulcer	1.772	0.508	5.888	2.172–15.964	0.0005
Major amputation	3.365	0.894	28.946	5.011–167.209	0.0002

95% CI 95% confidence interval, *eGFR* estimated glomerular filtration rate, *CVD* cardiovascular disease, *DFU* diabetic foot ulcer, *TcPO2* transcutaneous oxygen tension, *BMI* body mass index, *ABI* ankle brachial index

specific studies should evaluate whether revascularization can reduce DFU recurrence and minor amputation in selected patients with TcPO2 <46 mmHg and ABI <0.9.

Taken together, our study shows that a younger age and absence of complications and risk factors are often associated with a high percentage of healing, while both advanced end-organ disease (in particular renal and CVD), local conditions (previous DFU, previous or current amputation, osteomyelitis) are associated with progression to negative outcomes. Our data show two new important predictors of outcome: low BMI with possible malnutrition, and a reduced oxygenation of limbs, represented by TcPO2 <46 mmHg and ABI <0.9. Both conditions predict a progression of DFU. These findings seem to suggest the need for the implementation of nutritional programs and of a revascularization in a larger proportion of patients with PAD.

As for the incidence of complications in our study population, we think that some considerations may be of interest.

We observed that the incidence of DFU recurrence is quite similar to that described in the literature [4], but we found a greater proportion of persons who healed in a quite short average time (7.6 months) and a lower percentage of amputations, persistence of active ulcers and death. These

results may be due to differences among the studies: differences in age, gender, type of diabetes (type 1, type 2 or both), diabetes duration, setting (outpatient department or hospitalization), co-morbidities, duration of follow-up and so on. However, we cannot exclude that a rigorous and comprehensive protocol of care, characterized by the presence of a multidisciplinary team, a holistic approach, an aggressive and early treatment of all risk factors, complications and co-morbidities, a nutritional support, a conservative care and an early rehabilitation may have a great importance in reducing the progression of DFU, as also suggested by several studies [2–5, 16, 25, 32–35]. Another reason for our low incidence of worse outcomes may be linked to the fact that only patients with lesions occurred no more than 30 days before the recruitment were enrolled in the study. Indeed, if an ulcer is not adequately treated for a long time, the risk for worse outcomes can increase. This suggests that an early treatment in patients with DFU may improve their prognosis.

Another important topic regards the identification of tools to improve the outcomes. As already discussed, a higher percentage of revascularization in patients with less severe PAD may be a tool. In addition, PAD may be used to identify subjects with CVD that is a strong predictor of negative outcomes. Indeed, PAD is often associated with CVD [36], that may be also asymptomatic [6, 37]. On the other hand, TcPO2 and ABI seems to predict cardiovascular events [11] and an early treatment of subjects with asymptomatic CVD may improve their prognosis [7].

As for the local treatment of DFU, to obtain a rapid and complete healing, in addition to the well codified strategies (cleaning and dressing, periodic debridement, early treatment of infection, use of specific technologies, such as negative pressure therapy, pressure offloading, indication for specific footwear), emerging treatments should be considered [38], such as regenerative medicine, ozono or microbiota, considering the strong association between microbiota with diabetes and its complications [39, 40].

The present study shows both limitations and strengthens. The main limitation is the retrospective study design, even if the patients attending our center were followed-up by periodic examinations and telephone interviews even after they recovered. Strengthens appear to be the rigorous inclusion and exclusion criteria, a precise protocol of care performed for all the follow-up period by the same team and a complete evaluation and treatment of all risk factors and co-morbidities. In particular, to limit confounding factors we opted for a rigorous study protocol. Only patients with recent ulcer (occurrence <30 days) were recruited. This choice was made for the following reasons: (a) all the parameters evaluated at the moment of the recruitment exactly reflect the condition of the patient at the moment of the occurrence of the ulcer and therefore they can be reliable predictors of outcomes. (b) if an ulcer is "old", we cannot know how the parameters were at the moment of its occurrence, as weight, eGFR, HbA1c, co-morbidities, complications and so on can vary over the time. In many patients these parameters could be greatly varied and therefore their predictive power may be lost or modified. (c) If an ulcer is not adequately treated for a long time, the risk for worse outcomes can increase and therefore the predictive value of variables is less reliable. Patients with multiple ulcers were also excluded for the following reasons: (a) different ulcers often occur in different periods. (b) Different ulcers may have different outcomes that depend not only on the general features of the patient but also on local conditions, time of occurrence, early treatment and so on. All the above conditions are evident confounding factors. The follow-up period of the study was long. In the study protocol a follow-up period of at least 12 months is an inclusion criterium. But, if a hard outcome (death or amputation) occurs before this time frame, the follow-up is obviously stopped for the outcome death, while the follow-up continues for the patients with the amputation and only the outcome death is recorded in these patients. Among 583 patients, only 11 had a follow-up <12 months: ten patients died and one had a major amputation, but he was followed-up for 23 months after amputation and death did not occur. At last, we noted that some predictors of outcomes (in particular osteomyelitis for major amputation and major amputation for mortality) have wide 95% CI and this suggests that caution should be paid in drawing conclusions based on these results, even if our findings on the risk for major amputation linked to osteomyelitis and for mortality linked to major amputation are confirmatory of previous studies [3, 28, 29, 34, 35]. These wide 95% CI may be due to the small sample of patients with major amputation.

Conclusions

Our study confirms the role of several predictors of outcome of DFU, but shows new information on BMI, TcPO2, and ABI as potential predictors of outcomes of DFU. This may imply the importance of new strategies for nutrition support and revascularization. In addition, the low incidence of some complications, in particular of amputation, persistence of active ulcer and death, may be due to the holistic and conservative approach, to the multidisciplinary team, to the early treatment, and to an early and aggressive management of all risk factors and co-morbidities.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

1. P. Zhang, J. Lu, Y. Jing, S. Tang, D. Zhu, Y. Bi, Global epidemiology of diabetic foot ulceration: a systematic review and meta-analysis. Ann. Med. **49**, 106–116 (2017)
2. A.F. Mavrogenis, P.D. Megaloikonomos, T. Antoniadou, V.G. Igoumenou, G.N. Panagopoulos, L. Dimopoulos et al. Current concepts for the evaluation and management of diabetic foot ulcers. EFORT Open Rev. **3**, 513–525 (2018)
3. L.M.A. Dutra, M.C. Melo, M.C. Moura, L.A.P. Leme, M.R. De Carvalho, A.N. Mascarenhas et al. Prognosis of the outcome of severe diabetic foot ulcers with multidisciplinary care. J. Multidiscip. Healthc. **12**, 349–359 (2019)
4. D.G. Armstrong, A.J.M. Boulton, S.A. Bus, Diabetic foot ulcers and their recurrence. N. Engl. J. Med. **376**, 2367–2375 (2017)
5. W.J. Jeffcoate, L. Vileikyte, E.J. Boyko, D.G. Armstrong, A.J.M. Boulton, Current challenges and opportunities in the prevention and management of diabetic foot ulcers. Diabetes Care. **41**, 645–652 (2018)
6. International Working Group on the Diabetic Fooy. IWGDF guidelines on the prevention and management of diabetic foot disease (2019), https://iwgdfguidelines.org/wp-content/uploads/2019/05/IWGDF-Guidelines-2019.pdf. Accessed 14 Apr 2020
7. C. Gazzaruso, A. Coppola, T. Montalcini, C. Valenti, G. Pelissero, S.B. Solerte et al. Screening for asymptomatic coronary artery disease can reduce cardiovascular mortality in type 2 diabetic patients. Intern. Emerg. Med. **7**, 257–266 (2012)
8. C. Gazzaruso, A. Coppola, T. Montalcini, E. Baffero, A. Garzaniti, G. Pelissero et al. Lipoprotein(a) and homocysteine as genetic risk factors for vascular and neuropathic diabetic foot in type 2 diabetes mellitus. Endocrine **41**, 89–95 (2012)
9. C. Gazzaruso, S.B. Solerte, A. Pujia, A. Coppola, M. Vezzoli, F. Salvucci et al. Erectile dysfunction as a predictor of cardiovascular events and death in diabetic patients with asymptomatic coronary artery disease angiographically proven. A potential protective role for statins and 5-phosphodiesterase inhibitors. J. Am. Coll. Cardiol. **51**, 2040–2044 (2008)
10. C. Gazzaruso, A. Garzaniti, S. Giordanetti, C. Falcone, P. Fratino, Silent coronary artery disease in type 2 diabetes mellitus: the role of Lipoprotein(a), homocysteine and apo(a) polymorphism. Cardiovasc. Diabetol. **22**, 1–5 (2002)
11. C. Gazzaruso, A. Coppola, C. Falcone, C. Luppi, T. Montalcini, E. Baffero et al. Transcutaneous oxygen tension as a potential predictor of cardiovascular events in type 2 diabetes: comparison with ankle-brachial index. Diabetes Care **36**, 1720–1725 (2013)
12. W.T. Friedewald, R.I. Levy, D.S. Fredrickson, Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifugate. Clin. Chem. **18**, 499–502 (1972)
13. D.W. Cockcroft, M.H. Gault, Prediction of creatinine clearance from serum creatinine. Nephron **16**, 31–41 (1976)
14. C. Gazzaruso, S. Giordanetti, E. De Amici, G. Bertone, C. Falcone, D. Geroldi et al. Relationship between erectile dysfunction and silent myocardial ischemia in apparently uncomplicated type 2 diabetic patients. Circulation **110**, 22–26 (2004)
15. S.A. Eming, P. Martin, M. Tomic-Canic, Wound repair and regeneration: mechanisms, signaling, and translation. Sci. Transl. Med. **6**, 265sr6 (2014)
16. E. Ghanassia, L. Villon, J.F. Thuan Dit Dieudonné, C. Boegner, A. Avignon, A. Sultan, Long-term outcome and disability of diabetic patients hospitalized for diabetic foot ulcers: a 6.5-year follow-up study. Diabetes Care. **31**, 1288–1292 (2008)
17. O. Sarfo-Kantanka, F.S. Sarfo, I. Kyei, C. Agyemang, J.C. Mbanya, Incidence and determinants of diabetes-related lower limb amputations in Ghana, 2010-2015- a retrospective cohort study. BMC Endocr. Disord. **19**, 27 (2019)
18. S. Peled, R. Pollack, O. Elishoov, A. Haze, A. Cahn, Association of inpatient glucose measurements with amputations in patients hospitalized with acute diabetic foot. J. Clin. Endocrinol. Metab. **104**, 5445–5452 (2019)
19. K. Ikura, K. Hanai, T. Shinjyo, Y. Uchigata, HDL cholesterol as a predictor for the incidence of lower extremity amputation and wound-related death in patients with diabetic foot ulcers. Atherosclerosis. **239**, 465–469 (2015)
20. M.J. Young, J.E. McCardle, L.E. Randall, J.I. Barclay, Improved survival of diabetic foot ulcer patients 1995-2008: possible impact of aggressive cardiovascular risk management. Diabetes Care. **31**, 2143–2147 (2008)
21. A. Akturk, J.J. van Netten, R. Scheer, M. Vermeer, J.G. van Baal, Ulcer-free survival days and ulcer healing in patients with diabetic foot ulcers: a prospective cohort study. Int Wound J **16**, 1365–1372 (2019)
22. C. Gazzaruso, A. Coppola, T. Montalcini, C. Falcone, Antidiabetic agents and heart health: how to use new diabetes medications in a global strategy for the prevention of cardiovascular complications in type 2 diabetes. Ann. Transl. Med. **6**, 195 (2018)
23. L. Prompers, N. Schaper, J. Apelqvist, M. Edmonds, E. Jude, D. Mauricio et al. Prediction of outcome in individuals with diabetic foot ulcers: focus on the differences between individuals with and without peripheral arterial disease. The EURODIALE Study. Diabetologia **51**, 747–755 (2008)
24. M. Meloni, V. Izzo, L. Giurato, J.L. Lazaro-Martinez, L. Uccioli, Prevalence, clinical aspects and outcomes in a large cohort of persons with diabetic foot disease: comparison between neuropathic and ischemic ulcers. J. Clin. Med. **9**, 1780 (2020)
25. A.M. Quain, N.M. Khardori, Nutrition in wound care management: a comprehensive overview. Wounds **27**, 327–335 (2015)
26. T. Montalcini, S. Romeo, Y. Ferro, V. Migliaccio, C. Gazzaruso, A. Pujia, Osteoporosis in chronic inflammatory disease: the role of malnutrition. Endocrine **43**, 59–64 (2013)
27. L. Haughey, A. Barbul, Nutrition and lower extremity ulcers: causality and/or treatment. Int. J. Low. Extrem. Wounds **16**(4), 238–243 (2017)
28. M. Arias, S. Hassan-Reshat, W. Newsholme, Retrospective analysis of diabetic foot osteomyelitis management and outcome at a tertiary care hospital in the UK. PLoS ONE **14**, e0216701 (2019)
29. P. Sen, T. Demirdal, B. Emir, Meta-analysis of risk factors for amputation in diabetic foot infections. Diabetes Metab. Res. Rev. **35**, e3165 (2019)
30. J.A. Apelqvist, M.J. Lepäntalo, The ulcerated leg: when to revascularize. Diabetes Metab. Res. Rev. **28**(Suppl 1), 30–35 (2012)
31. J.L. Mills, M.S. Conte, D.G. Armstrong et al. Society for vascular surgery lower extremity guidelines committee. the society for vascular surgery lower extremity threatened limb classification system: risk stratification based on wound, ischemia, and foot infection (WIfI). J. Vasc. Surg. **59**(1), 220–234 (2014)
32. S.M. Taylor, B.L. Johnson, N.L. Samies, R.D. Rawlinson, L.E. Williamson, S.A. Davis et al. Contemporary management of diabetic neuropathic foot ulceration: a study of 917 consecutively treated limbs. J Am Coll Surg **212**, 532–545 (2011)
33. B. Hartmann, C. Fottner, K. Herrmann, T. Limbourg, M.M. Weber, K. Beckh, Interdisciplinary treatment of diabetic foot wounds in the elderly: low risk of amputations and mortality and good chance of being mobile with good quality of life. Diabetes Vasc. Dis. Res. **14**, 55–58 (2017)
34. Y.Y. Huang, C.W. Lin, H.M. Yang, S.Y. Hung, I.W. Chen, Survival and associated risk factors in patients with diabetes and

amputations caused by infectious foot gangrene. J. Foot Ankle Res. **11**, 1 (2018)
35. L. Monge, R. Gnavi, P. Carnà, F. Broglio, G.M. Boffano, C.B. Giorda, Incidence of hospitalization and mortality in patients with diabetic foot regardless of amputation: a population study. Acta Diabetol. **57**, 221–228 (2020)
36. C. Falcone, S. Bozzini, L. Guasti, A. D'Angelo, A.C. Capettini, E. M. Paganini et al. Soluble RAGE plasma levels in patients with coronary artery disease and peripheral artery disease. ScientificWorldJournal. **2013**, 584504 (2013)
37. C. Gazzaruso, S.B. Solerte, E. De Amici, M. Mancini, A. Pujia, P. Fratino et al. Association of the metabolic syndrome and insulin resistance with silent myocardial ischemia in patients with type 2 diabetes mellitus. Am. J. Cardiol. **97**, 236–239 (2006)
38. J.M. Ramirez-Acuña, S.A. Cardenas-Cadena, P.A. Marquez-Salas, I. Garza-Veloz, A. Perez-Favila, M.A. Cid-Baez et al. Diabetic foot ulcers: current advances in antimicrobial therapies and emerging treatments. Antibiotics **8**(4), pii: E193 (2019)
39. A. Pascale, N. Marchesi, C. Marelli, A. Coppola, L. Luzi, S. Govoni et al. Microbiota and metabolic diseases. Endocrine **61**, 357–371 (2018)
40. A. Pascale, N. Marchesi, S. Govoni, A. Coppola, C. Gazzaruso, The role of gut microbiota in obesity, diabetes mellitus, and effect of metformin: new insights into old diseases. Curr. Opin. Pharmacol. **49**, 1–5 (2019)