

ПРАКТИКУЮЧОМУ ЕНДОКРИНОЛОГУ

**THE USE OF 5-AMINOLEVULINIC ACID GEL
IN THE TREATMENT OF DIABETIC FOOT SYNDROME***

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The last 30 years have been characterized by a dramatic increase in the prevalence and incidence of diabetes mellitus (DM), mainly in industrialized countries, where the disease accounts for 2.3–9 % of the population [1, 2]. Diabetes ranks third in the morbidity structure after cardiovascular disease and cancer [3]. According to the International Diabetes Federation, there are currently 415 million people with diabetes worldwide [2, 4], but the actual number of patients with undiagnosed pathology is 3–4 times higher [5]. According to an expert evaluation of the prevalence of disease, the number of people with diabetes has a tendency to increase and may reach 642 million by 2040 [2].

Similar trends are taking place in Ukraine, where, according to the International Diabetes Federation, in 2013, there were 1.04 million

(2.99 %) patients with diabetes [2]. The incidence and prevalence of diabetes in Ukraine has doubled in 10 years. In reality, however, the number of patients is 2–3 times higher than the data of the Center for Medical Statistics of the Ministry of Health of Ukraine suggest, which is due to hidden forms of diabetes [6].

DM affects almost all body systems, but the most dramatic complications occur in the lower extremities. Diabetic foot syndrome (DFS) takes the leading position in the list of complications of DM, leading to early disability and mortality [7, 8]. DFS complicates the course of DM in almost 25 % of patients [9].

Chronic ulcers, infected wounds and previous surgery have been found to be predictors of foot suppuration (Diabetic Foot Study Group, 2015). In the etiology of purulent necrotic processes in patients with DFS, a mixed aerobic-

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anaerobic microflora with fungal infection is of great importance, which significantly complicates rational antibiotic therapy [10, 11].

The composition of pathogenic flora in the trophic ulcer area depends on many factors: the depth of the ulcerous defect, the duration of the ulcer, and the presence of prior antibiotic therapy. Analysis of the results of bacteriological studies revealed that in patients with DFS, a mixed aerobic-anaerobic infection was present in 87.7 % of cases, aerobic infection alone in 12.3 %, and anaerobic infection in 7.8 % [12]. The microbial associations include 2 to 14 species of aerobic, facultative-aerobic and obligate-anaerobic nonsporulating bacteria. Also, patients with DFS on admission are characterized by a high microbial contamination of the tissue of the purulonecrotic foci [13]. Gram-positive cocci (*S. aureus*, *S. pyogenes*) predominate in first-onset superficial tissue defects [14]; in deep, long-term ulcers, the polymicrobial nature of the infection process is more common [15]. It should be noted that in most cases, resistant flora is identified.

The aim of surgical treatment in DFS is to preserve the life of the patient and preserving the limb and its function. Surgical intervention should be timely, consistent with the principle of reasonable sufficiency (tissue-sparing, maximum preservation of foot function), and should be performed while stabilizing the patient's general condition, unloading the affected limb, correcting carbohydrate metabolism disorders, and providing antibacterial and pathogenetic therapy [16, 17].

Clinical observations illustrate the possibility of preserving limb weight-bearing with correctly chosen tactics of complex surgical treatment in patients with ischemic and mixed forms of DFS with purulent-necrotic tissue changes, despite the variety of medical problems that the specialist has to face (purulent-necrotic plastic material, postoperative orthopedic features) [18, 19].

In terms of improving the treatment outcomes of these patients, the existing standard surgical techniques are almost exhausted, which determines the search for new approaches in solving this problem.

Despite the large arsenal of such remedies, the problem of treatment of purulent and long-

term non-healing wounds is far from being resolved. In recent years, physical and chemical wound treatment methods have begun to be used both separately and in combination to address this problem. Phototherapy and photodynamic therapy (PDT) seem to be promising directions in treating purulent and long-term non-healing wounds.

At present, a new technology — PDT — is being intensively developed worldwide. This technique can be used in many different fields of medicine, including purulent surgery. PDT has undeniable advantages over conventional antibiotic therapy. The effectiveness of PDT does not depend on the antibiotic sensitivity spectrum of pathogens [20]. Pathogens, unlike antibiotic exposure, do not develop resistance to PDT. Photodynamic damage is local, and the bactericidal effect is limited by the laser irradiation zone of the photosensitized tissue, which avoids the local PDT side effect observed when using antibiotics and antiseptics to treat surgical infection. The effectiveness of PDT treatment was found to vary depending on the bacterial flora isolated from the wound contents. The need to study the effects of PDT on the course of the wound process caused by different groups of bacterial flora is evident to all researchers in this field.

The mechanism of PDT is based on the basic principles of photobiology. Bunsen-Roscoe law: the photochemical effect is determined by the dose of initiating radiation and is calculated as the product of the power of light flux on the time of exposure. Stark-Einstein law: one molecule of a substance participating in a photochemical reaction absorbs one light quantum. The Grotthus-Drawper law: the radiation used to start the photochemical reaction must have a wave of the appropriate length because only the absorbed light is able to initiate its course [21].

The chemical structure of modern photosensitizers (PS) is based on porphyrin, a macrocyclic compound consisting of four pyrrole rings linked by methine bridges. The general idea of the development of PDT therapy is in the guidelines for photosensitizers in clinical PDT which outline relevant points that need to be considered. Among them, the essential requirements that a product must fulfil are: harmlessness for the patient and physician; high selectivity of

accumulation in tumor, microbial and damaged cells; high quantum yield of the triplet state in vivo (with an energy of at least 94 kJ/mol); maximum absorption in the 600–800 nm wavelength range; pronounced luminescence; weak accumulation in healthy skin and mucous membranes low light toxicity when used in therapeutic doses; rapid elimination from the body; good solubility in water or other solvents used for intravenous administration; stability during storage and administration; availability of production or synthesis [22].

A special place among photosensitizers is occupied by 5-aminolevulinic acid (5-ALA), a natural precursor of the endogenous photosensitizer protoporphyrin IX (PP-IX). 5-ALA itself is not a photosensitizer and does not accumulate in cells, but when administered exogenously, it temporarily «overloads» the normal haem biosynthesis pathway. Due to reduced activity of ferro chelatase, an enzyme that limits the rate of this process, and iron deficiency, an intermediate product of biosynthesis, PP-IX accumulates in tumor tissue, unlike in normal tissue. PP-IX is a relatively active PS due to the presence of an intense absorption band with a maximum at 630 nm and the ability to generate singlet oxygen efficiently. Moreover, due to the selective accumulation of PP-IX, specifically in malignant cells, ALA is an effective fluorescent tumor marker [23].

Drug products based on 5-ALA are successfully used to treat keratosis, basal cell cancer, diagnosis and treatment of Barrett's esophagoma, bladder cancer, brain tumors, gastric tumors, and cosmetic practice. Particularly effective is the use of 5-ALA-based preparations for endoscopic diagnosis of tumors, the visual detection of which is difficult under normal white light [23, 24].

MATERIALS AND METHODS

10 patients with type 2 DM and ischemic and mixed forms of diabetic foot syndrome of both sexes aged 52–75 yrs, who were treated in the department of acute vascular pathology of SI «V. T. Zaitsev Institute of General and Emergency Surgery of NAMS of Ukraine», were under observation in 2021. Inclusion criteria were: the presence of type 2 DM and infringuinal lesions of the great arteries of the

For therapeutic purposes, 5-ALA is applied topically or administered systemically and penetrates indiscriminately into all cells. It is metabolized into the active sensitizer PP-IX using the haem biosynthesis pathway. As malignant or neoplastic (fast-growing) cells usually have lower intracellular iron (Fe^{2+}) levels than normal healthy cells, PP-IX cannot be rapidly converted into its final product haem and therefore accumulates only in malignant cells. Once the photosensitizer selectively accumulates in the tumor, it can be activated by illumination with light of an appropriate wavelength and dose. The PP-IX molecule absorbs light, and then the absorbed energy is transferred to molecular oxygen, creating the singlet oxygen state $^1\text{O}_2$. The singlet oxygen, which is produced by the PDT process on PP-IX, leads to oxidative damage of intracellular macromolecules and hence, cell death. This cytotoxic effect realizes, particularly on proliferating cells, due to singlet oxygen reactions, e.g., with the cell membrane and mitochondria or triggers the formation of free radicals that damage cells. A variant of PDT-induced cell death is usually a mixture of apoptosis, necrosis and autophagy. It is generally accepted that in addition to direct cellular cytotoxicity, two other important factors contribute to the overall effect of PDT: vascular occlusion (which stops angiogenesis and thus prevents metastasis) and the local inflammatory response [25, 26]

This study aimed to investigate the possibility of using 5-aminolevulinic acid as a photosensitizer in photodynamic therapy in the complex treatment of chronic wounds in patients with ischemic and mixed forms of diabetic foot syndrome.

limbs and the possibility for revascularization. The most common comorbid conditions were hypertension (85.2%), overweight/obesity (76.4%), hyperlipidemia (75.2%), chronic kidney disease (18.1%) and cardiovascular disease (28.6%).

The patients were treated according to the following scheme: DM compensation (switching to insulin therapy), metabolic therapy, anticoagulant and angiotropic therapy, physical thera-

py, and therapy aimed at treating osteoporosis (calcium preparations).

Analysis of clinical, laboratory, non-invasive and invasive methods of examining the patients made it possible to determine the degree of impairment of the main blood flow, the nature of the collateral circulation and microcirculation. The diagnosis was performed for all patients according to a standard algorithm: history, clinical and laboratory examination: duration of the disease, intermittent claudication, pain at rest, presence and nature of necrosis, pulsation of the main arteries, analysis of laboratory tests and history of co-morbidities. Non-invasive examination included: determination of regional systolic pressure index in the arteries of the foot using a portable ultrasound machine «Super Dopplex» (China); ultrasonic dopplerography using a machine «Hitachi EUB 7500» (Japan) with L 5–10 MHz linear transducer; Transcutaneous tensions of oxygen (TcO₂) in foot tissues percutaneous using TCM 400 device produced by Radiometer Copenhagen (Denmark); invasive examinations: Seldinger angiography using the Philips Integris Allura 9 (Holland).

In the postoperative period control, regional systolic pressure index determination in the foot arteries, ultrasound examination of the arterial reconstruction zone, and TcO₂ determination in the foot tissues were performed. X-rays of the bones of the foot and tibia (if necessary) were also performed. The degree of ischemia was assessed using the GLASS system (2014).

A prognostic criterion for wound healing is the transcutaneous oxygen (O₂) tension in the tissue, which was determined using a TCM-400oximonitor (Radiometer, Denmark). In the

studied patients, this index before revascularization was below 30 mmHg.

On admission, all patients underwent microbiological sampling. Wound exudate was microbiologically examined, and the quantitative and qualitative composition of the microflora, as well as their sensitivity to antibiotics, was determined. Identification of the isolated strains was performed using standard bacteriological methods (morphological, tinctural, cultural, biochemical properties, and pathogenicity), following the Bergi classification (2011). Antibiotic sensitivity of bacteria was assessed in accordance with ICC 4.2 1890-04.

The severity of pain sensations was evaluated during treatment (visual analogue scale). The rate of marginal epithelialization was determined by the formula of A.N. Povova (1945).

In the clinical assessment of the wound process, we considered the terms of edema disappearance, the appearance of single granulations, the wound filling with the mature granulation tissue, the beginning of epithelialization and full epithelialization. For this purpose, the wound dynamics assessment system, presented in Table 1, was used.

We developed and applied the following technique to treat ischemic and mixed forms of DFS. As a preparation for PDT, daily dressings with hydrogel were applied (3–5 days). During this period, limb revascularization was performed, the aim of which was to restore the blood flow to the foot, preferably through the anterior or posterior tibial artery. It should be noted that endovascular interventions were performed in all studied patients. The presence of blood flow in the foot and the localization of necrosis determined the choice of the artery for recanalization. The main procedure

Table 1

Treatment outcome assessment system

Assessment criteria	Treatment outcomes (days)		
	Good	Satisfactory	Unsatisfactory
Disappearance of perifocal oedema	< 4	5–8	> 9
Appearance of primary granulation tissue	< 6	7–10	> 10
Filling the wounds with mature granulation tissue	< 10	11–16	> 16
Appearance of marginal epithelialization	< 11	12–17	> 17
Total epithelialization	< 30	31–35	> 35

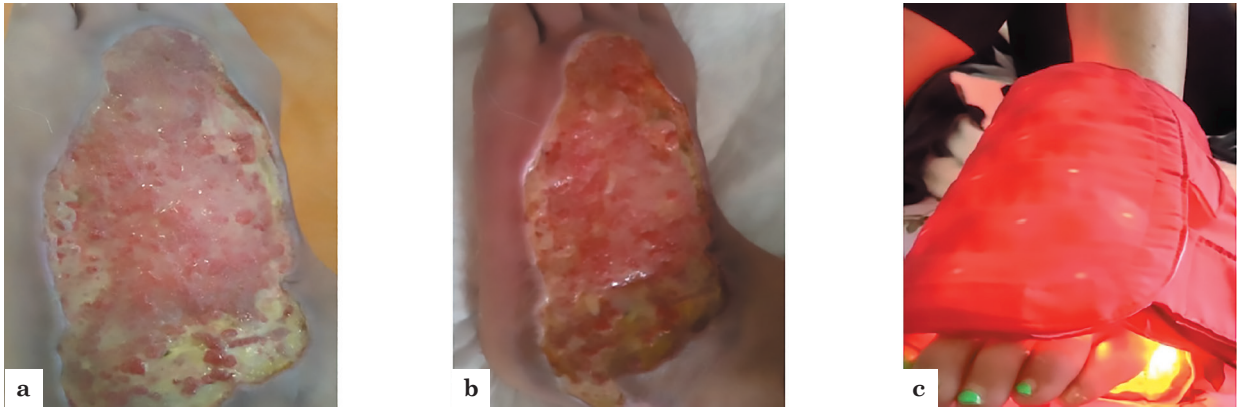


Fig. 1. Patient B., 58 years old. Type 2 diabetes mellitus, extensive chronic wound of the left foot: a) wound on admission; b) wound with PS; c) wound irradiation.

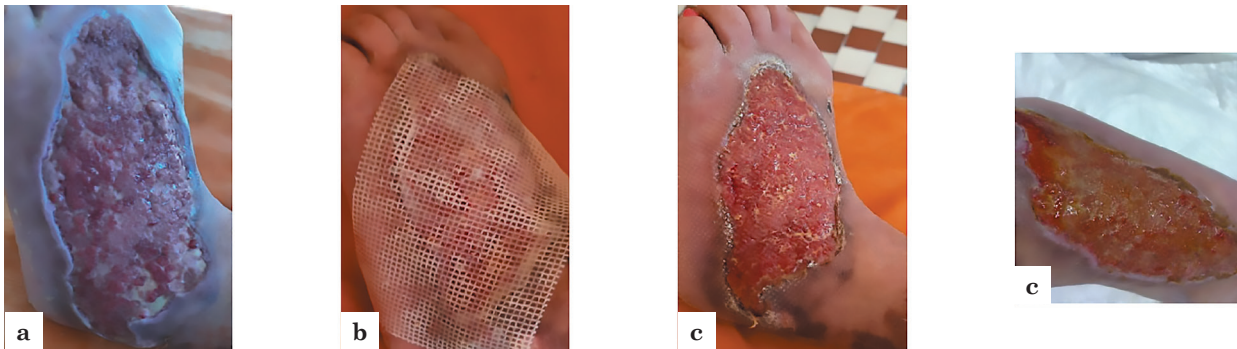


Fig. 2. Patient B., 58 years old. Type 2 diabetes mellitus, extensive chronic wound of the left foot: a) wound on the 3rd day after PDT; b) the wound was covered by human amniotic membrane; c) wound on the 5th and 10th day after irradiation.

was percutaneous balloon angioplasty. In case of residual hemodynamically significant stenosis or obstructive dissection, nitinol self-eluting stents were implanted in the femoro-popliteal segment, and balloon-eluting coronary stents in the tibial artery (2 observations).

After the restoration of the main blood flow, PDT was performed: after sanitation of the wound surface with physiological NaCl solution, Levuderm gel (6 % gel phosphate of 5-aminolaevulinic acid) was applied and covered with occlusive dressing for 2 hours, after which the gel remains were removed and irradiation with Korobov photonic matrices «Barva Flex» with 660 nm wave length was carried out. Energy applied to the wound averaged 30–40 J/cm². The light power density was between 0.1–1.0 W/cm², and the exposure time varied according to the wound area (Fig. 1).

After that, we switched to phototherapy (irradiation of wounds with red light (λ 660–630 nm) daily, the duration of sessions was 10–15 min, the number of sessions depended on the wound area) until conditions for plastic closure of de-

fects or healing by wound contraction appeared. For plastic closure, various wound coverings (as an alternative to autodermoplasty) — lyophilized xenografts, synthetic polycaprolactone and polylactide wound coverings and human amniotic membrane — were used (Fig. 2).

In cases of manifestation of infection in the wounds, PDT was repeated (1 observation). Patients signed an agreement to participate in the study in all observations. The Ethical Committee approved the whole list of the studies and the used treatment methods of the SI «V.T. Zaitsev Institute of General and Emergency Surgery of the National Academy of Medical Sciences of Ukraine».

Statistical tests: unpaired two-tailed student tests were performed using Statistica 8.0, Microsoft Excel software (USA). Differences were considered statistically significant with a p-value < 0.01. Results are expressed as means \pm standard error of the mean. Statistical analysis was performed using the standard software package SPSS for Windows 13.0. The significance level of differences p was assumed to be 0.05.

RESULTS AND THEIR DISCUSSION

After revascularization, all of the patients studied achieved stabilization of their local hemodynamic parameters (Table 2).

On admission, the majority of observations (6) had Gr+ microorganisms: staphylococci (in 4 patients) with an average colonization $(8.46 \pm 0.82) \times 10^9$ colony-forming unit (CFU). Micrococci were second (in 1 patient) with $(7.58 \pm 0.87) \times 10^9$ CFU). Gr- non-fermenting pathogens in monoculture were identified in 3 patients (in 2 cases *Pseudomonas aeruginosa* with an average colonization of 7.42×10^9 CFU and in 1 observation *Acinetobacter baumani* – 7×10^7 CFU). Microbial associations occurred in 2 cases: in one patient *St. aureus* $\times 10^7$ CFU + *Pseudomonas aeruginosa* $\times 10^8$ CFU and in one case *Pr. mirabilis* 5×10^6 CFU + *Kl. pneumonia* 4×10^5 CFU + *E. faecalis* 6×10^6 CFU.

Control cultures in the majority of patients (8) did not show growth, in 2 cases the infectivity level decreased below critical values (*St. aureus* $< 10^3$), in one case, a second session of PDT was needed, which was carried out on the 10th day of treatment.

After PDT on days 2–3, perifocal oedema decreased. By days 5–7, the quality of granulation and the degree of wound epithelialization significantly improved. The wound healing

rate on days 5–7 of treatment was 1.58 ± 0.44 , and on days 10–12 — 4.72 ± 0.63 ($p < 0.01$). Transcutaneous O₂ tension reached 50–80 mm Hg in the studied patients 5–7 days after treatment.

The evaluation of the wound healing rate showed good and satisfactory results: By day 32, the wound in the study group was completely epithelialized in the majority of patients (88.9 %) (Table 3).

In all investigated patients, the treatment period did not exceed 35 days (depending on the wound area and peculiarities of the wound process).

At the same time, no allergic reactions or pronounced pain syndromes were observed during the procedures.

Thus, the method proposed by the author can optimize the wound process, reduce the time of granulation formation and wound filling with mature granulation tissue by 3 and 3–14 days, respectively, and increase the frequency of epithelialization by 29.8 % compared with conventional therapy. The use of PDT in patients by means of the developed method contributed to the rapid (within 2–3 days) clearance of wounds from purulent-necrotic masses (with traditional treatment, the duration of

Indices of local hemodynamics before and after revascularization

Table 2

Index	Terms	
	Before revascularization (n = 10)	After revascularization (n = 10)
Regional Systolic Pressure Index	0.27 ± 0.03	0.67 ± 0.08 $p < 0.05$
TcpO _{2foot} , mmHg	9.4 ± 3.5	39.6 ± 6.7 $p < 0.05$

Assessment of treatment outcome

Table 3

Assessment criteria	Results of treatment (days and % of patients)		
	Good	Satisfactory	Unsatisfactory
Disappearance of perifocal oedema	6 (66.9 %)	3 (22.8 %)	1 (10.3 %)
Appearance of primary granulation tissue	6 (56.6 %)	4 (43.4 %)	–
Filling the wounds with mature granulation tissue	6 (66.7 %)	4 (33.3 %)	–
Appearance of marginal epithelialization	–	9 (88.9 %)	1 (11.1 %)
Total epithelialization	5 (66.9 %)	2 (12.9 %)	3 (20.2 %)

this stage is about 4–5 to 7–10 days). The first granulations appeared on the 4th–5th day, and peripheral epithelization — on the 6th–7th day of treatment. It is known that with traditional treatment, these indices are reached by 7 and 10–11 days respectively. Necrotic masses were already actively sloughed after the first session of PDT. There was almost no purulent exudate from the wound. No neurectomies were required in any of the cases during treatment. Microbiological examination showed that the wounds were decontaminated, which eliminated the need for an antibiotic therapy treatment.

PDT using ALA as a PS was initially used to treat skin cancer and some other cancers. However, it has been shown to be effective in treating some benign skin conditions (common acne, sebaceous hyperplasia and suppurative hidradenitis). Studies have shown that this type of exposure promotes an increase in type I collagen fibers in photodamaged skin. An increase in type I and III procollagen in photodamaged skin after PDT with ALA has also been demonstrated [26].

In addition, a significant increase in procollagen types I and III, collagen precursors were shown, reflecting increased skin collagen synthesis, as well as increased immunoreactivity (TGF- β and T β RII) and decreased matrix metalloproteinase levels, indicating an anti-inflammatory effect of PDT with ALA. On the other hand, the incoherent red-light source itself may, to some extent, influence histological changes in the skin [21, 27].

The final mechanism of the positive effect of PDT on the course of the wound process is far from being finally understood; however, some aspects of this mechanism, in our opinion, can be formulated as follows: The leukocytes play an essential role in regulating the changing stages of the wound process. However, several factors disturb the ordinary course of the wound healing process. The main one in the long-lasting wound is infection. A high level of bacterial contamination leads to an increased concentration of bacterial endotoxins. On the other hand, endotoxins are known to have a biphasic effect on the functional activity of leukocytes: Polymorphonuclear leukocytes and mononuclear cells depending on their concentration. At high concentrations of endotoxins, inhibition

of leukocyte functional activity and destruction of leukocytes is observed. It is also known that the wound healing process occurs through interrelated and controlled stages: inflammation, proliferation, granulation tissue maturation and epithelialization. Microbial antigens, upon entry into the body, are neutralized by complement system proteins C3, C4, C5 and immunoglobulins IgA, IgM, IgG. Antibodies with antigens and complement (AT+AG+ complement) form circulating immune complexes. They can be eliminated from the body or their further processing can be done by phagocytosis. Phagocytosis is a powerful barrier factor, but with chronicity of the process and bacterial expansion, all phagocytosis stages of antigen-presenting cells are affected and cannot be medically corrected.

In this regard, we can assume that the dynamics of the wound process under PDT exposure are as follows: With an initially high level of wound contamination and, as a consequence, a high concentration of endotoxins in the exudate, polymorphonuclear leukocytes are present in small numbers and have a low level of functional activity. During PDT in the presence of PS, there is a lesion of the bacterial cell, a decrease in the level of infection and concentration of endotoxins. Along with this, there is an activation of chemotaxis, adhesion and endocytosis of bacteria by phagocytizing neutrophils due to oxygen-dependent endocytosis and «oxygen explosion» due to generated singlet oxygen. In this case, polymorphonuclear leukocytes are not inhibited; on the contrary, their phagocytic activity is activated, resulting in faster wound cleansing. When microbial antigens enter, granulocytic neutrophils perform their processing — phagocytosis. The processing of microorganism destruction is carried out with the help of lysosomal enzymes inside the phagosome. The hydrolytic enzymes of the phagosome — granzymes, lactoferrins, proteases, nucleases, lipases, phospholipases, phosphatases, sulphurates — are actively involved in this process. Phagocytosis of microorganisms also occurs with the participation of ROS — superoxide anion (O_2^-) with the formation of hydrogen peroxide (H_2O_2), hydrochloric acid (HCl), myeloperoxidases (MPO) and phospholipases A_2 catalyze the reactions of cleava-

ge of substances composing the structure of microorganisms. At the end of this process, the neutrophils are killed. The combined action of singlet oxygen and hydrolytic enzymes causes destruction of the phagocyte membrane by the mechanism of lipid peroxidation and oxidative capacity of phagocytic neutrophils. The cells uptake and oxidize the dye pellets with varying intensity, depending on the reducing ability of the enzymes, under the influence of superoxide anion (O_2^-) formed in the NADPH oxidase reaction.

The change in the stages of the healing process of chronic wounds, the transition of the wound process to the granulation phase, is accompanied by a change in the composition of the leukocyte population in the wound: mononuclear cells, monocytes and macrophages, capable of producing a large number of different cytokines, come to the inflammation focus instead of neutrophils.

Thus, the pathophysiological basis for the use of PDT in the treatment of wounds is a change in the internal energy potentials of the cell when absorbing a quantum of light followed by a response at different levels — from the subcellular to the systemic. The abovementioned events in the wound are caused by the affecting effect of PDT on the microflora. This is due to the high level of accumulation of exogenous PS by bacterial cells and gross initiation of secondary free radical reactions. At the same time, intrinsic cells located in the wound defect are not able to intensively accumulate

significant amounts of PS and bind it in lesser amounts, compared to bacteria. In this case, light exposure is accompanied by cell stimulation. In leukocytes, exposure to PDT in the presence of small amounts of PS can have a direct effect, with two effects: an increase in bactericidal activity and activation of regenerative processes. The high level of enzyme induction by the photosensitizer contributes to the normalization of immunoreactivity. Despite high spontaneous oxidative and low induced phagocyte activity, granulocyte oxidative reserve increases after light exposure.

The increase in bactericidal capacity of leucocytes is due to a photoinduced increase in oxygen radical production by increasing NADPH oxidase activity, while the activation of synthesis leads to the formation of very important, in terms of the events considered, proteins: inducible NO synthetase and cytokines. The formation of inducible NO synthetase is accompanied by an increase in nitric oxide production, which is the factor responsible for the vasodilation of microvessels. All of this leads to the improvement of microcirculation that is observed during PDT exposure in the process of wound healing. Due to these and other factors, PDT in the treatment of long-term non-healing wounds gives the primary stimulus, accelerating the torpid wound process and then consistently shortening the time of all phases of wound healing on the background of activation of the barrier function of phagocytosis.

CONCLUSIONS

Thus, the growth of resistance to antibacterial agents is a significant challenge to their effective clinical use. The difficulty of maintaining a balance between the need for adequate antimicrobial therapy and limiting the growth of resistant flora, necessitates the search for new approaches to the strategy of anti-bacterial use. The method we have developed for treating chronic wounds using photodynamic therapy is pathogenetically feasible and highly effective

compared to conventional therapies. Combined light exposure using photosensitizer stimulates the formation of reactive oxygen species, increasing the effect of phagocytic cells, which is manifested in the activation of neutrophil chemotaxis, adhesion, and endocytosis. Photodynamic therapy using 5-aminolevulinic acid as a photosensitizer, enhances molecular mechanisms of intercellular interaction at all stages of primary immunity activation.

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IN THE TREATMENT OF DIABETIC FOOT SYNDROME

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Background. Diabetes mellitus affects almost all body systems, but the most dramatic complications occur in the lower extremities. Diabetic foot syndrome takes the leading position in the list of complications of diabetes mellitus, leading to early disability and mortality.

At present, a new technology — photodynamic therapy is being intensively developed worldwide. This technique can be used in many different fields of medicine, including purulent surgery.

This study aimed to investigate the possibility of using 5-aminolevulinic acid as a photosensitizer in photodynamic therapy in the complex treatment of chronic wounds in patients with ischemic and mixed forms of diabetic foot syndrome.

Materials and Methods. In 2021, 10 patients with type 2 diabetes mellitus and ischemic and mixed forms of diabetic foot syndrome of both sexes, aged 52 to 75 years, were observed. As a preparation for photodynamic therapy, daily dressings with hydrogel were applied (3–5 days). During this period, limb revascularization was performed, the aim of which was to restore the blood flow to the foot, preferably through the anterior or posterior tibial artery. After the restoration of the main blood flow, photodynamic therapy was performed: after sanitation of the wound surface with physiological NaCl solution, Levuderm gel (6 % gel phosphate of 5-aminolaevulinic acid) was applied and covered with occlusive dressing for 2 hours, after which the gel remains were removed and irradiation with Korobov photonic matrices «Barva Flex» with 660 nm wave length was carried out. Energy applied to the wound averaged 30–40 J/cm². The light power density was between 0.1–1.0 W/cm², and the exposure time varied according to the wound area.

Results. After photodynamic therapy on days 2–3, perifocal oedema decreased. By days 5–7, the quality of granulation and the degree of wound epithelialization significantly improved. The wound healing rate on days 5–7 of treatment was 1.58 ± 0.44 , and on days 10–12 — 4.72 ± 0.63 ($p < 0.01$). The evaluation of the wound healing rate showed good and satisfactory results: By day 32, the wound in the study group was completely epithelialized in the majority of patients (88.9 %).

Conclusions. The method we have developed for treating chronic wounds using photodynamic therapy is pathogenetically feasible and highly effective compared to conventional therapies. Combined light exposure using photosensitizer stimulates the formation of reactive oxygen species, increasing the effect of phagocytic cells, which is manifested in the activation of neutrophil chemotaxis, adhesion, and endocytosis. photodynamic therapy using aminolaevulinic acid as a photosensitizer, enhances molecular mechanisms of intercellular interaction at all stages of primary immunity activation.

Key words: diabetes mellitus, diabetic foot syndrome, photodynamic therapy, photosensitizer aminolaevulinic acid.

ЗАСТОСУВАННЯ 5-АМІНОЛЕВУЛІНОВОЇ КИСЛОТИ У ЛІКУВАННІ СИНДРОМУ ДІАБЕТИЧНОЇ СТОПИ

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Вступ. Цукровий діабет вражає майже всі системи організму, але найбільш серйозні ускладнення виникають у нижніх кінцівках. Синдром діабетичної стопи займає лідируючу позицію в переліку ускладнень ЦД, що призводять до ранньої інвалідизації та смертності.

Зараз у світі інтенсивно розвивається нова технологія — фотодинамічна терапія. Ця методика може використовуватися в самих різних областях медицини, в тому числі в гнійній хірургії.

Метою даного дослідження було вивчити можливість використання 5-амінолевулінової кислоти як фотосенсибілізатора при фотодинамічній терапії у комплексному лікуванні хронічних ран у хворих на ішемічну та змішану форми синдрому діабетичної стопи.

Матеріал та методи. У 2021 році під спостереженням перебувало 10 хворих на цукровий діабет 2 типу та ішемічну і змішану форми синдрому діабетичної стопи обох статей віком від 52 до 75 років. В якості підготовки до фотодинамічної терапії застосовували щоденні пов'язки з гідрогелем (3–5 днів). У цей період проводили ревазуляризацію кінцівки, метою якої було відновлення кровотоку в стопі, переважно по передній або задній великогомілковій артерії. Після відновлення магістрального кровотоку проводили фотодинамічну терапію: після санації поверхні рани фізіологічним розчином NaCl наносили гель Левудерм (6 % гель фосфата 5-амінолевулінової кислоти) і накладали оклюзійну пов'язку на 2 години, після чого видаляли залишки гелю та проводили опромінення фотонними матрицями Коробова «Varva Flex» з довжиною хвилі 660 нм. Енергія, прикладена до рани, становила в середньому 30–40 Дж/см². Щільність потужності світла становила 0,1–1,0 Вт/см², а час експозиції змінювався залежно від площі рани.

Результати. Після фотодинамічної терапії на 2–3 добу зменшився перифокальний набряк. На 5–7 добу якість грануляції та ступінь епітелізації рани значно покращилися. Швидкість загоєння ран на 5–7 добу лікування становила $1,58 \pm 0,44$, а на 10–12 добу — $4,72 \pm 0,63$ ($p < 0,01$). Оцінка швидкості загоєння рани показала добрі та задовільні результати: на 32 добу рана в групі дослідження була повністю епітелізована у більшості пацієнтів (88,9 %).

Висновок. Розроблений нами метод лікування хронічних ран із застосуванням фотодинамічної терапії є патогенетично здійсненним і високоефективним порівняно з традиційною терапією. Комбіноване світлове опромінення з використанням фотосенсибілізатора стимулює утворення активних форм кисню, посилюючи дію фагоцитуючих клітин, що проявляється в активації хемотаксису нейтрофілів, адгезії та ендоцитозу. Фотодинамічна терапія з використанням амінолевулінової кислоти в якості фотосенсибілізатора посилює молекулярні механізми міжклітинної взаємодії на всіх етапах первинної активації імунітету.

Ключові слова: цукровий діабет, синдром діабетичної стопи, фотодинамічна терапія, фотосенсибілізатор амінолевулінова кислота.

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