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Evaluation of long-term treatment results of patients with critical limb ischemia and diabetes mellitus with different management approaches

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Abstract

Background. In all cases of critical lower limb ischemia revascularization should be performed. However, the overlay of diabetic angiopathy significantly worsens the prognosis of the surgery, and the conservative therapy also frequently turns out to be ineffective even in the nearest future. Therapeutic stimulation of angiogenesis in this group of patients is an additional application point for the possible improvement of treatment results.
Aim. Evaluating treatment results of patients with critical ischemia and underlying diabetes after 5 years from the beginning of supervising, as well as assessing the benefits of additional angiogenesis as a part of treatment.
Material and methods. The study included data from 140 patients with critical ischemia and diabetes mellitus, divided into 4 groups, who received surgical [groups IA (n=45) and IB (n=30)] or therapeutic [groups IIA (n=40) and IIB (n=25)] treatment. Additional angiogenic therapy was also carried out in two groups (groups IB and IIB). Treatment outcomes were assessed over a 5-year period. Efficacy criteria were evaluated in the form of limb preservation and patient survival. The results were studied using descriptive statistics methods. Qualitative data were analyzed by constructing 2×2 contingency tables and using the xi-square method.
Results. The best indices of limb preservation and the lowest mortality of patients were obtained in the group after revascularization with additional angiogenic therapy. Gene therapy induction significantly improved the course of disease in limb preservation for periods from 6 months to 2 years (30% amputations by 2 years of follow-up, p=0.045). Angiogenic induction of angiogenesis also made it possible to improve the treatment results in the groups of conservative therapy but reliably better indicators of limb preservation were obtained in two groups with surgical interventions.
Conclusion. The optimal management tactic for patients with critical lower limb ischemia and underlying diabetes mellitus is direct revascularizing intervention on the lower limb arteries with additional angiogenic therapy in the postoperative period. Additional gene therapy induction of angiogenesis makes it possible to reduce the number of amputations both as part of combined surgical or therapeutic treatment , but it cannot be an alternative to surgery.
Keywords: atherosclerosis, critical ischemia of the lower limbs, diabetes mellitus, angiogenic induction, gene therapy.

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Background

Critical lower-limb ischemia (CLLI) in the vast majority of cases (80%–90%) is caused by atherosclerosis [1]. Diabetes mellitus increases the incidence of atherosclerosis in lower-limb arteries by 53 times compared with patients without diabetes [2]. Atherosclerosis and diabetes mellitus are two independent diseases that potentiate their negative effects on peripheral arteries through several main mechanisms. Systemic hyperglycemia causes endothelial dysfunction in the body due to the inhibition of the synthesis of nitric oxide (NO) [3]. In the hemostasis system, there is a shift toward hypercoagulability, due to the activation of protein kinase C, manifestations of a systemic inflammatory reaction occur, and oxidative stress due to an increase in the amount of free radicals [4, 5].

With the chronic course of diabetes mellitus and inadequate correction of glycemia, the “diabetic paradox” is especially noteworthy, which consists

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in the manifestation of diabetic retinopathy due to hyperangiogenesis of highly permeable vessels in the retina, on the one hand, and insufficient arteriogenesis in lower-limb and heart muscles, on the other hand. Thus, severe ischemia and trophic ulcers occur, which ultimately leads to lower-limb amputations [6, 7].

To date, undoubtedly, all patients with critical ischemia should be considered candidates for direct revascularization surgery (i.e., open, endovascular, and hybrid) in the near future due to the high risk of the limb loss. Thus, according to the National Guidelines, angiogenic therapy is not indicated in these patients due to the lack of time and the first signs of revascularization emerged 3–4 weeks later [8].

Unfortunately, in this group of patients, limb revascularization is performed in approximately 50% of cases, whereas 25% undergo conservative therapy, and the remaining 25% are doomed to primary amputation because of frequent concomitant damage to the tibial arteries due to diabetic macro- and microangiopathy [9]. The number of amputations that are performed within 3 years after the establishment of a clinical diagnosis of diabetes-associated CLLI reaches 62.5% among patients who received conservative therapy and 42% in patients who underwent revascularization surgeries [10], the 5-year mortality of patients with diabetes-associated CLLI reaches 13%–43%, and 10-year mortality rate is 30%–70% [11].

Even if it is technically possible to perform reconstructive interventions, their results are not satisfactory. Thus, direct revascularization surgeries are feasible in 41.8% of patients, whereas in 41% of cases, thrombosis of the reconstruction zone occurs within 1 year [12]. Perioperative mortality in patients with critical ischemia reaches 14%. Considering the above data, the amputation rate of 20.4% within 6 months appears consistent [12, 13].

When using endovascular techniques, 1 year after surgery, 71% of patients did not undergo amputation, and limb preservation rate after 3 years was 52%–57% [13]. Moreover, although technically successful result is achieved in 85%–100% of cases and the clinical effect varies within 65%–91%, the 3-year primary patency in the infrainguinal segment was 52% [14].

The treatment results of patients with CLLI and diabetes mellitus are poor because of several main mechanisms:

– Unsatisfactory outflow paths and, as a result, a small proportion of initially feasible reconstructions;

– Frequent postoperative complications;

– Disease progression;

– Thrombosis of the reconstruction zone;

– Purulent-septic complications, endothelial dysfunction.

An additional method that can improve treatment results of patients is the stimulation of endogenous angiogenesis processes [15, 16]. The induction of angiogenesis is one of the techniques that can improve treatment results. According to the literature, exogenous stimulation of angiogenesis (plasmid p-VEGF-165) has no side and adverse effects and provided good treatment results in patients with intermittent claudication [17]. The administration of vascular endothelial growth factor result in the relief of ischemia after 6 months of follow-up, and the effect of the therapy was retained for up to 2 years [18]. Moreover, the long-term 5-year results of treatment of patients with diabetes-associated CLLI using angiogenic technologies have not been previously evaluated.

Aim

This study aimed to analyze the treatment of patients with diabetes-associated CLLI for 5 years, compare conservative and surgical approaches, and evaluate the benefits of additional angiogenesis as part of combined with surgical intervention and complex treatment (as part of conventional therapy).

Materials and methods of research

A prospective randomized controlled trial was conducted in 2015–2021 at the clinical sites of the Department of Cardiovascular, X-ray Endovascular Surgery, and Radiation Diagnostics of the I.P. Pavlov Ryazan State Medical University of the Ministry of Health of Russia (Department of Vascular Surgery, Regional Clinical Cardiology Dispensary, Regional Clinical Hospital, Ryazan," and Emergency Care Hospital).

The study was approved by the local ethics committee of the I.P. Pavlov Ryazan State Medical University of the Ministry of Health of Russia (Protocol No. 10 dated October 6, 2015).

Treatment outcomes were evaluated in 140 patients diabetes-associated CLLI. The study included patients who were first admitted to the department of vascular surgery for treatment, diagnosed with critical ischemia, and had diabetes mellitus as concomitant disease.

Patients were distributed into two main treatment approaches, namely, surgical (I) and conservative (II). The conservative therapy group had a peripheral vascular bed unsuitable for reconstruction and did not have urgent indications for limb amputation (including a minor one) or refused surgical intervention independently. Non-reconstructability of the vascular bed was indicated for

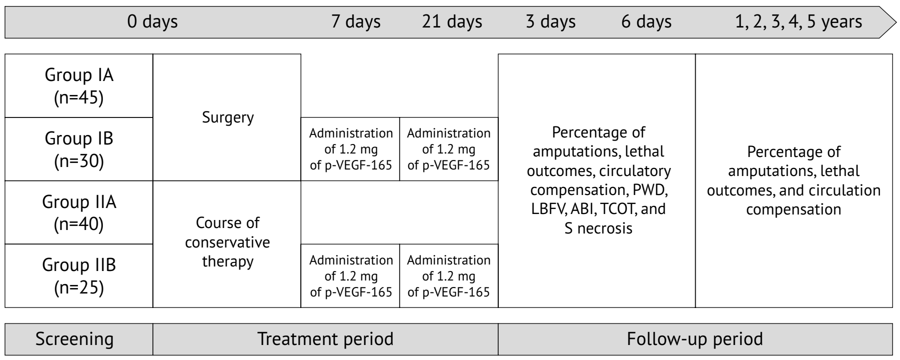

Fig. 1. Study design. PWD, painless walking distance; LBFV, linear blood flow velocity; ABI, ankle-brachial index; TCOT, transcutaneous oxygen tension.

lesions distal to the tibial arteries with the absence of satisfactory outflow tracts (>7 points according to the Rutherford classification) and widespread multifocal lesions of lower-limb arteries, in which a major reconstructive surgery is impossible because of a high intraoperative risk and general somatic background of the patient.

The inclusion criteria were as follows:

– Age ≥40 years, regardless of gender

– CLLI of atherosclerotic genesis (including the presence of dry necrosis with an area of no more than 3 cm^2; Wagner stages 0–2)

– Type 2 diabetes mellitus

– Lack of urgent indications for amputation

– Possibility of daily monitoring by patients of the level of glycemia

– Patients consented to comply with the requirements for examination and treatment

In addition, if distributed into the surgical or therapeutic group, the inclusion criteria were as follows: successful revascularization intervention on lower-limb arteries within the last 7 days or the impossibility of performing a revascularization surgery due to distal damage to the peripheral arteries of the leg (refusal from surgery).

The exclusion criteria were as follows:

– Age < 40 years;

– Limb ischemia of non-atherosclerotic genesis (thromboangiitis, arteritis, trauma, and anomalies in the development of blood vessels);

– Lack of patient adherence to treatment;

– History of myocardial infarction and stroke in the last 6 months;

– Decompensation of the cardiovascular system, kidney, or liver function;

– Decompensated type 2 and type 1 diabetes mellitus;

– Severe comorbidities with a life expectancy of <3 years;

– Malignant neoplasms (active or in history);

– Infectious conditions or sepsis;

– Pregnancy or lactation.

Group IA (n = 45) included patients who underwent direct revascularization surgeries using open, hybrid, or X-ray endovascular techniques. In group IIA (n = 40), patients who were recognized unsuitable for reconstructive interventions received a course of conservative therapy. The study groups (n = 55 total), including the surgical intervention (group IB, n = 30) and conservative treatment (group IIB, n = 25), received additionally a drug for therapeutic angiogenesis (plasmid superhelical deoxyribonucleic acid pCMV-VEGF-165).

Angiogenesis was induced by injection. In total, two series of injections (2–3 injections each) were performed into the ischemic zone of the anterolateral and posterior muscle groups in the middle lower third of the lower leg. Angiogenesis was induced, taking into account the angiosomal principle, in the projection lines of the anterior tibial artery and branches of the tibioperoneal trunk. The drug was administered at a dose of 1.2 μg twice, with an interval of 14 days.

Subsequently, this ischemic limb was monitored by a cardiovascular surgeon at the specialized department for 5 years through in-person (mandatory for 6 months) and off-site visits. Control visits were made every 6 months, 1 year, and then annually up to 5 years. During the first 6 months of follow-up, patients underwent ultrasound duplex scan of lower-limb arteries, measurement of transcutaneous oxygen tension, determination of the ankle-brachial index, and treadmill test. Subsequently, post-clinical follow-up was performed to determine the primary points of efficiency in the form of saving the patient's life and freedom from amputation (Fig. 1).

We analyzed the results of treatment of 140 patients (87 men and 53 women) who underwent inpatient treatment in the department of vascular surgery in 2015–2016. The mean age of the patients was 67.2 ± 7.7 years. Between the groups, there was a comparable distribution by gender and age,

Table 1. Distribution of patients according to the level of lesion within groups.

Lesion level	Group IA (n = 45)	Group IB (n = 30)	Group IIA (n = 40)	Group IIB (n = 25)
Iliac segment	6 (13.3%)	–	4 (10%)	2 (8%)
Femoropopliteal segment	25 (57.8%)	23 (76.7%)	24 (60%)	13 (52%)
Distal segment	4 (8.9%)	4 (13.3%)	8 (20%)	8 (32%)
Multifocal	10 (20%)	3 (10%)	4 (10%)	2 (8%)

Table 2. Summary data on comorbidities at inclusion in the study.

Comorbidity	Group IA (n = 45)	Group IB (n = 30)	Group IIA (n = 40)	Group IIB (n = 25)
Hypertension	35 (77.8%)	24 (80%)	24 (60%)	18 (72%)
Ischemic heart disease	32 (71.1%)	19 (63.3%)	14 (35%)	16 (64%)
Postinfarction cardiosclerosis	5 (11.1%)	12 (40%)	11 (27.5%)	5 (20%)
Cerebrovascular disease	9 (20%)	2 (6.7%)	3 (7.5%)	6 (24%)
Acute cerebrovascular accident	8 (17.8%)	1 (3.3%)	2 (5%)	4 (16%)
Chronic nonspecific lung diseases	3 (6.7%)	1 (3.3%)	4 (10%)	2 (8%)
Peptic ulcer of the stomach or duodenum	2 (4.4%)	2 (6.7%)	2 (5%)	1 (4%)

disease stage (35 men and 10 women in group IA, 67.5 ± 7.9 years; 20 men and 10 women in group IB, 63.3 ± 7.8 years; 18 men and 22 women in group IIA, 69.6 ± 7 years; 14 men and 11 women in group IIB, 67.8 ± 6.8 years).

The distribution of patients within groups depending on the disease stage is as follows. In group IA, 15 patients had stage III and 30 had stage IV. Group IB included 9 patients with stage III chronic ischemia and 21 with stage IV. In group IIA, 12 patients had stage III and 28 had stage IV. Group IIB included 7 patients with stage III chronic ischemia and 18 with stage IV.

Stage III chronic lower-limb ischemia according to the classification of A.V. Pokrovsky-Fontein was established in 43 patients, and stage IV was revealed in 97 patients.

According to the degree of pathology of the vascular bed, the predominant lesion of the arteries of the aortoiliac segment was registered in 12 patients. The most common was the femoral–popliteal level of occlusion (n = 85). Distal occlusion was detected in 24 patients, multifocal lesion was noted in 19 patients, except for the iliac segment, and patients had similar characteristics for other levels of the lesion (Table 1).

Concomitant type 2 diabetes mellitus was diagnosed in 100% of the patients, whereas tablet correction was used for 43.6% of them, and 56.4% of the patients received combination therapy with subcutaneous insulin (short or long-acting). The mean duration of diabetes mellitus was 3.9 ± 1.5 years in group IA, 4.1 ± 1.9 years in group IB, 4.2 ± 2.2 years in group IIA, and 4.4 ± 2.7 years in group IIB. The distribution according to the method of hyperglycemia correction in all groups was comparable.

The most common comorbidities for all four groups were cardiac diseases, namely, ischemic heart disease (57.9%) and hypertensive disease (72.1%) (Table 2). This, of course, can be attributed to prognostically unfavorable signs in relation to the risk of infarction and other vascular events; however, the influence of concomitant diseases on the prognosis of the course of the atherosclerotic process was not evaluated in this study.

During the initial hospitalization, patients in groups IA and IB underwent open or endovascular surgery on lower-limb arteries, depending on the treatment approach accepted. The scope of open surgical interventions (n = 25) included bypass surgeries (15 autovenous bypasses and 5 bypasses using a synthetic prosthesis) and endarterectomy with angioplasty (5 surgeries). Hybrid surgical interventions were performed in 20 cases (6 stentings of the iliac arteries, followed by bypassing of the femoropopliteal segment, and 14 bypassing surgeries followed by balloon angioplasty of the distal sections). Another 35 patients were treated endovascularly, and they underwent balloon angioplasty of the arteries of the femoral–popliteal–tibial segment. Bare metal stents and balloons without drug coating were used in all cases. Groups with surgical approach were comparable in terms of the method of surgical treatment.

In the postoperative period, group IB underwent injection induction of angiogenesis on days 7 and 21 after surgery. After making a decision

Table 3. Summary data on painless walking distance in all patients.

Group		Baseline	After 6 months	Intra-group value p
IA	Number of cases	45	29	$p_{0–6\ mon} = 0.001$
	Mean value	4.6 m	151.7 m	
IB	Number of cases	30	29	$p_{0–6\ mon} = 0.001$
	Mean value	8.2 m	132.1 m	
Intra-group value $p_{IA–IB}$		0.625	0.240	–
IIA	Number of cases	40	20	$p_{0–6\ mon} = 0.001$
	Mean value	0 m	48.5 m	
IIB	Number of cases	25	19	$p_{0–6\ mon} = 0.001$
	Mean value	0 m	72.9 m	
Intra-group value $p_{IIA–IIB}$		1.000	0.032	—

about the non-reconstructability of the vascular bed, groups IIA and IIB (conservative approach) underwent a course of conservative therapy, which included lipid-lowering drugs and an individually selected dosage, antiplatelet therapy (acetylsalicylic acid 100 mg/day), angioprotective therapy (sulodexide at a dose of 600 lipophilic units per day, followed by a transition to 250 lipophilic units 2 times a day; deproteinized hemoderivate 2000 mg/day), metabolic drugs (α-lipoic acid preparations 600 mg/day), and prostaglandin group drugs (prostaglandin E1 and prostacyclin I2 at a dose of 50–200 µg once a day). Moreover, injection stimulation of angiogenesis was performed in group IIB.

All patients included in the study underwent modification of risk factors within 5 years of follow-up, namely, smoking cessation, selection of a lipid-lowering diet, correction of carbohydrate intake, lifestyle modification (physical activity), and diet No. 9.

Patients were randomly distributed to the surgical or therapeutic group (using a random sequence), whereas patients were comparable in terms of disease stage, nature of the lesion, and method of revascularization. Subsequently, patients were followed up for 5 years; received continuous maintenance vascular therapy, correction of risk factors, and repeated courses of infusion therapy every 6 months; and made annual follow-up visits. The active follow-up period with the assessment of instrumental parameters was 6 months, followed by an assessment of the primary points of efficiency (Fig. 1).

Mortality rates and number of amputations performed on the side of the ischemic limb were taken as the primary efficacy criteria. Secondary efficacy factors were instrumental indicators of the degree of circulatory compensation, which were assessed in detail during the first 6 months of patient follow-up.

Data obtained were evaluated using descriptive statistics. Parametric and nonparametric criteria were used. For each group, the correspondence to normal distribution using the Shapiro–Wilk test was analyzed. Analysis of quantitative data during the follow-up period with a normal distribution was performed by comparing paired Wilcoxon cases. For intergroup comparison, the nonparametric Mann–Whitney *U* test was used. Qualitative data were analyzed by constructing 2 × 2 contingency tables and using the χ^2 method with Yates correction and Fisher's test.

Results

During the 5-year evaluation of the treatment results of the patients, no serious adverse reactions were observed after angiogenic therapy, as well as local complications immediately after the drug administration. Injections into ischemic tissues were performed without anesthesia. They caused minor pain during the procedure itself, which stopped immediately after the injection. Laboratory methods were used to all patients over time for studying blood parameters, whereas were no significant fluctuations were noted in the general clinical parameters. When patients underwent repeated annual examinations, no findings of neoplasms were noted.

The painless walking distance was assessed during the first 6 months of follow-up for all patients (Table 3).

The data analysis revealed that the painless walking distance increased significantly in all patients, with limb preservation for 6 months (patients with amputation were excluded from the assessment of this criterion).

The largest increase occurred in groups after surgery, whereas additional angiogenesis in group IB did not affect this indicator, which was due to the satisfactory results of revascularizations and,

Table 4. Summary results of treatment of patients in terms of limb preservation and survival during 5 years of follow-up.

Indicator	Lethal outcomes						Number of amputations					
Follow-up period	6 months	1 year	2 years	3 years	4 years	5 years	6 months	1 year	2 years	3 years	4 years	5 years
Group IA, surgery (*n* = 45)	5 (11.1%)	8 (17.7%)	13 (28.9%)	16 (35.6%)	20 (44.4%)	22 (48.9%)	13 (28.8%)	17 (37.8%)	23 (51.1%)	26 (57.8%)	27 (60%)	28 (62.2%)
Group IB, surgery + angiogenesis (*n* = 30)	1 (3.3%)	4 (13.3%)	7 (23.3%)	9 (30%)	12 (40%)	14 (46.7%)	3 (10%)	5 (16.7%)	9 (30%)	12 (40%)	15 (50%)	16 (53.3%)
$p_{IA–IB}$	0.223	0.430	0.510	0.610	0.703	0.850	0.032*	0.041*	0.045*	0.058	0.393	0.443
Group IIA, conservative therapy (*n* = 40)	6 (15%)	10 (25%)	19 (47.5%)	27 (67.5%)	30 (75%)	32 (80%)	17 (42.5%)	24 (60%)	30 (75%)	35 (87.5%)	35 (87.5%)	35 (87.5%)
Group IIB, conservative therapy + angiogenesis (*n* = 25)	2 (8%)	4 (16%)	8 (32%)	10 (40%)	13 (52%)	16 (64%)	5 (20%)	9 (36%)	14 (56%)	16 (64%)	18 (72%)	18 (72%)
$p_{IIA–IIB}$	0.165	0.296	0.358	0.420	0.057	0.153	0.041*	0.0496*	0.050	0.061	0.117	0.117

Note: *Significant difference.

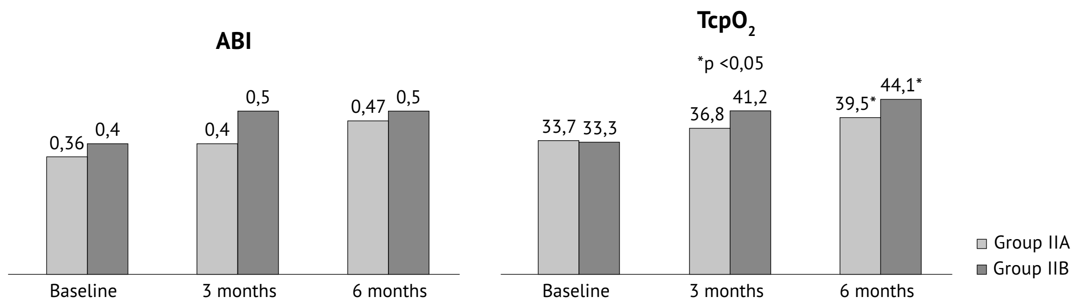

Fig. 2. Dynamics of the ankle-brachial index (ABI) and transcutaneous oxygen tension (TcpO2).

as a result, a rapid increase in painless walking distance in both groups by more than 10 times.

However, in the conservative groups, after 6 months of follow-up with additional angiogenic induction, a significantly better painless walking distance (72.9 m versus 48.5 m, $p = 0.032$) was registered, which indicates a better development of the microvasculature in patients in the absence of restoration of blood flow along the main artery.

We also analyzed the ankle-brachial index (ABI), transcutaneous oxygen tension, and linear blood flow velocity during ultrasound duplex scanning. These instrumental parameters after revascularization, while maintaining the patency of the reconstruction area for the next 6 months, for obvious reasons, were significantly better than those upon inclusion in the study; therefore, their separate analysis is not of interest.

Moreover, in the conservative group, we analyzed separately the parameters of the ABI and transcutaneous oxygen tension for 6 months. The change in the ABI over time for groups IIA and IIB was comparable at baseline (group IIA, 0.36; group IIB, 0.4), after 3 and 6 months (0.36–0.4–0.47 by month 6 for group IIA, and 0.4–0.5–0.5 for group IIB), and the increase was 0.1–0.11 for both groups. Moreover, no significant differences were found between them (0.47 in group IIA and 0.5 in group IIB, $p = 0.569$ by month 6 of follow-up), which indicates the absence of changes in pressure in the main arteries in the lower limbs (Fig. 2).

In the analysis of transcutaneous oxygen tension in group IIA, the value was 33.7–36.8–39.5 mm Hg over time by month 6 of follow-up with a total increase of 17.2%. For group IIB, this indicator was 33.3–41.2–44.1 mm Hg with an increase of 32.4% by 6 months of follow-up. In an intergroup comparison from initially comparable groups, by month 6 of follow-up, a significant difference was obtained in favor of group IIB ($p = 0.028$), which indicates better oxygen saturation of tissues with additional angiogenic induction of angiogenesis (Fig. 2).

The increase in transcutaneous oxygen tension and ABI in these groups was attributed to the poor treatment results in both groups with progression of ischemia underwent amputation of the limb, and they were *ipso facto* excluded from this analysis. Thus, the graphs analyzed the data of those patients who showed improvement during conservative treatment.

The 5-year survival rates of patients with this pathology, regardless of the chosen treatment approach, were comparable with the outcomes of the course of various malignant neoplasms (46.7–80% of lethal outcomes) (Table 4) [7].

The best results in terms of primary efficacy points were obtained in group IB of direct revascularization with angiogenesis (46.7% mortality, 53.3% amputations), and the intended tendency appeared starting from month 6 of follow-up and persisted throughout the study period (5 years). The worst results of survival and limb preservation were obtained in group IIA of standard conservative therapy (80% of lethal outcomes, 87.5% of amputations by year 5 of follow-up; Table 4).

Stimulation of angiogenesis within a complex therapeutic approach by year 5 of follow-up also reduced the frequency of amputations (87.5% in group IIA vs. 72% in group IIB), which led to fewer lethal outcomes in this group (64% in group IIB vs. 80% in group IIA). In general, revascularization surgery (groups IA and IB) showed better results than conservative therapy in terms of primary points of efficiency (limb preservation, survival, and circulatory compensation), regardless of the presence or absence of additional angiogenic stimulations (Table 4).

Additional gene therapy induction in the postoperative period reduced the frequency of amputations in the long-term period (62.2% in group IA and 53.3% in group IB by year 5 of follow-up), but had almost no effect on the mortality rate (48.9% in group IA and 46.7% in group IB after 5 years). As a result of the study, significantly better results were obtained in an intergroup comparison in both surgical (51.1% vs. 30%, $p = 0.045$) and conservative (75% vs. 56%, $p = 0.050$) groups at terms from 6 months to 2 years with additional induction of angiogenesis according to the limb preservation parameter. Moreover, no significant difference was found between the mortality of patients (28.9% vs. 23.3%, $p = 0.510$ in surgery groups, 47.5% vs. 32%, $p = 0.358$ in the conservative group).

In the intergroup comparison of surgical and conservative approaches at all follow-up periods, the best treatment results were obtained in the surgical intervention groups; however, we did not separate this statistical sample because of the impracticality of comparing the surgical and conservative approaches separately.

Discussion

In view of the results, at the present stage, despite the multitudes of treatment approaches to patients with critical ischemia and diabetic angiopathy of the lower limbs, none of them is optimal and does not allow achieving satisfactory treatment results; therefore, the additional use of angiogenic technologies as a postoperative induction or as part of complex therapy can become an additional lever that enables further improvement of results. The administration of therapeutic agents that improve peripheral microcirculation appears promising in the absence of urgent indications for amputation in the patient, possibility of pain control, care of trophic altered tissues, and correction of risk factors.

Patients with diabetes-associated diabetes mellitus are the most difficult group to manage in terms of prognosis. The treatment outcomes of such patients in the long-term period (up to 5 years) are comparable with those of patients with malignant neoplasms and can be poor [7].

Undoubtedly, methods for limb revascularization should be revealed for all patients, as it is the most effective treatment method and helps stop ischemia quickly [7]; however, in the long-term, due to the disease progression and systemic vascular damage by the atherosclerotic process, the frequency of the limb preservation and saving lives are desirable.

Considering the obtained data, the absence of an adequate peripheral channel is a prognostically unfavorable indicator, which may indicate a more severe degree of damage not only to lower-limb arteries, but also to other vascular systems, and a specific painless clinical presentation of disease course due to diabetic neuropathy leads to patients seeking help later and, as a result, worse treatment outcomes.

Conclusions

1. The optimal approach for managing patients with diabetes-associated CLL is direct revascularization intervention on lower-limb arteries with additional angiogenic therapy in the postoperative period (46.7% limb preservation by year 5 of follow-up).

2. The best parameters of limb preservation and the lowest mortality of patients were obtained in the group with additional angiogenic therapy after revascularization. Moreover, gene therapy induction improved significantly the disease course

in terms of limb preservation from 6 months to 2 years (30% of amputations by year 2 of follow-up, $p = 0.045$).

3. Angiogenic induction of angiogenesis also improved the treatment results in the conservative therapy groups; however, for both surgical groups, the best parameters of limb preservation were obtained.

Author contributions. R.E.K. was the work supervisor. I.A.S. created the concept and design of the study and edited the text. A.A.K. performed data analysis, conducted surgical treatment and diagnostic studies, and wrote the text. N.D.M. collected and processed the materials and reviewed the literature. A.S.P. and V.V.K. performed surgical treatment and diagnostic studies. S.A.V. conducted diagnostic studies. N.A.S. analyzed the data obtained and wrote the text.

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Conflict of interest. The authors declare no conflict of interest.

REFERENCES

1. Prevalence, epidemiology, statistics of peripheral arterial disease. *Surgeryzone Medical Site*. http://surgeryzone.net/info/info-hirurgia/rasprostranennost-epidemiologiya-statistika-zabolevanijperifericheskix-arterij.html (access date: 13.09.2021). (In Russ.)

2. Yang SL, Zhu LY, Han R, Sun L, Li J, Dou J. Pathophysiology of peripheral arterial disease in diabetes mellitus. *J Diabetes*. 2017;9(2):133–140. DOI: 10.1111/1753-0407.12474.

3. Strelnikova EA, Trushkina PYu, Surov IYu, Korotkova NV, Mzhavanadze ND, Deev RV. Endothelium *in vivo* and *in vitro*. Part 1: histogenesis, structure, cytophysiology and key markers. *Nauka Molodykh (Eruditio Juvenium)*. 2019;7(3):450–465. (In Russ.) DOI: 10.23888/HMJ201973450-465.

4. Kalinin RE, Suchkov IA, Klimentova EA, Egorov AA, Povarov VO. Apoptosis in vascular pathology: present and future. *Rossiyskiy mediko-biologicheskiy vestnik imeni akademika IP Pavlova*. 2020;28(1):79–87. (In Russ.) DOI: 10.23888/PAVLOVJ202028179-87.

5. Kalinin RE, Pshennikov AS, Suchkov IA. Reperfusion injury of tissues in lower limb arterial reconstructive surgery. *Novosti Khirurgii*. 2015;23(3):348–352. (In Russ.) DOI: 10.18484/2305-0047.2015.3.348.

6. Veerman KJ, Venegas-Pino DE, Shi Y, Khan MI, Gerstein H, Werstuck GH. Hyperglycaemia is associated with impaired vasa vasorum neovascularization and accelerated atherosclerosis in apolipoprotein-E deficient mice. *Atherosclerosis*. 2013;227(2):250–258. DOI: 10.1016/j.atherosclerosis.2013.01.018.

7. Hamasaki H, Moriyama S, Yanai H. A crosstalk between macroangiopathy and microangiopathy in type 2 diabetes. *Int J Cardiol*. 2013;168(1):550–551. DOI: 10.1016/j.ijcard.2013.01.199.

8. *National guidelines on the diagnosis and treatment of diseases of the arteries of the lower extremities, 2019*. Russian conciliation document. http://www.angiolsurgery.org/library/recommendations/2019/recommendations_LLA_2019.pdf (access date: 13.09.2021) (In Russ.)

9. Global vascular guidelines on the management of chronic limb-threatening ischemia. *J Vasc Surg*. 2019;58(1):1–109. DOI: 10.1016/j.ejvs.2019.05.006.

10. Forsythe RO, Hinchliffe RJ. Management of critical limb ischemia in the patient with diabetes. *Journal of Cardiovascular Surgery*. 2016;57(2):273–281.

11. Troitsky AB, Lysenko ER, Khabazov RI, Orekhov PYu, Parshin PYu, Korolev VI, Ustyantseva NV, Malyutina ED, Nishchenko AV. Results of reconstructive operations in patients with lesions of the leg arteries. *Angiologiya i sosudistaya khirurgiya*. 2003;(1):102–108. (In Russ.)

12. Dibirov MD, Dibirov AA, Gadzhimuradov RU, Tereshchenko SA, Soboleva SR, Ramazanova YuI, Bazhenov MS. Distal reconstructions under critical lower limb ischemia in elderly patients. *Khirurgiya. Zhurnal imeni NI Pirogova*. 2009;(1):49–52. (In Russ.)

13. Adam DJ, Beard JD, Cleveland T, Bell J, Bradbury AW, Forbes JF, Fowkes FGR, Gillepsie I, Ruckley CV, Raab G, Storkey H. Bypass versus angioplasty in severe ischaemia of the leg (BASIL): multi-centre, randomized controlled trial. *Lancet*. 2005;366:1925–1934. DOI: 10.1016/S0140-6736(05)67704-5.

14. Dorros G, Jaff MR, Dorros AM, Mathiak LM, He T. Tibioperoneal (outflow lesion) angioplasty can be as primary treatment in 235 patients with critical limb ischemia: five-year follow-up. *Circulation*. 2001;104:2057–2062. DOI: 10.1161/hc4201.097943.

15. Kalinin RE, Suchkov IA, Deev RV, Mzhavanadze ND, Krylov AA. Gene-mediated induction of angiogenesis in inoperable patients with atherosclerosis and diabetes mellitus. *Angiologiya i sosudistaya khirurgiya*. 2018;24(2):33–40. (In Russ.)

16. Berlanga-Acosta J. Diabetic lower extremity wounds: the rationale for growth factors-based infiltration treatment. *Int Wound J*. 2011;8(6):612–620. DOI: 10.1111/j.1742-481X.2011.00840.x.

17. Deev RV, Kalinin RE, Chervyakov YV, Gryaznov SG, Mzhavanadze ND, Kiselev SL, Isaev AA, Schwalb PG, Staroverov IN, Nersessian EG. Results of gen-therapeutic drug "Neovasculgen" in patients with chronic lower limb ischemia: 1 year of observation. *Bulletin of Pirogov National Medical & Surgical Center*. 2011;6(4):20–25. (In Russ.)

18. Deev RV, Bozo IYa, Mzhavanadze ND, Nersesyan EG, Chukhralia OV, Shvalb PG, Chervyakov YuV, Staroverov IN, Kalinin RE, Voronov DA, Gavrilenko AV, Isaev AA. Efficacy of using VEGF165 gene in comprehensive treatment of patients with stage 2A–3 lower limb chronic ischaemia. *Angiologiya i sosudistaya khirurgiya*. 2014;20(2):38–48. (In Russ.)

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