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Evaluation of vulvar leukoplakia photodynamic therapy efficiency by fluorescent diagnostics method with local «Alasens®» photosensitizer application

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ABSTRACT

Purpose: Using continuous-pulse irradiation mode application for performing photodynamic therapy sessions to introduce a new method, and conclude results of clinical research focused on vulvar leukoplakia photodynamic therapy efficiency in combination with topical aqueous «Alasens®» solution administration.
Method: Seventy patients (average age of 61 years) diagnosed with vulvar leukoplakia disease (2018 ICD-10-CM Diagnosis Code N90.4) were examined. The following values represent doses of combined mode photodynamic therapy session: 12 J/cm^2 pulse radiation dose; 3.5 J/cm^2 continuous radiation dose. Non-invasive spectroscopic and visual control of drug accumulation in real time was carried out by fluorescence diagnostic method before and after each therapy session.
Results: Single-therapy session efficiency was estimated by a fluorescent signal reduction in the pathological region after irradiation, and the direct correlation between photosensitizer photobleaching and disease regression was registered. Photodynamic therapy course included three procedures, with each session applied in 24 -h intervals, and when necessary, an additional course of therapy was applied 60 days afterward. Significant post-treatment results took effect after 2–3 courses with symptom presence reduced or fully regressed depending on the initial severity of the disease. Additionally, side effects and sequelae remained absent in all cases.
Conclusion: The result of methods applied during the clinical research period indicate strong potential in utilizing such promising technology to contribute to the possible prevention of malignant transformation and the treatment of vulvar leukoplakia.

1. Introduction

Vulvar leukoplakia is a noninflammatory disorder and pathological modification of external genitalia multilayered flat epithelium that is accompanied by skin and mucosa cornification (according to the international classification 2018 ICD-10-CM with Diagnosis Code N90.4). Vulvar leukoplakia is a prolonged course, characterized by the severity of clinical manifestations, frequent relapses, and a high likelihood of vulva malignant transformation against the background of prior pathological processes. The frequency of vulvar leukoplakia malignant transition into cancer varies between 13–31% according to previous clinical experience. Leukoplakia often accompanies dermatoses and the development of inflammatory diseases due to infection vulnerability through cracks in mucosa [1–5]. Traditional methods of treatment in similar nature pathologies are surgical intervention and non-surgical techniques. Examples of non-surgical methods are CO_2 laser coagulation, vaporization of lesion area with high-intensity radiation, application of high-intensity laser radiation, ultrasound exposure, chemotherapy, and immune-modulatory therapy [6–12]. The imperfection of existing conservative and operative treatment methods results in the imperative need for improved and optimized theranostics' approaches.

Modern physical theranostics methods (fluorescent diagnostics [FD] and photodynamic therapy [PDT]) are considered to be effective in terms of radical and palliative treatment of oncological lesions of

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different localization and nature. FD and PDT are distinguished by low invasiveness, selective impact on pathologically altered tissues, the possibility of multiple sessions, the absence of severe systemic and local complications, and excellent cosmetic effect [13–21].

FD and PDT methods are based on the interaction between photosensitizer (PS), light, and oxygen located in the irradiation zone biotissue. Theranostics begins with PS medication being administrated to the patient. Due to specific processes and features occurring in pathological tissue, PSs accumulate predominantly in the lesion region without developing in surrounding healthy tissue. In relation to photonics science, mechanisms taking place during FD and PDT are described hereinafter. While absorbing the radiation of a wavelength corresponding to PS absorption band, PS transfers from the ground energy state to a singlet excited one. The reverse transition of PS to the ground state occurs in the form of fluorescence emission described by a short lifetime of 10 ns order. Meanwhile, a portion of excitation energy passes to the long-lived triplet level of PS while selection rules forbid transition to the ground state (phosphorescence). Phosphorescence is characterized by a durable lifetime of 10^{-2}-10^{-4} s depending on the extinguishers' presence in PS' microenvironment. Molecular oxygen present in the PS surrounding dissolves in tissues and cells that allows energy from triplet state of PS to be transferred to molecular oxygen (that is excited from triplet to singlet state). The critical fact is that singlet oxygen is a strong oxidant. It oxidizes biological molecules located in the nearest microenvironment (so-called chemical quenching) and also can oxidize the very molecule of PS (photobleaching) [22]. As a result of oxidation, photodestruction of PS molecule occurs with the subsequent formation of photo-reduction products (presumably, chlorine type photoproducts called "photoproduct 1,2" in the literature, also photodynamically active) [23]. The process of PS oxidation allows qualitative evaluation of PDT efficiency that provides an opportunity of changing and controlling treatment tactics when necessary. For instance, if fluorescence intensity dropped to 10–20% of initial value during irradiation, it is concluded that at a minimum of 2 mm depth (depth of probing) PS has oxidized, and if lesion depth does not exceed 2 mm, then irradiation can be completed. PS absence at a depth of 2 mm defeats the purpose of carrying out PDT session because therapy efficiency is directly related to "oxygen effect." For effective pathological cells destruction, it is essential to wait until oxygenation and influx levels of PS are restored. That would be followed by lesion irradiation or subsequent additional PS' dose administration in case of insufficient PS concentration in pathologically altered tissue. If lesion depth exceeds 2 mm, it is recommended to continue irradiation considering model calculations of light passage through tissue. If over time a rapid restoration of fluorescence intensity signal is observed in certain zones, it means that destruction of affected areas was not complete. Especially in terms of microvessels destruction and therefore additional irradiation is required.

An essential advantage of spectroscopic analysis is the opportunity to obtain information about the dynamics of various biological processes in real time. FD allows data collection of PS accumulation levels in affected cells by measuring fluorescent signal intensity. This enables FD to detect and specify localization of pathological formation, select optimal parameters for conducting PDT sessions, and evaluate therapy efficiency post session.

For the treatment of vulvar leukoplakia and diseases of similar nature (lichen sclerosus, vulvar intraepithelial neoplasia (VIN) of I-III grades and other dermatoses) PDT method is considered the most effective in comparison with existing ones [24–27]. Potential of applying standard continuous irradiation mode for PDT of these diseases was previously studied using PSs such as 5-aminolevulinic acid (5-ALA) (10% and 20%) and Photolon [27–29].

The novelty of this study lies in the application of combined continuous-pulse irradiation mode for PDT of vulvar leukoplakia. Evaluation of proposed PDT method efficiency using a 0.5% aqueous solution of 5-ALA as PS defines the purpose of this research.

2. Materials

Studies were performed at the City Clinical Hospital №40 Moscow Health Department. Seventy patients diagnosed with vulvar leukoplakia (according to the international classification 2018 ICD-10-CM Diagnosis Code N90.4) participated in the research and the age of patients varied from 47 to 75 years. All patients had continuous anamnesis between 3 to 5 years, severe clinical symptoms in the form of pruritus, and in 9 cases itching led to sleep disturbance. Conservative manipulations in the form of various ointments application had a temporary effect.

5-ALA-induced protoporphyrin IX is an effective photodynamic agent widely used in clinical practice for oncological diseases treatment and diagnostics [30–35]. 5-ALA (Alasens®) is officially approved for clinical application trials in Russia with the regulation of all PS parameters [36]. In this study, protoporphyrin IX, being an intermediate product of reaction chains of heme biosynthesis from 5-ALA, serves as PS. 5-ALA-induced protoporphyrin IX is characterized by high fluorescent contrast and enhanced accumulation in types of rapidly proliferating tissues with ferrochelatase deficiency. Protoporphyrin IX spreads not only inside the intercellular cell matrix but also in its extracellular space. The significant amounts of it persist in the lesion for several hours, while in contrast with healthy cells it rapidly transforms into non-photoactive porphyrin under ferrochelatase impact. In other words, protoporphyrin IX accumulation in pathological cells is due to the lack of ferrochelatase enzyme within the cells in comparison with healthy ones

Thus, Alasens® medicine (synthesized by Organic Intermediates & Dyes Institute (NIOPIK), Russia) was used as PS for FD and PDT in the current study. Sterile aqueous PS solution with a concentration of c = 5 g/l was prepared one hour before use by dissolving the required amount of Alasens® powder in distilled water. 5-ALA in solution is utilized more efficiently in comparison with gel form. Cotton- gauze swab soaked in drug solution was applied topically on external genitals, and local administration process of PS was carried out for 3–4 hours. The procedure was followed by visual and spectroscopic pretreatment-FD, PDT session, and posttreatment-FD. Lesion surface was not treated in a particular way before PS application; patients carried out regular water-hygienic procedures.

Preliminary studies carried out on the selection of PS dosage have demonstrated that above-mentioned 5-ALA concentration for topical administration is sufficient for protoporphyrin IX stimulation, followed by conducting theranostics. Additionally, treatment without PS administration revealed that any photodynamic effects were non existent. 5-ALA 0.5% dose proved to be optimal and more profitable in terms of price and effect.

Biopsy tests were carried out regardless of the preliminary cytological examination results. Biopsy samples were collected from each patient on the lesion region with maximum fluorescent contrast during preliminary visual FD. All 70 patients showed histological results were consistent with leukoplakia and in all cases, the major part of vulva was affected. In 14 patients transition of dystrophic changes to the perianal region was observed, and due to the prolonged course of the disease, 5 patients experienced affected inguinal-femoral folds. Phototheranostics session was conducted no earlier than one month after biopsy analysis.

3. Methods

Studies devoted to pain effect assessment were conducted on volunteers and showed that pain effects occur even in cases of applying low radiation doses. Furthermore, pain effect also had individual nature. To neutralize the pain phenomenon, an approach of the pulsed light application was implemented, and it was noted that the use of pulsed radiation leads to anesthesia effects of irradiated tissue containing PS [37]. However, pulsed radiation occurs in a wide spectral range, and thus, a significant amount of light energy goes for tissue

heating and not photodynamic response achievement.

For this reason, continuous laser radiation of wavelength corresponding to the absorption maximum of PS was involved in the PDT process simultaneously (in parallel) with pulsed radiation. Directed continuous laser exposure provokes active singlet oxygen generation that undeniably affects PDT efficiency. Radiation dosage was selected in correspondence of total energy being sufficient for photodynamic effect achievement. In the case of leukoplakia treatment, the total dose amounted to 15 ± 2 J/cm2, and pulsed excitation in combination with continuous one increased the depth of radiation penetration which allowed affecting deeper lesion layers during therapy.

Light-emitting diode (LED) system was used for PDT of vulvar leukoplakia in combined continuous-pulse irradiation mode. After 3–4 hours of topical administration of the drug, the efficiency of the proposed therapy method was assessed by carrying out visual and spectroscopic fluorescence monitoring before and immediately after PDT

3.1. Visual FD and PDT

For visual FD and PDT, a unique LED system with a pulsed source based on xenon gas-discharge lamp was used (Fig. 1). This system contains an annular pulse lamp (providing radiation with energy of 600 J/pulse) and a group of LEDs emitting in red (630 +/- 5 nm) and blue (400 ± 5 nm) optical spectral ranges. The first LEDs are intended for PDT, and others are for visual FD which allows the possibility to conduct a visual assessment of lesion boundaries and identifying the quantity of lesions in a specified area after PS accumulation. LEDs and the annular pulse lamp are placed specifically to provide simultaneous continuous-pulse mode irradiation of the pathological tissue site. As previous studies on phantom models have shown, simultaneous pulsed and continuous light exposure leads to an increase in penetration depth of radiation by 10–15%, and a decrease of patients' sensitivity to intense heat during irradiation. This provides an opportunity to enhance the efficiency of PDT session and shorten its duration by increasing the penetration depth of radiation and reducing the pain patients experience during therapy.

Optimal "complete" effective light dose is chosen by the patients' sensitivity to intense heat prior to PDT treatment. Total light dose of pulsed radiation during PDT session amounted to 12 J/cm^2 (energy density in a pulse – 0.3 J/pulse*cm^2, number of pulses per session – 40); continuous light dose – 3.5 J/cm^2 (power density – 70 mW/cm^2, exposure time – 50 s). Average total light dose reached 15 ± 2 J/cm^2 which is 10–20 times lower than the commonly accepted irradiation parameters. The critical light dose responsible for photodynamic effect achievement was contributed by laser diode radiation with λ = 630 +/-5 nm.

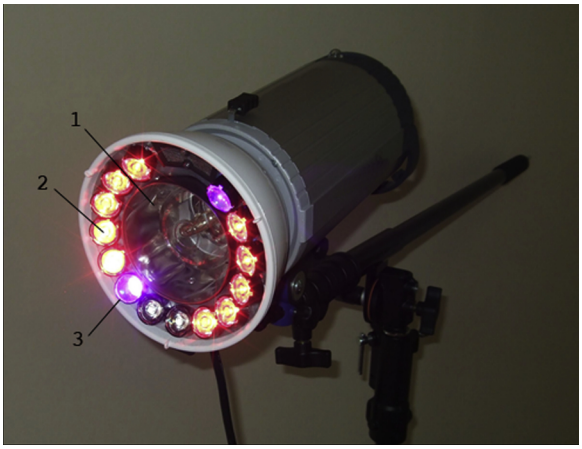

Fig. 1. LED system with pulsed source based on the gas-discharge lamp for FD and PDT of skin non-oncological and precancerous diseases. Gas-discharge lamp (1), LEDs with λ = 630 +/- 5 nm (2), LEDs with λ = 400 +/-5 nm (2).

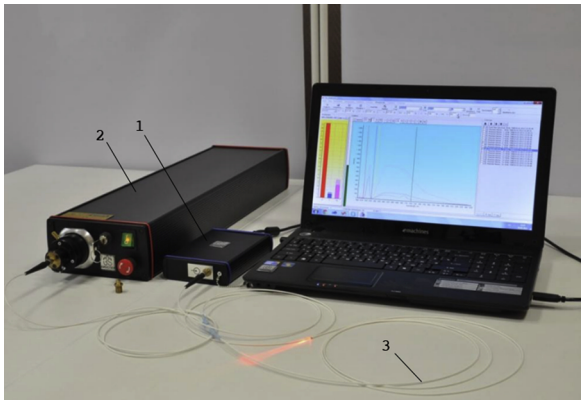

Fig. 2. Fiber optic spectrometer LESA-01 "BIOSPEC". Laptop with specialized software, spectrum analyzer (1), laser diagnostic source λ = 632.8 nm (2), fiber-optic diagnostic probe (3).

3.2. Spectroscopic FD

Spectroscopic control of PDT was carried out via fiber-optic spectrum analyzer LESA-01-BIOSPEC (Fig. 2) by quantitative registration of 5-ALA-induced protoporphyrin IX fluorescence intensity. PS fluorescence was excited by a laser diagnostic radiation source with a wavelength of λ = 632.8 nm, and with power on the output of fiber tip set to Pex = 4–5 mW. Monitoring the development of the lesion area was performed using a diagnostic fiber-optic probe with emitting and receiving fibers. Laser radiation supply on zones of interest was provided using optical fiber. Passing an optical path in biotissue, photons migrate along a "banana" shaped trajectory and are registered by receiving fibers connected to a spectrometer. Once a signal is obtained it is then processed using dedicated spectrometer software.

4. Results

Despite the variety of methods for PS application, intravenous administration is associated with high drug costs and sensitization of the whole organism. Therefore, topical drug administration is a preferred method for the local procedure. Protocol parameters carried out in this study demonstrated a significant positive result. The concentration of 5-ALA 0.5% was selected empirically by conducting numerous experiments on volunteers diagnosed with leukoplakia. Optimal PS dosage providing the maximum therapeutic and minimal pain effect was achieved by testing 5-ALA solutions of different concentrations.

High fluorescent contrast (red glow observed in blue light) relative to surrounding tissue is the result of ALA-induced protoporphyrin IX accumulation. This property provides a visual assessment to help determine the amount of lesions and their boundaries. Color and fluorescent images of leukoplakia region were obtained before PDT (Fig. 3). At the end of the PDT session, "photobleaching" effect was observed which manifested when the reduction of visible fluorescence drug intensity decreased by up to 30%. The result of visual FD indicated a heterogeneous nature of examined lesions.

Spectroscopic FD was conducted by probing fluorescent foci identified by visual FD. Drug concentration in different pathological tissue sites varied based on quantitative spectroscopic results. Lesion area with maximum fluorescence was categorized as a control zone, and single PDT session efficiency was assessed by PS photobleaching after combined continuous-pulse irradiation of pathological centers. Fig. 5 presents a common characteristic dependency: fluorescence spectra averaged over one localization (in the control zone) for one patient after 3–4 hours of PS administration, and immediately after PDT. Optical signal was detected in the spectral range from 600 to 800 nm and included scattered laser excitation peaks. Normalizing obtained spectra by backscattered laser line allowed quantifying PS amount in tissue.

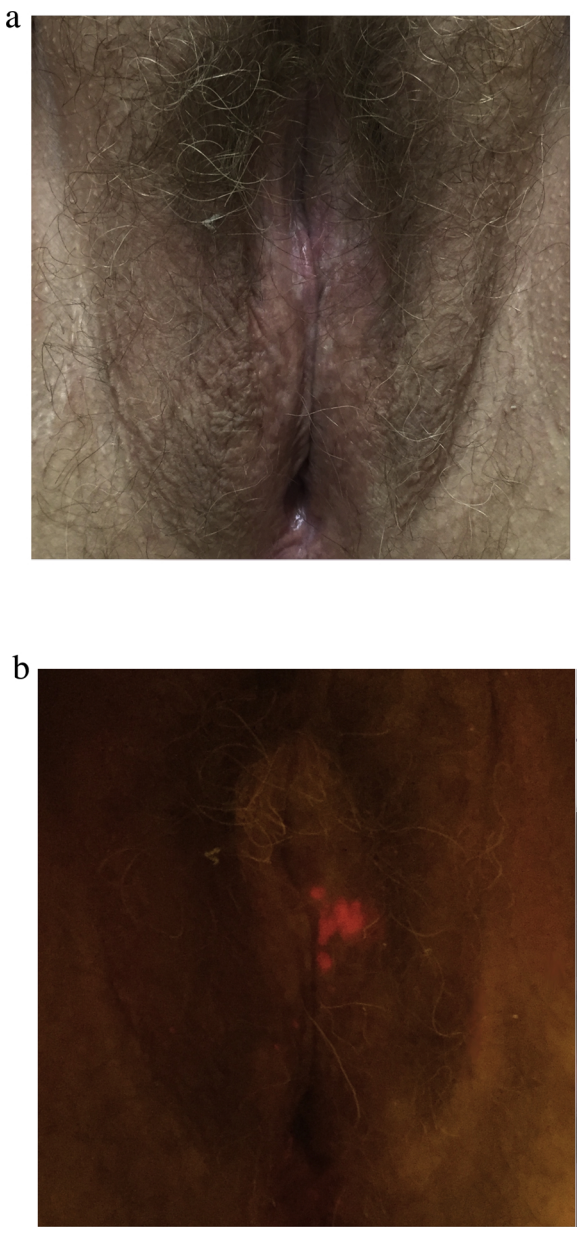

Fig. 3. Lesion region before PDT (after 3–4 hours of topical PS application). a-Visible light image; b - Fluorescent (λ_{ex} = 405 nm) image.

It is well known that oxygen is an active agent provoking cells destruction and subsequent death induced through necrosis/apoptosis mechanism. Heat is proven to be synergetic with PDT as microcirculation activity that is proportional to oxygen supply increases during the heating process. During PDT, irradiation temperature of the exposed surface is increased by 1.6 °C and was recorded using thermal monitoring with a highly sensitive IR camera.

Healthy tissue existing outside the pathology region accepted as a norm was free of any drug accumulation as no pronounced fluorescent signal was detected. Two maximum units of fluorescence spectra registered in the lesion area correspond to ALA-induced protoporphyrin IX (λ = 706 nm), and to endogenous porphyrins (λ = 690 nm) (Fig. 4). That enables observation of active PS accumulation in pathological cells and their increased content of endogenous porphyrins. The intensity of the fluorescent signal corresponding to both endogenous porphyrins [38,39] and protoporphyrin IX decreased post-treatment. Endogenous porphyrins that are a mixture of uro-, copro- and protoporphyrins are characterized by weaker photodynamic activity, as a more rapid reduction in intensity of protoporphyrin IX was observed in comparison with endogenous porphyrins.

Five spectral value characteristics were collected from each of the 70 patients examined by taking measurements in lesion areas that contained the highest PS accumulation revealed by visual fluorescent diagnostics. These areas were probed before and after PDT to monitor the impact of therapy. Data gathered was averaged out over one localization for an individual (Fig. 4) and two target groups were created based on the results of pre and post therapy intensity values for each patient. The first group of patients contributed to a poor outcome as less than 50% loss of PS concentration values after PDT was observed. Despite this outcome, the rest of patients did account for beneficial therapeutic results as PS concentration value after PDT decreased more than by 50%. In all cases, the standard deviation approach of error estimation was applied to a set of obtained data values to quantify the amount of variation. Grouping method was introduced in order to help clinicians simplify the selection of disease treatment tactics.

PS concentration reduction of 2x or higher in the lesion area was observed in 60% of patients (Fig. 5a) after PDT and based on previous studies, a decrease in fluorescence intensity of 2x or higher is a good prognosis when reflecting on the quality of the conducted therapy session. 40% of patients (Fig. 5b) showed slight declines of PS concentration which can be explained by prolonged drug accumulation due to the lack of vascular damage during PDT and by possible protoporphyrin IX diffuse redistribution.

The suggested assumption could be justified by the sensitivity of laser line of λ = 632.8 nm used for excitation of PS fluorescence to hemoglobin at a certain depth. So, by the intensity of the registered signal it is possible to monitor the dynamics of tissue oxygenation. Detected scattered back light is inversely associated with absorption of hemoglobin and oxygen concentration in tissue. Thus, blood flow disturbance accompanied by oxygen reduction leads to an increase in laser line intensity. To present diagnostics results, spectra were normalized by laser peak (Fig. 4) in order to see comparisons of pre and post 5ALA-induced PPIX fluorescence intensity. Data of non-normalized spectra that characterize the specified results was not presented.

5. Discussion

Because of active light absorption in the upper tissue layers, lower lesion layers experience a reduction in light intensity. In lesion depths of 10 mm or more, lower layers are affected to a lesser extent or not exposed to PDT at all. This causes a significant reduction in the effectiveness of therapy and may result in a relapse of disease. When this happens, influencing the entire pathological site including the lowest layers cannot be enhanced by increasing drug dose administered due to shielding of lower layers by a highly sensitized surface. Enhancement of photodynamic therapy impact on lower lesion layers has its hardware limitations when increasing the power density of continuous radiation interaction with the tissue. Furthermore, the high power density of radiation may cause overheating of the pathological site and adjacent tissues.

Due to the potential of light dosage applied reaching up to a dozen of J / cm2 for several milliseconds, pulsed radiation provides high peak power density that contributes to photobleaching of superficial endogenous and exogenous chromophores. That outcome results in a significant decrease of absorption coefficient by surface tissue layers. Previous studies on phantom models confirmed that simultaneous pulsed and continuous light exposure leads to an increase in penetration depth of radiation by 10–15%, and a decrease of patients' sensitivity to intense heat during irradiation [40,41]. This phenomenon is theoretically possible taking into account the existence of triplet long-living level of PS molecule that can contribute to the clearing effect of the medium.

Spectroscopic non-invasive analysis after PDT session demonstrated a decrease of fluorescence signal intensity of both ALA-induced protoporphyrin IX and endogenous porphyrins, which could have also contributed to obtaining therapy effect. The concentration decrease of PS

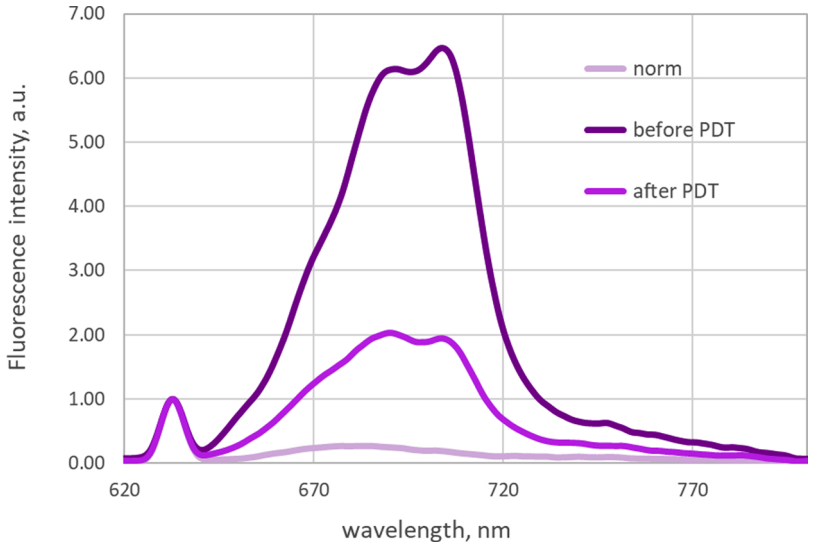

Fig. 4. Fluorescence spectra of healthy tissue, lesion region before (after 3–4 hours of topical PS application) and after PDT session. Spectra are averaged over one localization (for a zone with maximum fluorescent contrast) for one particular patient.

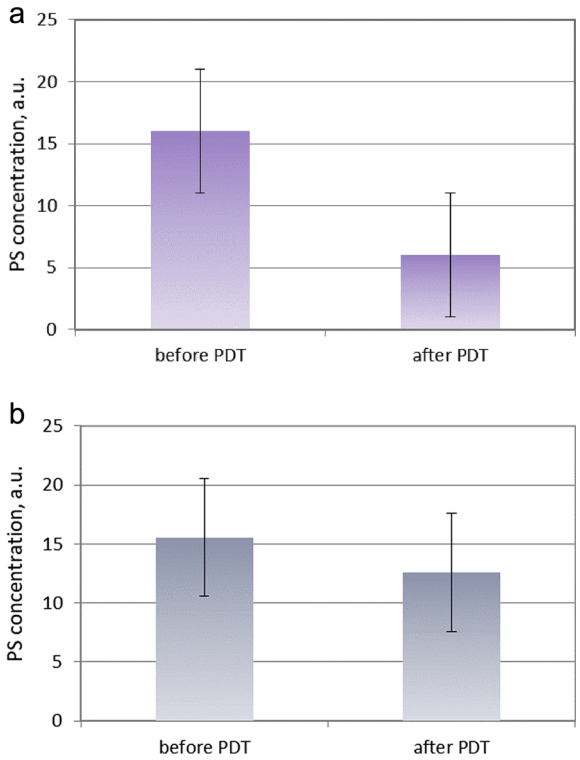

Fig. 5. Level of PS concentration in the lesion region before (after 3–4 hours of topical PS application) and after PDT session. A – in 60% of patients; b – in 40% of patients. Histograms are presented for zones with maximum fluorescent contrast.

within the lesion area after PDT is explained by partial elimination of PS molecules and the transformation of PS into photo-reduced forms under the radiation excitation on the wavelength corresponding to the absorption maximum of PS. These photo-reduced forms are called "photoproducts" in the literature. Fluorescence intensity reduction indicates that radiation has reached its goal and protoporphyrin IX located in cells produced a photodynamic effect. As a result of the photodynamic impact, a portion of generated singlet oxygen was consumed on inactivation of inflammation cells and destruction of stromal tissue part, and another portion of it was consumed on the transformation of PS. Based on previous research experience [42], the simultaneous destruction of biological tissue and oxidation of PS occurs during experiments related to PS photobleaching in biological tissues. Furthermore, the percentage ratio between energy used on the elimination of biological molecules located in PS 'microenvironment, and energy of PS itself, depends on the type of surrounding molecules and the enrichment of tissue with oxygen. A comprehensive evaluation of such parameters was not carried out in this study considering the complex nature of creating an appropriate experimental model.

Destruction of the stromal part of tissue caused by PDT plays an essential role in the treatment process. Weak therapeutic effects are demonstrated in cases of stroma remaining untouched which is confirmed by the recovery of fluorescence intensity after PDT (Fig. 5b). A direct correlation between photobleaching and the effects of therapy was noticed.

Significant fluorescent signal reductions within the lesion area were observed in 42 patients after a single therapy session. This is a good prognosis and indicator for effective treatment. A slight decrease in PS concentration was registered in 28 cases that can be attributed to the insufficient vascular damage during PDT and possible protoporphyrin IX diffuse redistribution. It is permissible to increase the irradiation dose if the therapy proves to be weak but not without taking into account the pain threshold of patients.

6. Conclusion

Combined continuous-pulse irradiation mode for vulvar leukoplakia PDT with a 0.5% 5-ALA application was investigated for the first time in this study. Phototheranostic sessions were conducted in 70 patients, who were scheduled to undergo three courses of treatment that consisted of 3 therapy sessions every other day with a one-month interval between courses. In 7 patients, significant beneficial effects such as itch relief and visual change regression were observed after the first course of therapy. A direct correlation between PS photobleaching and the patients' ability to heal was also recorded. In no case did the disease transition into a malignant form while patients were closely monitored before, during, and after treatment.

Combined continuous-pulse mode of PDT exposure accompanied by topical 0.5% 5-ALA application is a promising approach towards treatment of pathological vulvar diseases and an effective way to prevent the development of cancerous vulvar transformations against the background of illness. Described in this study technique is characterized by methodological simplicity of execution, selective influence on

pathological tissues, the absence of adverse reactions and complications, and excellent cosmetic effect. Also, improved PDT treatment approach is less time-consuming and provides with an opportunity to carry out theranostics daily in 20 patients.

Conflict of interest

The authors declare that they have no conflict of interest.

Ethical approval

"All procedures performed in studies involving human participants were by the ethical standards of the institutional and national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards."

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References

[1] G.L. Tasker, F. Wojnarowska, Lichen sclerosus, Clin. Exp. Dermatol. (28) (2003) 128–133, https://doi.org/10.1046/j.1365-2230.2003.01211.x.
[2] A.T. Goldstein, S.C. Marinoff, K. Christopher, M. Srodon, Prevalence of vulvar lichen sclerosus in a general gynecology practice, J. Reprod. Med. 50 (7) (2005) 477–480.
[3] Lien N. Hoang, Kay J. Park, Robert A. Soslow, Rajmohan Murali, Squamous precursor lesions of the vulva: current classification and diagnostic challenges, Pathology 48 (4) (2016) 291–302, https://doi.org/10.1016/j.pathol.2016.02.015.
[4] Z. Knio, M. Kurban, O. Abbas, Lichen sclerosus: clinicopathological study of 60 cases from Lebanon, Int. J. Dermatol. 55 (10) (2016) 1076–1081.
[5] P. Vieira-Baptista, J. Lima-Silva, J. Cavaco-Gomes, J. Bieres, J. Martinez-de-Oliveira, What differentiates symptomatic from asymptomatic women with lichen sclerosus, Gynecol. Obstet. Invest. 79 (4) (2015) 263–268, https://doi.org/10.1159/000367788.
[6] L.J. Burrows, H.A. Shaw, A.T. Goldstein, The vulvar dermatoses, J. Sex. Med. 5 (2) (2008) 276–283, https://doi.org/10.1111/j.1743-6109.2007.00703.x.
[7] S. Mashayekhi, C. Flohr, F.M. Lewis, The treatment of vulval lichen sclerosus in prepubertal girls: a critically appraised topic, Br. J. Dermatol. 176 (2) (2017) 307–316, https://doi.org/10.1111/bjd.15202.
[8] K. Olek-Hrab, D. Jenerowicz, A. Osmola-Mankowska, A. Polanska, E. Teresiak-Mikolajczak, Selected vulvar dermatoses, Ginekol. Pol. 84 (11) (2013) 959–965, https://doi.org/10.17772/gp/1667.
[9] Y. Gu, L. Zhu, X. Li, et al., Surgical treatment of usual type vulvar intraepithelial neoplasia: a study at three academic hospitals, Chin. Med. J. 127 (2014) 784–786, https://doi.org/10.3760/cma.j.issn.0366-6999.20131816.
[10] C. Westermann, A. Fischer, A. Clad, Treatment of vulvar intraepithelial neoplasia with topical 5% imiquimod cream, Int. J. Gynaecol. Obstet. 120 (2013) 266–270, https://doi.org/10.1016/j.ijgo.2012.09.020.
[11] Chengzhi Li, Duhong Bian, Wenzhi Chen, Chunliang Zhao, Niangang Yin, Zhibiao Wang, Focused ultrasound therapy of vulvar dystrophies: a feasibility study, Obstet. Gynecol. 104 (5:1) (2004) 915–921, https://doi.org/10.1097/01.AOG.0000139835.34168.b0.
[12] V. Dunaevskaya, D. Tzerkovsky, Photodynamic therapy in vulvar dystrophy treatment, Arch. Clin. Biomed. Res. 2 (2) (2018) 42–51, https://doi.org/10.26502/acbr.5017040.
[13] V.B. Loschenov, V.I. Konov, A.M. Prokhorov, Photodynamic therapy and fluorescence diagnostics, Laser Phys. 10 (6) (2000) 1188–1207.
[14] A.A. Stratonnikov, N.E. Edinac, D.V. Klimov, K.G. Linkov, V.B. Loschenov, E.A. Luckjanets, G.A. Meerovich, E.G. Vakulovskaya, The control of photosensitizer in tissue during photodynamic therapy using absorption spectroscopy, SPIE 2924 (49) (1996) 1–12, https://doi.org/10.1117/12.260790.
[15] A.A. Stratonnikov, G.A. Meerovich, V.B. Loschenov, Photobleaching of photosensitizers applied for photodynamic therapy, SPIE 3909 (2000) 81–91, https://doi.org/10.1117/12.373885.
[16] A.Yu. Douplik, A.A. Stratonnikov, V.B. Loschenov, V.S. Lebedeva, V.M. Derkacheva, A.I. Vitkin, V.D. Rumyanceva, S.G. Kusmin, A.F. Mironov, E.A. Lukyanets, Study of photodynamic reactions in human blood, J. Biomed. Opt. 5 (3) (2000) 338–349, https://doi.org/10.1117/1.430005.
[17] A.P. Castano, T.N. Demidova, M.R. Hamblin, Mechanisms in photodynamic therapy: part two-cellular signaling, cell metabolism, and modes of cell death, Photodiagn. Photodyn. Ther. 2 (1) (2005) 1–23, https://doi.org/10.1016/S1572-1000(05)00030-X.
[18] M. Ascencio, P. Collinet, M. Cosson, D. Vinatier, S. Mordon, The place of photodynamic therapy in gynecology, Gynecol. Obstet. Fertil. 35 (11) (2007) 1155–1165, https://doi.org/10.1016/j.gyobfe.2007.07.035.
[19] R. Sroka, H. Stepp, G. Hennig, G.M. Brittenham, A. Ruehm, L. Lilge, Medical laser application: translation into the clinics, J. Biomed. Opt. 20 (6) (2015) 061110, , https://doi.org/10.1117/1.JBO.20.6.061110.
[20] A.S. Sharova, Yu.S. Maklygina, G.M. Yusubalieva, I.A. Shikunova, V.N. Kurlov, V.B. Loschenov, Sapphire implant based neuro-complex for deep-lying brain tumors phototheranostics, J. Phys. Conf. Ser. 945 (2017) 012009, , https://doi.org/10.1088/1742-6596/945/1/012009.
[21] E.G. Borisova, L.P. Angelova, E.P. Pavlova, Endogenous and exogenous fluorescence skin Cancer diagnostics for clinical applications, IEEE J. Sel. Top. Quant. Electron. 20 (2) (2014) 7100412.
[22] M.M. Kim, A.A. Ghogare, A. Greer, T.C. Zhu, On the in vivo photochemical rate parameters for PDT reactive oxygen species modeling, Phys. Med. Biol. 62 (5) (2017) R1.
[23] S. Bagdonas, L.W. Ma, V. Iani, R. Rotomskis, P. Juzenas, J. Moan, Phototransformations of 5-Aminolevulinic acid-induced protoporphyrin IX in vitro: a spectroscopic study, Photochem. Photobiol. 72 (2) (2000) 186–192.
[24] P. Hillemanns, X. Wang, S. Staehle, et al., Evaluation of different treatment modalities for vulvar intraepithelial neoplasia (VIN): CO2 laser vaporization, photodynamic therapy, excision, and vulvectomy, Gynecol. Oncol. 100 (2) (2006) 271–275, https://doi.org/10.1016/j.ygyno.2005.08.012.
[25] A.F. Taub, Photodynamic therapy: other uses, Dermatol. Clin. 25 (2007) 101–109, https://doi.org/10.1016/j.det.2006.09.007.
[26] A. Olejek, K. Steplewska, A. Gabriel, I. Kozak-Darmas, A. Jarek, S. Kellas-Sleczka, F. Bydlinski, K. Sieron-Stoltny, S. Horak, A. Chelmicki, A. Sieron, Efficacy of photodynamic therapy in vulvar lichen sclerosus treatment based on immunohistochemical analysis of CD34, CD44, myelin basic protein, and Ki67 antibodies, Int. J. Gynecol. Cancer 20 (5) (2010) 879–887, https://doi.org/10.1111/IGC.0b013e3181d94f05.
[27] Tomasz Biniszkiewicz, Anita Olejek, Iwona Kozak-Darmas, Aleksander Sieron. Therapeutic effects of 5ALA-induced photodynamic therapy in vulvar lichen sclerosus, Photodiagn. Photodyn. Ther. 2 (2005) 157–160, https://doi.org/10.1016/S1572-1000(05)00062-1.
[28] P. Hillemanns, M. Untch, C. Dannecker, et al., Photodynamic therapy of vulvar intraepithelial neoplasia using 5-aminolevulinic acid, Int. J. Cancer 85 (5) (2000) 649–653, https://doi.org/10.1002/(SICI)1097-0215(20000301)85:5<649::AID-IJC9>3.0.CO;2-E.
[29] E.V. Filonenko, L.G. Serova, A case report of successful photodynamic therapy of the patient with severe vulvar dysplasia, Photodyn. Ther. Photo Diagn. 3 (4) (2014) 31–33.
[30] Michael H. Gold, Mitchel P. Goldman, 5-aminolevulinic acid photodynamic therapy: where we have been and where we are going, Dermatol. Surg. 30 (2004) 1077–1084, https://doi.org/10.1111/j.1524-4725.2004.30331.x.
[31] E.V. Filonenko, A.D. Kaprin, A.A. Raszhivina, A.N. Urlova, A.M. Nechipai, Fluorescence diagnostics of Colon Malignant and premalignant lesions using 5-Aminolevulinic acid, Int. J. Photoenergy (378673) (2014).
[32] L.A. Belyaeva, L.V. Adamyan, V.P. Kozachenko, A.A. Stratonnikov, E.F. Stranadko, V.B. Loschenov, Fluorescence diagnostics in oncological gynecology, Saratov Fall Meeting 2002: Optical Technologies in Biophysics and Medicine IV, Proceedings of The Society of Photo-Optical Instrumentation Engineers (SPIE) 5068 (2002) 55–60.
[33] K.N. Maloth, N. Velpula, S. Kodangal, M. Sangmesh, K. Vellamchetla, S. Ugrappa, N. Meka, Photodynamic therapy - a non-invasive treatment modality for precancerous lesions, J. Lasers Med. Sci. 7 (1) (2016) 30–36, https://doi.org/10.15171/jlms.2016.07.
[34] M. Fujino, Y. Nishio, H. Ito, T. Tanaka, X.K. Li, 5-Aminolevulinic acid regulates the inflammatory response and alloimmune reaction, Int. Immunopharmacol. 37 (2016) 71–78, https://doi.org/10.1016/j.intimp.2015.11.034.
[35] V.I. Ivanova-Radkevich, E.V. Filonenko, E.K. Slovokhodov, I.P. Smirnova, O.M. Kuznetsova, T.A. Lobaeva, Yu. Sh. Gushchina, Z.S. Khadasheva, The organization of clinical trials of photosensitizer, based on 5-aminolevulinic acid hexyl ester, Indian J. Sci. Technol. 9 (18) (2016) 1–7, https://doi.org/10.17485/ijst/2016/v9i18/93759.
[36] NIOPIK, State Scientific Center "NIOPIK"_Alasens, [ONLINE] Available at http://www.niopik.ru/products/pdt_and_fd/alasense/ . [Accessed 31 March 2018] (2018).
[37] P. Babilas, R. Knobler, et al., Variable pulsed light is less painful than light-emitting diodes for topical photodynamic therapy of actinic keratosis: a prospective randomized controlled trial, Br. J. Dermatol. 157 (2007) 111–117, https://doi.org/10.1111/j.1365-2133.2007.07959.x.
[38] A.A. Stratonnikov, V.B. Loschenov, D.V. Klimov, N.E. Edinac, V.A. Wolnukhin, I.A. Strashkevich, The spectral fluorescent properties of tissues in vivo with excitation in the red wavelength range, Optical Biopsies and Microscopic Techniques II 3197 (1997) 119–130, https://doi.org/10.1117/12.297959.
[39] A.A. Stratonnikov, V.S. Polikarpov, V.B. Loschenov, Photobleaching of endogenous Fluorochromes in tissues in vivo, SPIE 4241 (2000) 13–24, https://doi.org/10.1117/12.431555.
[40] S.Yu. Vasilchenko, M.S. Ivanova, S.N. Akhtjamov, Yu.S. Butov, S.G. Kuzmin, G.N. Vorozhtsov, Pulsed irradiation for photodynamic therapy of skin and Soft tissue diseases, Laser Med. 15 (2) (2011) 61.
[41] S.Yu. Vasilchenko, V.I. Makarov, V.B. Loshenov, R. Steiner, M.S. Ivanova, S.N. Akhtyamov, S.G. Kuzmin, E.A. Lukyanets, Fluorescence diagnostics and photodynamic therapy with the use of photosensitizers in molecular and nanoform in dermatology and transplantology, Materials of the Tenth International Conference "High Medical Technologies in XXI Century" 62 (2011).
[42] A.V. Ryabova, A.A. Stratonnikov, V.B. Loshchenov, Laser spectroscopy technique for estimating the efficiency of photosensitizers in biological media, Quant. Elec. (Woodbury) 36 (6) (2006) 562–568.