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Comparative analysis of microcirculation and oxidative metabolism indicators in elderly people with diabetes mellitus with and without diabetic foot using laser Doppler flowmetry and laser fluorescence spectroscopy

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Abstract. The combined use of laser Doppler flowmetry and laser fluorescence spectroscopy is a topical area of modern clinical diagnostics, allowing not only quantitative assessment of tissue perfusion in patients with diabetes mellitus, but also to analyse the mechanisms of microcirculation regulation, which is especially important in the treatment of patients with diabetic foot and chronic ischaemia. The aim of this study was to conduct a comparative analysis of microcirculation and oxidative metabolism indicators in elderly people with diabetes mellitus with and without diabetic foot. The study involved elderly patients in two groups, aged 60-75 years, with 20 people in each group ($n = 40$, men – 26 (65%), women – 14 (35%), $p < 0.001$). Analysis of the data showed that in 35% of patients with type 2 diabetes mellitus, microcirculation indicators were within normal limits compared to patients with type 2 diabetes mellitus complicated by diabetic foot (35% vs. 0, $p < 0.001$). The decrease in oxidative metabolism indicators in patients with type 2 diabetes mellitus complicated by diabetic foot was 3 times greater than in patients with type 2 diabetes mellitus ($p < 0.05$). This, in turn, led to a compensatory threefold increase in the amplitude of nicotinamide adenine dinucleotide coenzyme in patients with diabetic foot ($p < 0.05$). The presence of normal microcirculation indicators in laser Doppler flowmetry in patients with diabetes mellitus does not exclude the risk of developing diabetic foot. The decrease in oxidative metabolism indicators in patients with diabetic foot was a consequence of oxidative stress caused by chronic hyperglycaemia, microvascular disorders, and inflammation, which led to tissue trophism damage in this category of patients. Early understanding of the state of microcirculation and trophism of the foot in patients with diabetes and intervention in muscle, nerve, and endothelial function may be an effective way to improve microcirculation of the foot and prevent diabetic ulcers

Keywords: microcirculation; laser Doppler flowmetry; laser fluorescence spectroscopy; oxidative metabolism; type 2 diabetes mellitus; diabetic foot

Introduction

Diabetes mellitus (DM) is a global health problem. In 2021, the International Diabetes Federation estimated that the prevalence of diabetes among people aged 20 to 79 had become widespread, reaching 10.5% or 536.6 million people. Forecasts indicate that this figure will increase to 12.2% (783.2 million people) by 2045 [1]. One of the most common and serious complications of diabetes is diabetic foot (DF) [2]. However, as

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the body ages, there are also significant changes in the microcirculation and tissue metabolism, especially in the lower extremities, which can contribute to the development of chronic ischaemic conditions and tissue trophism disorders [3].

Contradictory results of comparisons of baseline microcirculation in elderly patients with diabetes and in relatively healthy volunteers suggest that the assessment of baseline perfusion associated with diabetes is not very informative [4,5]. Some authors found no significant differences, others noted a tendency towards increased baseline blood flow in patients with diabetes [6-8], and some researchers reported a decrease in baseline blood flow in patients with diabetes. Thanks to the portability of wireless technologies, laser Doppler flowmetry (LDF) and laser fluorescence spectroscopy (LFS) are suitable for daily use [9,10].

The results of using wireless portable dynamic light scattering sensors that implement laser Doppler flowmetry signal processing have confirmed that hardware testing of volunteers can detect microvascular changes caused by both diabetes and ageing [3,11,12]. Continuous and adequate blood flow is vital for cell function; thus, tissue perfusion, usually quantified by measuring the volume of blood passing through the microvascular network in a given volume of tissue over a given period of time, is a key indicator of organ or tissue health. Since regional blood flow strongly influences the metabolic activity of cells, it is extremely important for the authors of this work to understand the nuances of the characteristics and reactivity of microvessels, which change with age, in order to understand the progressive deterioration of tissue function due to age-related metabolic activity disorders. To better understand blood flow in microvessels, microscopy can be combined with surface electrode measurements of partial oxygen pressure. Alternatively, laser Doppler flowmetry can be used as a standalone method for assessing microvascular function, as it allows both flow and erythrocyte velocity to be measured [3,11].

Wearable LDF has a unique advantage in the rapid assessment of plantar microcirculation in the early stages of diabetic ulcers. In combination with wavelet analysis, it allows quantitative and qualitative analysis of the mechanism of foot microcirculation regulation. Compared to healthy adults, patients with diabetes had significantly lower foot microcirculation when using portable LDF, mainly in the plantar region near the distal end of the foot [8]. Diagnosis of blood circulation in the microvessels of the skin and oxidative metabolism of biological tissue in elderly people with diabetes, which allows predicting the presence of microcirculatory disorders in the body, assessing their severity and monitoring during therapy, is a relevant area of research. The aim of the study was to conduct a comparative analysis of microcirculation and oxidative metabolism indicators in elderly people with type 2 diabetes mellitus with and without diabetic foot using a combination of LDF and LFS of the lower extremities.

Materials and Methods

The study included 40 elderly patients aged 60-75 years. The study was conducted in the diabetic foot department of the M.A. Topchubashev Scientific Surgery Centre over a period of two months (April, May 2025). There were 26 men (65%) and 14 women (35%), with statistical differences observed between patients by gender ($\chi^2 = 7.200$, f = 0.008, $p < 0.001$). The patients were divided into two groups: 20 elderly patients with type 2 diabetes mellitus complicated by diabetic foot syndrome receiving inpatient treatment and 20 elderly volunteers with type 2 diabetes mellitus (Fig. 1).

Figure 1. Laser Doppler flowmetry and laser fluorescence spectroscopy of the lower extremities in patients with DF (left) and elderly volunteers with type 2 DM (right)

Source: created by the authors

The inclusion criteria were the presence of type 2 DM complicated by DF (stage 2, 3, or 4 DF according to Wagner) and without DF. The exclusion criteria were severe cardiovascular disorders, septic conditions, and significant cognitive impairments that prevented contact with the patient. The study area was the malar surface of the big toe. The studies were conducted at rest, without any functional tests, for 10 minutes, after 30 minutes of rest for the subjects. For the combined use of LDF and LFS, a portable "LAZMA PF" device was used. Informed consent was obtained from all patients for the study. The arithmetic mean values of perfusion (M), nutrient blood flow (Mnutr), oxidative metabolism indicators (OMI), linking the nutrient component of blood perfusion and the fluorescence amplitude of nicotinamide adenine dinucleotide coenzyme (Anadn) were analysed.

Statistical processing was performed using Statistics 16.0 software for Windows (StatSoft Inc, USA) and StatTech software (StatTech LLC, Russia). Quantitative indicators close to normal distribution were presented as arithmetic mean (M) and standard deviation (SD). Absolute values were compared using multi-field contingency tables with Pearson's chi-square test (χ^2) and Fisher's exact test (f). Differences between the compared values were considered statistically significant at a confidence level of $p < 0.050$ [13].

Results

The control values of microcirculation and oxidative metabolism indicators in persons over 50 years of age using the portable analyser "LAZMA PF" (in perfusion units) are as follows: M (microcirculation indicator) – 10.2-15.4; Mnutr (nutritive blood flow indicator) – 1.9-2.3; Anadn (fluorescence amplitude of nicotinamide adenine dinucleotide coenzyme) – 0.7-1.7; OMI (oxidative metabolism indicator) – 0.6-1.62. As can be seen from the data, out of 20 patients with type 2 diabetes mellitus, 7 patients (35%) had microcirculation (M) indicators within normal control values (Table 1).

Table 1. Results of examination of patients with type 2 diabetes mellitus who underwent microcirculation and oxidative metabolism testing (absolute and relative values)

Microcirculation and oxidative metabolism indicators	Number of patients with indicators within normal limits	Number of patients with indicators above the upper limit of control values	Number of patients with indicators below the lower limit of control values
M	7 (35%) ($p < 0.001$)	4 (20%) ($p > 0.05$)	9 (45%) ($p > 0.05$)
Mnutr	7 (35%) ($p < 0.001$)	9 (45%) ($p > 0.05$)	4 (20%) ($p > 0.05$)
OMI	5 (25%) ($p > 0.05$)	12 (60%) ($p > 0.05$)	3 (15%) ($p < 0.05$)
Anadn	6 (30%) ($p > 0.05$)	2 (10%) ($p < 0.05$)	12 (60%) ($p > 0.05$)

Source: created by the authors

Nine patients (45%) with type 2 DM had impaired microcirculation in the form of decreased microcirculation, and four patients (20%) with type 2 DM had impaired microcirculation in the form of increased microcirculation ($p > 0.05$). As for OMI, out of 20 patients with type 2 DM, 12 (60%) had an increased functional state of the microcirculatory-tissue system (FS MTS), which indicates that OMI values exceeded the upper limit of control values; in 3 (15%) patients, the FS MTS was in a state of decompensation. Only in 5 (25%) patients was the OMI within the control range (Table 1). In elderly patients with diabetic foot, the LDF pattern was slightly different (Table 2).

Table 2. Results of examination of patients with type 2 diabetes mellitus and diabetic foot who underwent examination of microcirculation and oxidative metabolism (absolute and relative values)

Microcirculation and oxidative metabolism indicators	Number of patients with indicators within normal limits	Number of patients with indicators above the upper limit of control values	Number of patients with indicators below the lower limit of control values
M	0 ($p < 0.001$)	8 (40%) ($p > 0.05$)	12 (60%) ($p > 0.05$)
Mnutr	0 ($p < 0.001$)	12 (60%) ($p > 0.05$)	8 (40%) ($p > 0.05$)
OMI	1 (5%) ($p > 0.05$)	10 (50%) ($p > 0.05$)	9 (45%) ($p < 0.05$)
Anadn	2 (10%) ($p > 0.05$)	6 (30%) ($p < 0.05$)	12 (60%) ($p > 0.05$)

Source: created by the authors

Microcirculation indicators M and Mnutr within the control values were not detected in any patient. Twelve (60%) patients with diabetic foot had microcirculation disorders in the form of decreased perfusion, and eight (40%) patients with diabetic foot had microcirculation disorders in the form of increased perfusion ($p > 0.05$). A decrease in OMI was found in 9 patients (45%) and an increase in OMI in 10 (50%) patients, and only in one patient (5%) was OMI within the control values (Table 2). There were significant statistical differences between the number of patients with type 2 diabetes mellitus and the number of patients with type 2 diabetes mellitus complicated by DF, in whom M and Mnutr values were within the normal range ($\chi^2 = 19.259$, $p < 0.001$) (Table 3).

Table 3. Results of statistical processing of microcirculation and oxidative metabolism indicators in patients with type 2 diabetes mellitus and the number of patients with type 2 diabetes mellitus complicated by diabetic foot

Microcirculation and oxidative metabolism indicators	Differences in values within the normal range	Differences in indicators above the upper limit of control values	Differences in indicators below the lower limit of control values
M	$\chi^2 = 19.259$	$\chi^2 = 1.905$ p = 0.168 f = 0.300	$\chi^2 = 0.902$ p = 0.343 f = 0.366
Mnutr	$\chi^2 = 19.259$	$\chi^2 = 0.902$ p = 0.343 f = 0.366	$\chi^2 = 1.905$ p = 0.168 f = 0.300
OMI	$\chi^2 = 3.137$ p = 0.077 f = 0.101	$\chi^2 = 0.404$ p = 0.526 f = 0.751	$\chi^2 = 4.289$ p = 0.039 f = 0.048
Anadn	$\chi^2 = 2.500$ p = 0.114 f = 0.139	$\chi^2 = 4.800$ p = 0.029 f = 0.065	$\chi^2 = 0.404$ p = 0.526 f = 0.546

Source: created by the authors

There were no statistical differences between the number of patients with type 2 DM and the number of patients with type 2 DM complicated by diabetic foot with OMI within the normal range and OMI exceeding the upper limit of control values ($p > 0.05$), but there were significant statistical differences in the OMI indicator below the lower limit of the control values ($\chi^2 = 4.289$, $p = 0.039$, $f = 0.048$, $p < 0.05$). When comparing Anadn indicators within the normal range and below the control values between the number of patients with type 2 DM and the number of patients with type 2 DM complicated by DF, no statistical differences were observed ($p > 0.05$). At the same time, statistical differences were observed when comparing these patients with Anadn indicators above the upper limit of control values ($p < 0.05$).

Discussion

Analysis of the data showed that in 35% of patients with type 2 DM, microcirculation parameters were within normal limits compared to patients with type 2 DM complicated by DF (35% vs. 0%, $p < 0.001$). This is because microcirculation disorders do not always manifest themselves in the early stages, and in some patients with diabetes, microcirculation may remain within normal limits until diabetic foot develops, as shown in the studies by I. Mizeva *et al.* [7] and A.V. Skripal *et al.* [11]. As for OMI indicators in LFS, statistical differences were observed when comparing groups of patients with OMI, in which this indicator was below the lower limit of control values in patients with type 2 diabetes and diabetic foot ($p < 0.05$). The decrease in OMI in patients with type 2 DM complicated by DF was 3 times greater than in patients with type 2 DM, which indicates the prevalence of trophic disorders in this group of patients. In this regard, a compensatory increase in Anadn by 3 times was noted in patients with diabetic foot ($p < 0.05$). This is explained by the fact that diabetic foot syndrome is accompanied by pronounced oxidative stress caused by chronic hyperglycaemia, microvascular disorders and inflammation, which leads to excessive formation of free radicals and tissue damage [5,11]. The lack of statistical significance between microcirculation disorders in patients with type 2 DM and patients with type 2 DM complicated by DF ($p > 0.05$) in LDF indicates that, despite the presence of diabetes, in 65% of patients, microcirculation in the tissues of the foot is not impaired to an extent that would be recorded by LDF as a statistically significant change.

Conclusions

Normal microcirculation parameters obtained by laser Doppler flowmetry in patients with diabetes mellitus do not exclude the risk of developing diabetic foot. Despite the presence of diabetes, in 65% of patients, microcirculation in the tissues of the foot is not impaired to such an extent that it can be recorded by LDF as a statistically significant change. Often, statistical significance and power are achieved not by increasing the magnitude of differences, but by including a larger number of subjects in the study. The decrease in oxidative metabolism indicators in patients with diabetic foot, as a statistically significant change, is a consequence of oxidative stress, which exacerbates cell and tissue damage, complex interactions between microcirculation disorders, neuropathy, and other factors associated with diabetes mellitus. A comprehensive assessment of the patient's condition and the use of additional diagnostic methods allow the causes of diabetic foot development to be identified and eliminated. In addition, LDF and LFS are instrumental methods for personalising treatment, allowing a transition from group statistical indicators to the analysis of the condition of a specific patient, which contributes to an improved prognosis and quality of life.

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Conflict of Interest

The authors declare that there is no conflict of interest.

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