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Low-temperature argon plasma in the wounds treatment after hemorrhoidectomy

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ABSTRACT *AIM: to improve the results of treatment in hemorrhoid's Grade IV.*
PATIENTS AND METHODS: the prospective randomized study included 101 patients with combined hemorrhoids Grade IV divided in two groups. Both groups were homogenous in age and gender. All patients underwent open hemorrhoidectomy with monopolar coagulation. Low-temperature argon plasma was implemented in postoperative period as an additional option in the main group at 2, 4, 6, 8, 14, 21, 30 days after surgery. Visual Analogue Scale (VAS, 0 to 10 points) was used to assess a pain intensity. Bacteriological and cytological tests performed at 2, 8, 14, 21, 30 days and then every 7 days until the wounds were completely healed. The area of the postoperative wound and the rate of healing were calculated using a planimetric method. Quality of life was assessed before surgery, and on days 8 and 30 using the SF-36 questionnaire.
RESULTS: on the 30th day after surgery, cytology confirmed wound healing occurred in 38 (76.0%) patients of the main group and in 18 (36.0%) patients in the control group, $p = 0.0001$. VAS score at day 8 after surgery was 3 (3; 4) and 4 (3; 5) points in main and control group, respectively, $p < 0.0001$. Quality of life improved and patients showed significant difference in the physical component between groups: 48 (44; 53) vs 42 (38; 48) points in the main and control group, respectively ($p < 0.05$). On the 30th day after the procedure, the physical component of the quality of life was 48 (44; 53) points in the patients of the main group, 42 (38; 48) in — the control group, $p = 0.005$. There was found significant difference in wound microbial content between groups: 104 vs 107 CFU on the 30th day after the surgery.
CONCLUSION: the low-temperature argon plasma accelerates wound healing, as well as reduces the pain intensity. A significant antimicrobial effect was detected.

KEYWORDS: *low-temperature argon plasma, plasma, wound treatment, hemorrhoids*

CONFLICT OF INTEREST: *The authors declare no conflict of interest*

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INTRODUCTION

Recently, the treatment of postoperative and non-healing wounds remains relevant. Hemorrhoids is a common cause of a patient's referral to a coloproctologist, its incidence is 130–145 people per 1,000 adult population [1].In many countries, hemorrhoidectomy remains a radical method of treating hemorrhoids [2,3]. When performing surgery, a postoperative wound is formed, the healing time of which is still long. This is due to the constant contamination of the wound surface with intestinal microflora, which leads to tissue inflammation, pain syndrome, and also reduces the quality of life. In this regard, the relevance of the use of physical methods that accelerate wound healing is increasing today.

According to some studies, low-temperature argon plasma (LTAP) accelerates the healing processes. However, there are no randomized studies of the use of LTAP in the treatment of wounds after anal surgery.

PATIENTS AND METHODS

A randomized prospective study of the use of LTAP in the treatment of patients with Grade IV hemorrhoids after open hemorrhoidectomy was carried out.

The hypothesis of the study: the use of low-temperature argon plasma accelerates the healing process in postoperative wounds.

Inclusion criteria:

- patients with external and internal hemorrhoids of Grade IV;
- open hemorrhoidectomy surgery using monopolar electrocoagulation;
- the age of patients: 18 years and older;
- the patient's consent to the proposed treatment plan.

Criteria of non-inclusion:

- patients with previous anal surgery;
- acute hemorrhoids;
- inflammatory bowel diseases;
- concomitant diseases of the perianal area and anal canal;
- patients with severe concomitant diseases in the decompensation.

Exclusion criteria:

- surgery by any other method;
- refusal of the patient to participate in the study at any stage, including non-compliance with recommendations.

The primary point of the study was the rate of healing of the postoperative wound (the number of patients whose wound healing was noted on day 30). The wound healing period was assessed by visual control and digital examination, confirmed by cytology.

The study was prospective, randomization of patients was carried out 1:1 using the Internet resource RandStuff.ru.

In the study, the classification of hemorrhoids according to clinical guidelines was used.

According to the classification of hemorrhoidal disease, there are acute and chronic forms. In turn, the chronic one is divided into 4 grades:

Grade I: No prolapse, just prominent blood vessels

Grade II: Prolapse upon bearing down, but spontaneous reduction

Grade III: Prolapse upon bearing down requiring manual reduction

Grade IV: Prolapse with inability to be manually reduced.

The wound was assessed using M.I. Kuzin's classification: phase 1 — the phase of inflammation, which is divided into the period of vascular changes and the period of wound cleansing, phase 2 — the phase of regeneration, formation and maturation of granulation tissue, phase 3 — the phase of scar formation and reorganization [4,5].

The diagnosis of hemorrhoids was established on the basis of complaints and history, digital examination and instrumental diagnostics.

From January to December 2019, 134 patients with external and internal hemorrhoids of Grade IV who met the inclusion criteria were admitted to the hospital. All the patients signed their consent to participate in the study. Thirty-three patients were excluded due to a violation of the study protocol. The main group included 50 patients, the control group — 51 patients. The informed consents to participate in the study were signed by 101 patients of the main and control groups.

Before the surgery, all the patients underwent transrectal ultrasound and colonoscopy. Gastroscopy was performed in patients older than 45 years or with the presence of stomach diseases. The patients underwent open hemorrhoidectomy using electrocoagulation.

After the surgery the patients were randomized into the main and control groups.

The patients of the main group underwent daily control of the postoperative wound using antiseptics and ointment. In addition, LTAP was applied for 4 minutes (the procedure was performed on days 1-8, 14, 21, 30 after surgery and then every 7 days until the wound healed). In the control group, the postoperative wound control was performed only with the use of antiseptic solutions and ointment.

The average age of the patients was 46 (39;56) and 47 (40;55) years in the main and control groups, respectively, (p = 0.65). There were 33 (66.0%) males and 17 (34.0%) females in the main group. The control group included

Table 1. *Preoperative factors and comorbidities*

	Main Group (n = 50)	Control Group (n = 51)	*P*
Gender Females Males	 17 (34.0%) 33 (66.0%)	 22 (43.0%) 29 (57.0%)	0.42
Age, years	46 (39;56)	47 (40;55)	0.65
BMI, kg/m^2	26.6 (23.7;29)	25.5 (23.7;29)	0.57
Surgery duration, minutes	25 (20;30)	27 (20;30)	0.37
Hospital stay, days	7.6 (7;7)	7.1 (7;7)	0.37
Hemoglobin, g/l	138.5 (127;143)	138 (127;146)	0.9
Hypertension	7 (14.0%)	10 (20.0%)	0.6
Smoking	8 (16.0%)	12 (23.5%)	0.46
Diabetes mellitus	1 (2.0%)	2 (4.0%)	1.0
Violation of blood supply to the lower limbs	4 (8.0%)	5 (10.0%)	1.0

29 (57.0%) males and 22 (43.0%) females. According to antromometric data, hospital stay, operation time, the level of hemoglobin, as well as the presence of comorbidities, the patients in the main and control groups did not differ significantly (Table 1).

The area of the postoperative wound was calculated using the LP_SQUARE 50 program. The change in the wound area and the healing rate were calculated using the formula of Troitsa A.E. and Peschansky V.S. (on days 2, 8, 14, 21, 30, 37, 45) (Fig. 1). The assessment of the wound healing was carried out using cytology of smears-prints from the surface of the postoperative wound.

After treating the wound with an antiseptic solution, all the patients underwent a microbiology of the wound secretion samples. Also, in the patients of the main group the microbiological culture samples were taken from the wound after the LTAP treatment. The quality of life was assessed using the SF-36 questionnaire (before surgery, on days 8 and 30 after surgery). The pain syndrome was studied using the visual analog pain scale (on days 1-8, 14, 21, 30).

STATISTICAL ANALYSIS

Continuous quantitative data in connection with an abnormal distribution were described by the median quartiles (Q25;Q75) with the minimum and maximum values. The comparison of the two groups was carried out by the Mann-Whitney test. The comparison of the frequency of indicators was carried out by the Fisher test. The statistical

$$S = (S_0 - St)/S_0 \times 100$$

The formula of Peschanskiy V.S.
S_o — the initial area of the wound
S_t — the area of the wound on the day of its measurement

$$V = (S_0 - St)/n$$

The formula of Troitsa A.E.
S_o — the initial area of the wound
S_t — the area of the wound on the day of its measurement
n — the number of days

Figure 1. *Rate of the wound healing calculating formulae and changes in wound area*

analysis was carried out in the program Statistica 13.3 TYBCO USA.

LOW-TEMPERATURE ARGON PLASMA PROCESSING TECHNIQUE

The wound treatment was carried out using a plasma-arc installation "Plasmoran" for the treatment of wounds. This installation has 6 modes (A1, B1, A2, B2, A3, B3). The letter A is a mode with a lower electric energy of the plasma, the letter B is a larger one. The numbers indicate the power of the gas flow. After removing the dressing, the wound was treated with an antiseptic solution and dried. The Plasmoran device was used in the B2 mode. In the first phase of the wound process, the distance from the plasma torch nozzle to the wound surface was 15 cm, in the second and third phases — 25 cm. The duration of the procedure was 4 minutes.

RESULTS

The time of wound healing was assessed visually and by digital examination, confirmed by the cytology. By day 30, wound healing in the main group was detected in 38 (76.0%) patients, while in the control group only in 18 (36.0%) patients, $p < 0.0001$. By day 37, complete wound epithelization occurred in 49 (98.0%) patients of the main group and in 41 (80.0%) patients of the control group, $p = 0.008$. By day 45, wound healing was detected in 51 (100.0%) and 49 (96.0%) patients in the main and control groups, ($p = 0.5$).

According to the cytology of smears-prints from the wound surface on the 2nd day after the surgery, few leukocytes, blood elements, amorphous masses, scanty cocci flora were found in all cases in the main and control groups (Fig. 2).

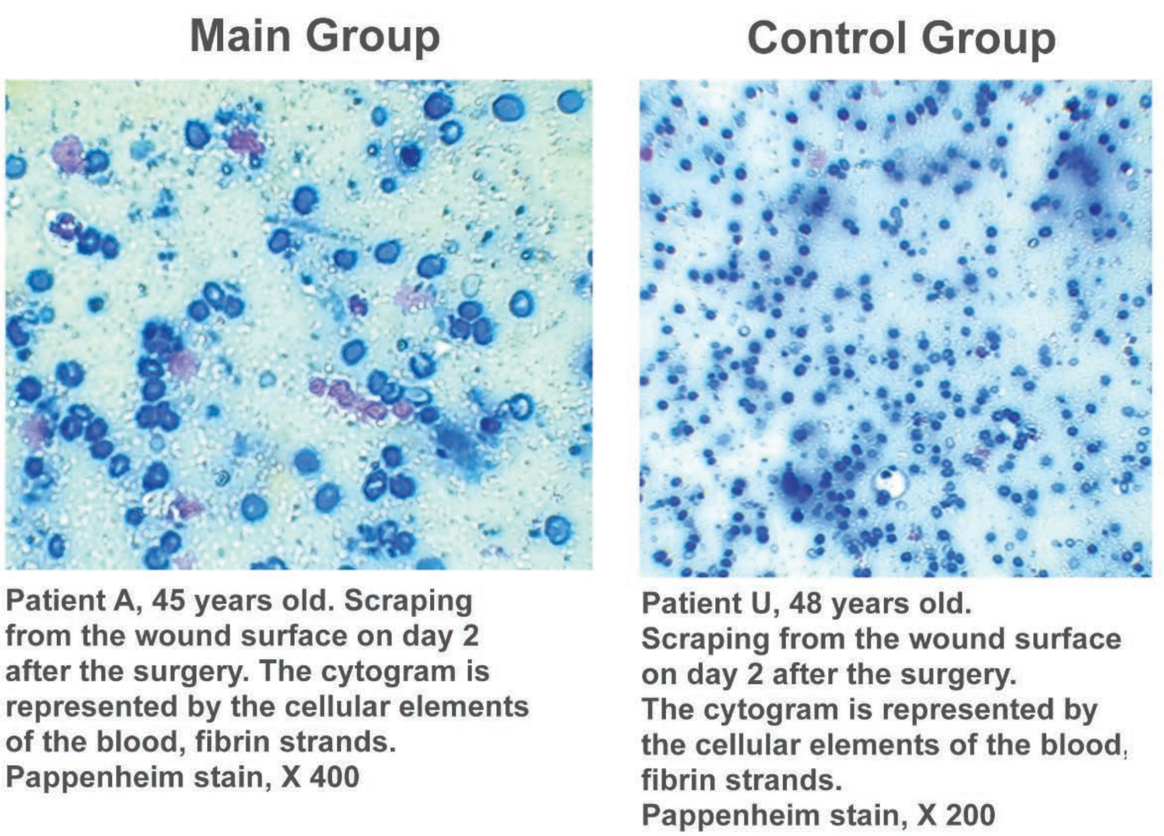

Figure 2. *Cytogram on day 2 after surgery*

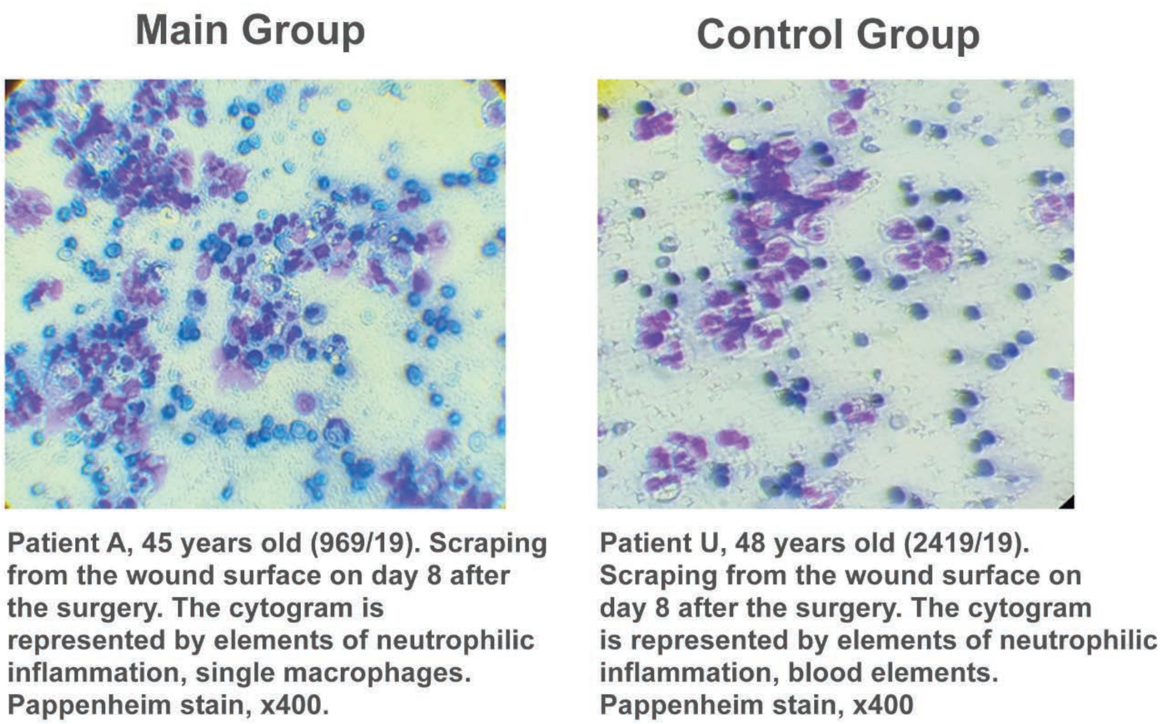

Figure 3. *Cytogram on the 8th day after surgery*

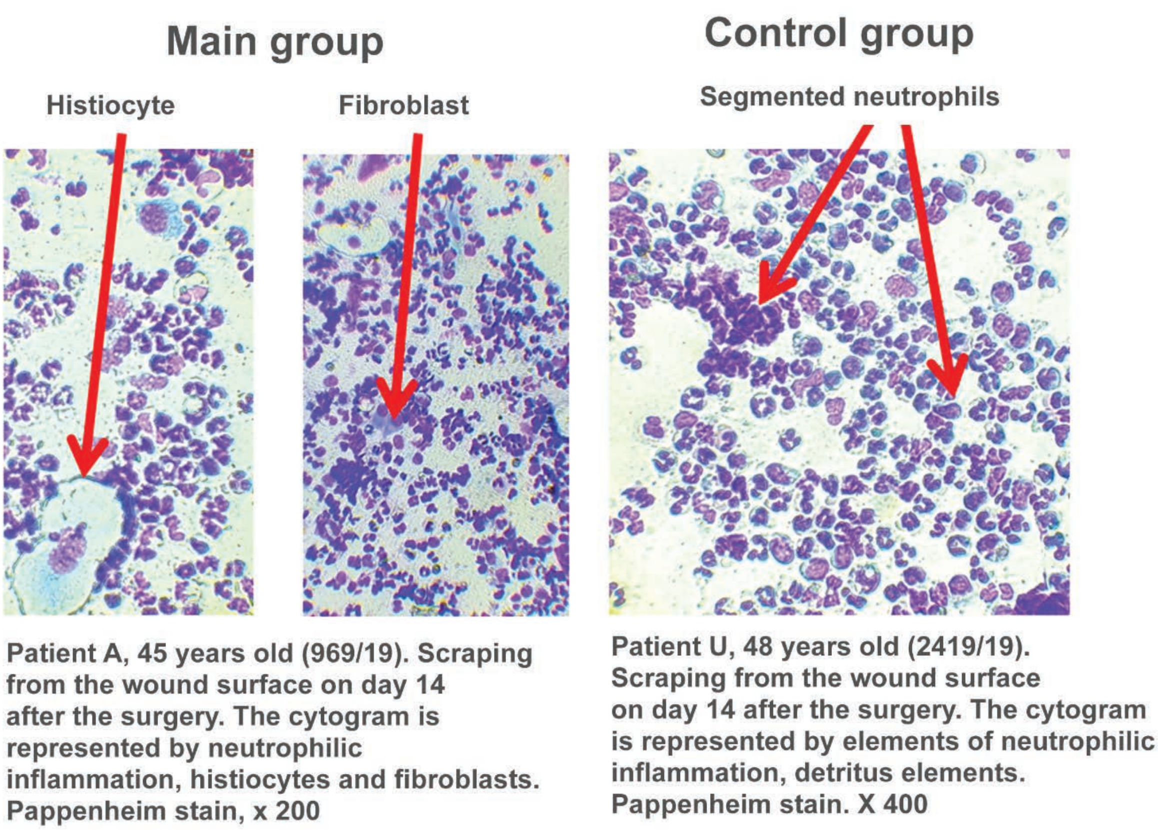

Figure 4. *Cytogram on the 14th day after surgery*

Table 2. *Assessment of the quality of life (SF-36)*

	Main Group, (n = 50)	Control Group, (n = 51)	P
Before surgery (physical health)	49 (43; 52)	45 (40; 52)	0.21
Before surgery (mental health)	44 (39; 56)	42 (35; 48)	0.1
Day 8 (physical health)	43 (36; 50)	38 (35; 44)	0.12
Day 8 (mental health)	45 (39; 54)	43 (37; 48)	0.16
Day 30 (physical health)	48 (44; 53)	42 (38; 47)	0.0007
Day 30 (mental health)	44 (40; 54)	45 (40; 52)	0.51

By day 8 after surgery, all the patients of the main and control groups had a moderate number of elements of neutrophilic inflammation (segmented leukocytes 90%, lymphocytes 5%, macrophages 5%), fibrin, blood elements, which indicates the first phase of the wound process (Fig. 3).

On day 14 after the surgery, 47 (94.0%) patients of the main group and 28 (55.0%) patients of the control group had fibroblasts and histiocytes, which indicates the transition of the healing process to the second phase of the wound process, $p = 0.001$ (Fig. 4).

By day 21 after the surgery, 47 (94.0%) and 28 (55.0%) patients of the main and control groups, respectively, had histiocyte cells, fibroblasts, and cells of immature squamous epithelium, which indicates the transition to the third phase of wound healing, $p = 0.001$ (Fig. 5).

On day 30 after the surgery, 38 (76.0%) and 19 (37.0%) patients of the main and control groups, respectively, had immatured and matured squamous epithelium cells in the smears-prints of the wound surface, while there were no signs of inflammation, which indicates complete healing of the postoperative wound, $p = 0.001$ (Fig. 6).

Due to microbiology results, the wound contamination in most cases consisted of the following clinically significant microorganisms: *E. coli, Enterococcus faecalis, Klebsiella pneumoniae, Pseudomanonas aeruginosa, Staphylococcus epidermidis hem+*.

On day 2 after the surgery in the patients of the main group before the use of LTAP, *E. coli* was detected in 29 (58.0%) patients — the average degree of contamination was 10*6.6 CFU, in 2 (4.0%) patients *Klebsiella pneumonia and Staphylococcus epidermidis hem+* in the amount of 10*7.0 and 10*4 CFU, respectively, in 6 (12.0%) patients *Enterococcus faecalis* — 10*6.3 CFU, *Pseudomanonas aeruginosa* was detected in 1 (2.0%) patient, the value was 10*7.0 CFU. After applying LTAP on day 2 after the surgery, the microbial content of the wound changed. *E. coli* was isolated in 18 (36%) patients at a value of 10*6.0 CFU, *Enterococcus faecalis* in 6 (12.0%) patients — 10*5.8 CFU, *Staphylococcus epidermidis hem+* in 2 (4%) patients — an average contamination of 10*6.2 CFU, *Klebsiella pneumoniae and Pseudomanonas aeruginosa* were not detected in any patient. In the control group, on day 2 after surgery, *E. coli* was most common in 24 (47.0%) patients in a concentration of 10*7.0 CFU, *Klebsiella pneumonia* — in 4 (8.0%) patients — 10*8 CFU, *Staphylococcus epidermidis hem+* — in 3 (6.0%) patients in a concentration of 10*5.5 CFU, *Enterococcus faecalis* was detected in 7 (14.0%) patients — 10*6.0 CFU, and *Pseudomanonas aeruginosa* in no patient. On day 8 after the surgery, *E. coli* was most often detected in the patients of the main group before the LTAP treatment — the average contamination value was 10*6.8 CFU, in 2 (4.0%) patients *Klebsiella pneumoniae* — 10*8.0 CFU, in 1 (2.0%) patient — *Staphylococcus pidermidis hem+* in a

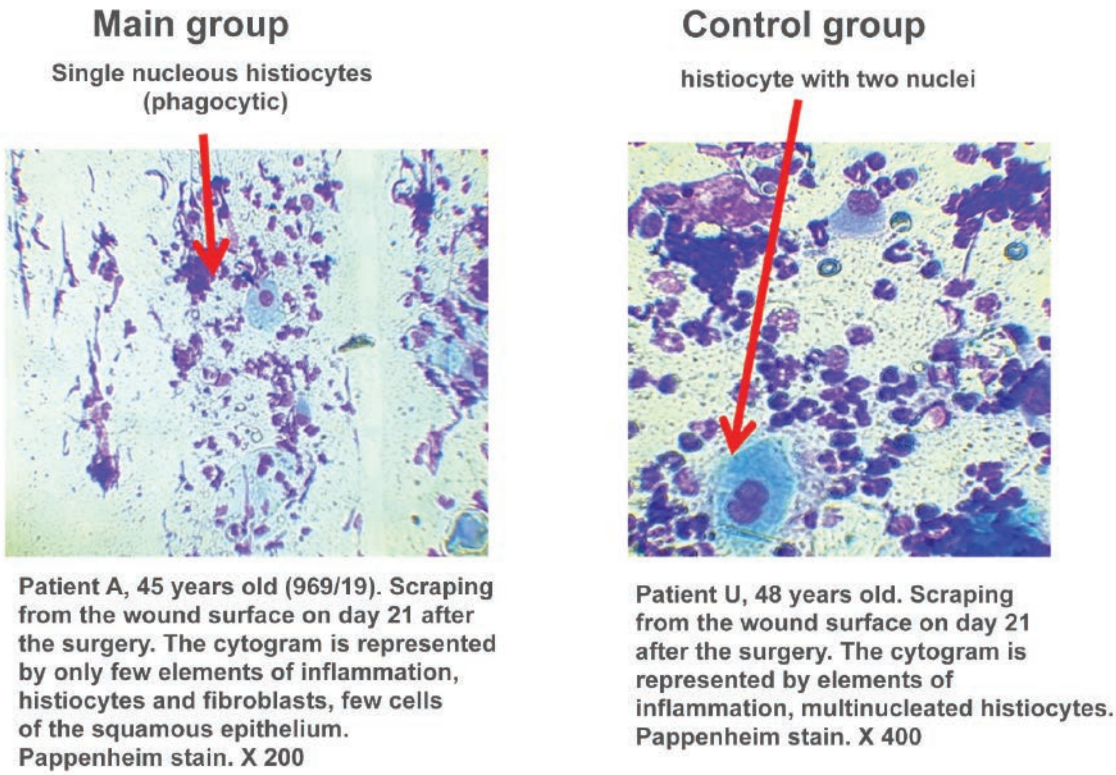

Figure 5. *Cytogram on day 21 after surgery*

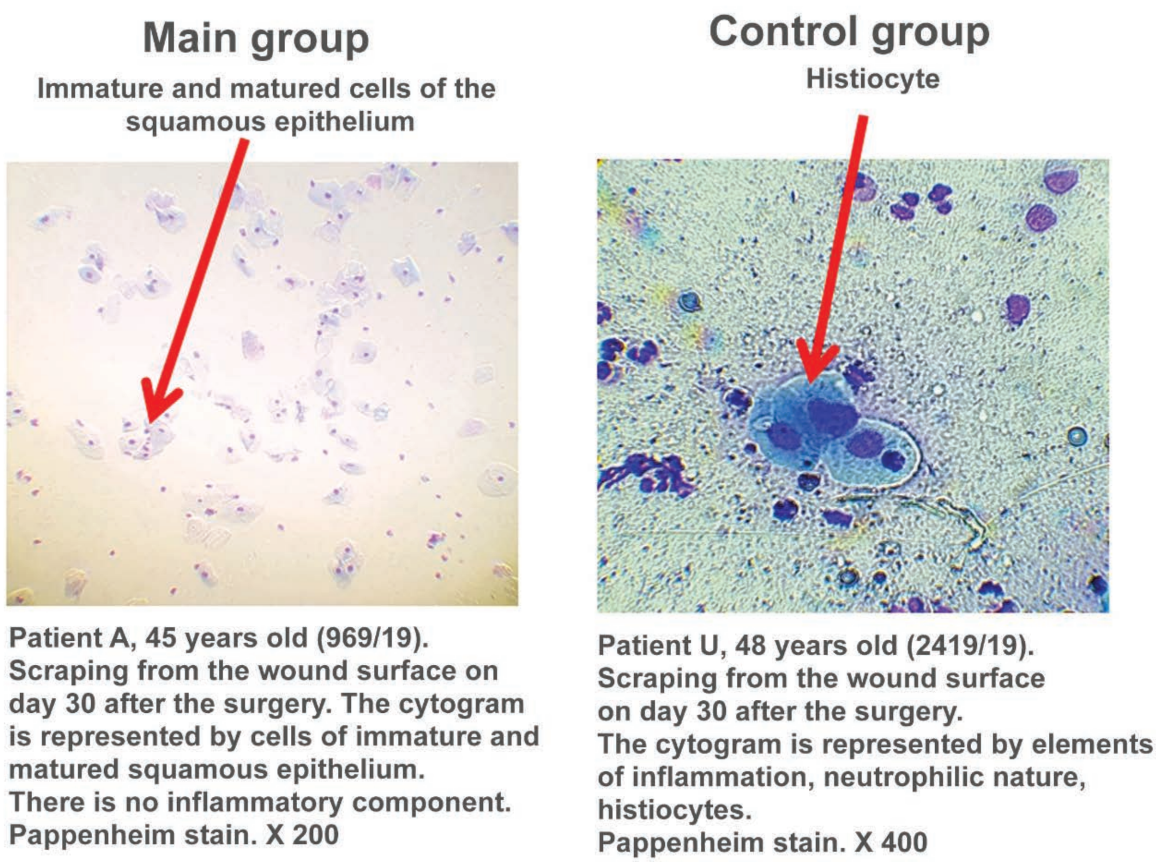

Figure 6. *Cytogram 30 days after surgery*

Table 3. *Postoperative wound area*

	Main Group, (*n* = 50)	Control Group, (*n* = 51)	*P*
Day 2, cm^2	2.2 (1.94; 2.45)	2.2 (1.9; 2.33)	0.49
Day 8, cm^2	1.81 (1.57; 1.96)	1.78 (1.55; 1.93)	0.8
Day 14, cm^2	1.3 (1.05; 1.49)	1.4 (1.16; 1.51)	1
Day 21, cm^2	0.6 (0.36; 0.87)	0.86 (0.71; 1.15)	0.23
Day 30, cm^2	0 (0; 0)	0.31 (0; 0.31)	0.16

concentration of 10*7.0 CFU, and *Enterococcus faecalis* and *Pseudomanonas aegidinos* were not detected in any patient. After the LTAP treatment, *E.coli* was most often detected in 39 (78.0%) patients in the amount of 10*6.0 CFU, *Staphylococcus epidermidis hem+* and *Enterococcus faecalis* were detected in 1 (2.0%) patient in the amount of 10*4.0 CFU in both cases, *Klebsiella pneumoniae* and *Pseudomanonas aeruginosa* were not detected in any patient. In the patients of the control group, on day 8 after the surgery, *E. coli* was detected in 38 (75.0%) patients in the amount of 10*7.0 CFU, *Klebsiella rheimopia* and *Enterococcus faecalis* — in 4 (8.0%) patients, the average value was 10*5.0 and 10*5.25 CFU, respectively. *Pseudomanonas aeruginosa* and *Staphylococcus epidermidis hem+* were not found in any patient. On day 30 after the surgery, *E.coli* was detected in 8 (16.0%) cases in the patients of the main group before the LTAP treatment, *Enterococcus faecalis* was detected in 3 (6.0%) cases — an average value of 10*4.6 CFU, *Staphylococcus epidermidis hem+* was detected in 1 (2.0%) patient in an amount of 10*7.0 CFU, and *Klebsiella pneumoniae* and *Pseudomanonas aeruginosa* were not detected in any patients. After treatment with the LTAP, *E. coli* was found in 7 (14%) patients — an average number of 10*4.0 CFU, *Enterococcus faecalis* in 3 (6.0%) patients in an amount of 10*6.7 CFU. *Klebsiella pneumoniae, Staphylococcus epidermidis hem+* and *Pseudomanonas aeruginosa* were not found in any patients. In the patients of the control group, on day 30 after the surgery, *E. coli* was most common in 20 (39.0%) cases in the amount of 10*7.0 CFU. *Klebsiella pneumonia* and *Enterococcus faecalis* in 2 (4.0%) patients — 10*6.0 CFU and 10*5.5 CFU, respectively. In 1 (2.0%) patient, *Staphylococcus epidermidis* + was detected in an amount of 10*6.0 CFU. *Pseudomonas aeruginosa* was not detected in any patients.

Before the surgery, the quality of life of the patients according to the SF-36 questionnaire did not differ. On day 8 after the surgery, the quality of life (mental and physical components) between the groups also did not differ significantly. By day 30 after the surgery, the analysis of the quality of life (physical component) revealed a significant difference between the main and control groups, while no such difference was found when evaluating the psychological component (Table 2).

On day 2 after the surgery, the area of the postoperative wound did not differ significantly in the patients of the main and control groups.

However, starting from day 14, it was found that the wound area in the main group was smaller than in the control group, but the difference was not significant (Table 3).

An important component in the rehabilitation of patients after surgery is the level of pain.

On day 2 after the surgery, the severity of the pain did not differ between the main and control groups. However, already on day 3, it is clear that the level of pain syndrome in the patients of the main group was less and amounted to 4.5 (4;5) points, whereas in the patients of the control group it was 5 points (4;6), $p = 0.02$. In the future, it can be seen that the level of pain

Table 4. *Pain syndrome assessment*

	Main Group, (*n* = 50)	Control Group, (*n* = 51)	*P*
Day 2	5 (4; 5)	5 (4; 6)	0.42
Day 3	4.5 (4; 5)	5 (4; 6)	0.02
Day 4	4 (4; 5)	5 (4; 6)	0.03
Day 5	4 (3; 5)	5 (3; 6)	0.13
Day 6	4 (3; 5)	4 (4; 6)	0.02
Day 7	3.5 (3; 4)	4 (3; 5)	0.003
Day 8	3 (3; 4)	4 (3; 5)	0.00004
Day 14	2 (2; 3)	3 (2; 4)	0.00004
Day 21	1 (1;2)	3 (2; 3)	0.00001
Day 30	0 (0; 0)	1 (0; 2)	0.00001

syndrome in the patients of the main group is lower than in the patients of the control group, and the difference is significant (Table 4).

DISCUSSION

The American scientist Langmuir I. in 1928 for the first time wrote in his work about plasma as the fourth aggregate state of matter. Initially, high-temperature argon plasma was obtained, which was used in the sterilization of implants and various medical instruments, and in practical medicine for vascular coagulation and tissue dissection [6]. After that, it was possible to obtain low-temperature argon plasma, which became possible to be safely used on human soft tissues.

Chuangsuwanich A. (2016) in randomized prospective study used LTAP in the treatment of bedsores of various sites in 50 patients [7]. In addition to daily dressings, the patients of the main group were treated with the LTAP once a week, while the patients of the control group had only daily dressings. On day 28 after the start of treatment, the wound area in the main group decreased in 23 (88.0%) patients, while in the control group only in 3 (26.1%) patients ($p < 0.001$). The amount of exudate released on day 28 in the patients of the main group decreased in 18 (69.2%) patients, and in the control group — in 3 (13.0%) patients ($p < 0.001$). We conducted a study that studied the effect of LTAP on wound healing after open hemorrhoidectomy. Thus, by day 30, 38 (76.0%) patients of the main group had complete epithelization of the wound, which is 40% faster than in the patients of the control group.

Acceleration of wound healing occurs due to the fact that LTAP promotes wound healing in all 3 phases of the wound process. In the first phase of the wound healing, it is important to clean the wound as quickly as possible. The LTAP promotes the formation of MCP-1 and IL-6, which accelerate the migration of macrophages into the wound [8]. In the 2nd phase of the wound process, the active formation of granulation tissue occurs. Fibroblasts play a leading role in its formation. The LTAP promotes the migration of fibroblasts to the wound surface as early as 12 hours after its use [8-10].

In the 3rd phase of the wound process, wound epithelization and scar formation occur. Due to the action of the LTAP, endogenous NO and HIF1α protein are produced [11-13]. In turn, NO stimulates the formation of type 1 collagen.

The HIF1α protein is actively involved in neoangiogenesis, which is an important part in the wound healing process. The LTAP promotes the production of β1-integrins, which are involved in the migration, proliferation and differentiation of epidermal cells [14,15].

A decrease in the microbiological contamination of the wound leads to its faster healing. According to the world literature, LTAP has bactericidal and bacteriostatic properties [16,17]. This is due to hydrogen peroxide, UV radiation, ozone and reactive oxygen species [18,19]. According to the results of the microbiology, the bactericidal effect of LTAP was determined against clinically significant microorganisms associated with infection of the wound surface in patients after general anal surgeries. The randomized study showed that the use of LTAP promotes faster wound healing and this technique can be widely used in medicine. The technique itself does not require special training, the Plasmoran device is mobile and easy to use, and the consumables do not require large expenses.

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AUTHORS CONTRIBUTTON:

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