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Sublingual administration of 5-aminolevulinic acid for laser-induced photodiagnostics and photodynamic therapy of oral cavity and larynx cancers

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ABSTRACT

Background: The study aimed to develop a method for sublingual administration of 5-aminolevulinic acid to patients and evaluate its effectiveness in fluorescence diagnostics and photodynamic therapy of neoplasms of the oral cavity and larynx.
Methods: The boundaries of the neoplasms were established by the video-fluorescence diagnostics and clarified using spectral-fluorescent diagnosis before and after photodynamic therapy.
Results: The fluorescence diagnostics demonstrated a high accumulation of protoporphyrin IX, induced by sublingual administration of 5-aminolevulinic acid to patients before the photodynamic therapy and photobleaching of protoporphyrin IX in pathologically altered tissues after the photodynamic therapy. Glucose contained in the sublingual dose supports active transport of 5-ALA into the cells. It increases the PpIX accumulation in the cells, therefore improving the PD and PDT efficacy.
Conclusion: The study and the initially obtained results demonstrated the possibility and effectiveness of laser-induced photodiagnostics and photodynamic therapy with sublingual administration of 5-ALA to patients with premalignant lesions of the oral cavity and larynx. It can eliminate the threat of the transformation of these diseases into malignant tumors and prevent the need for surgical treatment.

1. Introduction

The relevant problems of modern oncology are timely diagnosis and effective treatment of precancerous conditions of the oral cavity and larynx. Also, early diagnosis of precancerous lesions and oral cancer is complex. It is often associated with an asymptomatic disease course and differentiating pathological tissue from relatively healthy tissue is often difficult. Precancerous diseases, often called 'oral potentially malignant diseases' (OPMD) are morphologically altered tissues with a significantly increased risk of cancer [1]. The lack of timely diagnosis and treatment of precancerous conditions contributes to their further transformation into a malignant tumor. Mortality from oral cancer in most countries is high, and the 5-year relative survival rate is about 50 % [2].

Among the methods of accurate diagnosis of the oral cavity and larynx neoplasm, the most promising are spectral diagnosis and fluorescence imaging [3–6]. These methods are based on the principles of interaction of light with tissue that contains endogenous fluorophores or an exogenously introduced photosensitizer (PS). Due to the pathological process, the cellular structure of the tissue changes. It affects the degree of photosensitizer accumulation and the processes of interaction of light with tissue [7].

The main treatments for neoplasms of the oral cavity and larynx are surgical removal, radiation therapy, with or without chemotherapy.

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However, these treatments have many side effects, such as pain, cosmetic defects, bacterial and fungal infections, taste changes, difficulty eating, hair loss, and others [8]. Photodynamic therapy (PDT) is an alternative treatment for malignant neoplasms [9]. The PDT principle consists of administering PS to the patient, which accumulates mainly in the tumor tissue. Then the tissue is irradiated with light at a wavelength that corresponds with one of the PS absorption wavelengths. As a result of these actions, highly reactive singlet oxygen ($^1O_2$) is generated. It destroys tumor cells as a result of a complex cascade of chemical, biological, and physiological reactions. Chlorin-type [10,11] and porphyrin-type PS [12,13] are most often used for the diagnosis and treatment of neoplasms of the oral cavity and larynx.

The porphyrin-type PS 5-aminolevulinic acid (5-ALA) was used for PDT in this study. 5-ALA induces protoporphyrin IX (PpIX) in biotissue cells. It is a fluorescent precursor in the heme biosynthetic pathway. PpIX accumulates in the tumor over several hours and maintains its high accumulation for 1–2 days. In healthy cells, PpIX is rapidly converted to photoinactive heme. As a result of PpIX accumulation, a high fluorescence of the tumor is observed in comparison to the surrounding normal tissues [14].

PpIX has an absorption maximum at $\lambda = 404$ (Soret band), 503, 536, 576, and 633 nm. Excitation at any of these wavelengths causes intense fluorescence in the red range up to $\lambda = 720$ nm with a maximum at 635 nm [15]. PpIX has a low molecular weight, a short phototoxicity period (24–48 h), good tissue penetration, and a high singlet oxygen yield.

The method of PS administration into the patient significantly affects the degree of PS accumulation in the tumor tissue.

The main methods of 5-ALA administration into the patient are topical, oral, instillation and inhalation. The method of 5-ALA administration depends on the nature of the diagnostic study and the localization of the pathological process.

The local administration method includes applying an ointment or an aqueous solution based on 5-ALA to the pathologically altered area [16]. Usually, to obtain a 5, 10, or 20 % ointment, 0.125, 0.25 or 0.5 g of 5-ALA, respectively, is dissolved in 0.6 mL of a solution containing 0.9 % sodium chloride, 4% ethylenediaminetetraacetic acid sodium salt, and 2% dimexide, then mixed with 1.2 g of lanolin and 0.2 g of petroleum jelly. Photodiagnosis (PD) and PDT of patients are carried out 4–6 h after applying the ointment.

Oral administration of 5-ALA for PDT is a safe and effective procedure [17]. The recommended concentration of 5-ALA can vary from 20 to 60 mg/kg. PD and PDT of patients are performed 2–6 h after oral administration. During surgery or laparoscopy, this interval from the solution administration to the anesthesia administration is almost halved.

The method of intravesical instillation of a sterile 5-ALA solution is used for bladder cancer PDT [18]. Usually, to obtain a 1.5 or 3% solution for instillation, 0.75 g or 1.5 g of 5-ALA, respectively, is dissolved in 50 mL of sterile 5% sodium bicarbonate solution. PD and PDT of patients are performed 1.5–2 h after instillation.

Inhaled administration of 5-ALA is the preferred method of administration for the treatment of bronchial neoplasms. Usually, 1 or 1.5 g of 5-ALA is dissolved in 10 or 25 mL of 5% sodium bicarbonate solution, respectively, to obtain a 10 % sterile solution for inhalation. PD and PDT are carried out 1.5–2 h after inhalation.

Sublingual administration of 5-ALA (Alasens) to patients is investigated in this paper. The novelty of the research consists in the possibility of using a new method of 5-ALA administration for neoplasms of the oral cavity and larynx.

PDT with the sublingual administration of 5-ALA has several advantages compared to other methods of administration:

1 Medical and diagnostic procedures in patients can be carried out independently of the tumor location.
2 Glucose contained in the sublingual dose supports the active transport of 5-ALA into cells. It promotes an increase in the PpIX accumulation in cells [19], and, accordingly, an increase in the efficiency of PD and PDT.
3 The possibility of aspiration during the anesthesia administration existing with oral administration is avoided.

Table 1
Clinical characteristics.

Case	Age	Diagnosis	Investigated zone	Power density, W/cm^2	Energy dose, J/cm^2
1	50	Papillomatosis of the larynx	Laryngeal mucosa	0.32	
2	65	Papillomatosis of the oral mucosa		0.32	200
3	46			0.05	
4	73			0.05	
5	31			0.26	100
6	60			1.05	150
7	58			1.07	100
8	30	Oral leukoplakia	Oral mucosa	0.26	100
9	57			0.12	150
10	54			0.07	150
11	61			0.28	100
12	60			0.04	150
13	67			0.55	100
14	64			0.99	150
15	55			0.76	150
16	62	Oral dysplasia		0.27	100

The study aimed to develop a method for sublingual administration of 5-ALA to patients and to evaluate its effectiveness in fluorescent diagnosis and photodynamic therapy of neoplasms of the oral cavity and larynx.

The following tasks were formed:

1 Development of a 5-ALA drug form for sublingual administration;
2 Assessment of the 5-ALA-induced PpIX accumulation in normal and pathological tissues of the oral cavity and larynx using spectral- and video-fluorescent methods of diagnosis before and after PDT;
3 Investigation of the possibility of PDT with sublingual administration of 5-ALA.

2. Materials and methods

The study was carried out at the University Clinical Hospital No. 1 of the I.M. Sechenov First Moscow State Medical University (Sechenov University). The study protocol was approved by the Local Ethics Committee of Levshin Institute of Cluster Oncology. Each patient was informed about the treatment protocol and signed an agreement to participate in the clinical study.

Sixteen patients aged 56 ± 12 with precancerous diseases of the oral cavity and larynx were included in the study. Table 1 shows the age of the patient, clinical diagnosis, investigated zone, power density, and energy dose.

Before the treatment, the patients took a previously prepared 5-ALA solution at a concentration of 20 mg/kg sublingually. The solution is a 5-ALA sugar syrup in liquid form. Sublingual administration of the 5-ALA includes locating and keeping the 5-ALA sugar syrup into hyoid cavity for 10−15 min. We have established the optimal duration of receiving the entire volume sublingual solution based on patient feelings. The sublingual solution was sequentially administered into the hyoid cavity in small portions (approximately 1 mL). The keeping time of one solution portion was about 2 min. There was no sublingual rinse. The volume of the sublingual solution was not the same for each patient, as the weight of patients was different. The sublingual solution had a slightly sour taste. Patients did not swallow the solution.

Sublingual drug delivery through the oral mucosa is a promising alternative to oral administration. Drugs are quickly absorbed into the

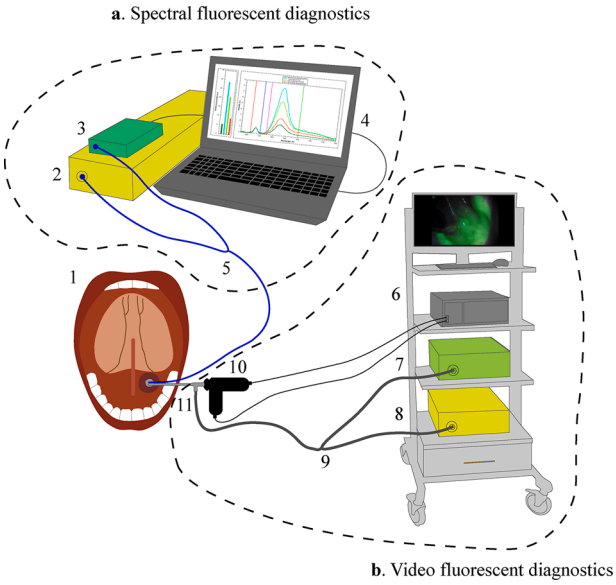

Fig. 1. The scheme of the study of pathological tissue. **a.** Spectral-fluorescent diagnosis; **b.** Video-fluorescent diagnosis; **1.** The oral cavity of a patient with neoplasm; **2.** Laser with a wavelength of 632.8 nm; **3.** Fiber spectrometer; **4.** Laptop; **5.** Y-shaped fiber cable for spectral fluorescence diagnosis; **6.** Laptop; **7.** White light source; **8.** Laser with a wavelength of 635 nm; **9.** Y-shaped optical fiber for video- and -spectral diagnosis; **10.** Universal device for recording backscattered and fluorescent radiation, equipped with black-and-white and color cameras; **11.** Endoscope.

oral mucosa through the blood vessels under the tongue, bypassing the extremely unfavorable environment in the gastrointestinal tract. This method of drug administration provides a fast pharmacological effect [20].

The drug penetrates the laryngeal mucosa by systemic absorption from the oral cavity and not by physical contact. The boundary between tumor and normal tissue can become blurred when 5-ALA is applied topically, making it difficult to diagnose and treat the tumor [21].

The total volume of sublingual solution per a patient was calculated with the formulas below.

$$m_A = c_A \times m_p \tag{1}$$

$$V = \frac{m_A}{k} \tag{2}$$

where m_A *[mg]* is a 5-ALA mass per sublingual solution for a patient; $c_A = 20$ *[mg/kg]* is a 5-ALA concentration per sublingual solution for a patient; m_P *[mg]* is a mass of a patient; V *[mL]* is a 5-ALA volume of sublingual solution per a patient; $k = 200$ *[mg/mL]* is a 5-ALA concentration per 1 mL of sublingual solution.

The sublingual solution was prepared based on crystalline sugar, the density of which is $\rho = 1.586$ *[g/mL]*. Sugar and water were diluted in a 3:1 ratio.

$$V = V_{H_2O} + (f \times V_{H_2O}) \tag{3}$$

$$V_S = f \times V_{H_2O} \tag{4}$$

$$m_s = \rho \times V_S \tag{5}$$

where $f = 1.89$ is a sugar-to-water ratio; V_S is a volume of sugar syrup, and V_{H_2O} is a volume of water for preparing a stock solution; m_s is a mass of sugar.

For example, the total volume of sublingual solution is 7 mL when 5-ALA is administered at a concentration of 20 mg/kg to a patient weighing 70 kg. The 5-ALA concentration for each patient was the same and amounted to 20 mg/kg.

The 5-ALA solution with a pH of 2–3 was alkalized to a pH of 5–6, as the sublingual application of the highly acidic solution could lead to burns of the oral mucosa, esophagus, and stomach. A $NaHCO_3$ solution diluted 1:10 was used for alkalization.

It was established experimentally that one sublingual dose requires N g of 5-ALA and N ml of sodium bicarbonate solution. The remaining liquid volume obtained from Eq. (3) is water, which is added to the 5-ALA solution before neutralization. Before the diagnosis and treatment, patients were informed about the side effects of the solution under investigation, the sequence of the study, and the rules for keeping the light regime after the medical procedure. Patients were also asked about chronic diseases and allergic reactions to the components of the solution under investigation to avoid adverse treatment outcomes.

Diagnostic methods allow determining the boundaries of the tumor and healthy tissue (Fig. 1). Spectral-fluorescent diagnosis was carried out using a LESA-01-BIOSPEC fiber spectrometer with PS fluorescence excitation by a helium-neon laser (λ =632.8 nm, Pmax =15 mW). Radiation with a wavelength of 632.8 nm makes it possible to diagnose deep-seated tumors. External neoplasms can be diagnosed using radiation in the blue range of the spectrum.

Delivery of laser radiation to the surface of the investigated area and subsequent registration of fluorescent radiation was carried out using a Y-shaped fiber cable. The fluorescence spectra were recorded before and 2 h after the administration of 5-ALA, and after the PDT.

Spectral-fluorescent diagnosis was conducted using a fiber spectrometer. It made it possible to obtain fluorescence spectra at various points of the investigated zone. The fluorescence spectra were normalized to the laser line. Then the fluorescence index was calculated. The method for calculating the fluorescence index has been previously reported [22]. The registration and processing of the spectra were carried out using the Uno Momento program developed at the Laboratory of Laser Biospectroscopy of the GPI RAS.

Video-fluorescent diagnosis was performed before PDT to visualize the boundaries of the tumor and healthy tissue. The fluorescent images of the investigated zones were recorded using a two-channel video system, the diagram of which is shown in Fig. 1.

The video-system consists of the following components:

- A white light source allows observing the investigated zones in color mode;
- A laser radiation source with a wavelength of 635 nm that excites PS fluorescence in biological tissues and makes it possible to study the investigated zones in a fluorescent mode (black-and-white image);
- Y-shaped optical fiber provides transport of light from light sources to the surface of biological tissue;
- A universal device equipped with black-and-white and color video cameras registers backscattered and fluorescent radiation;
- An endoscope makes it possible to investigate the zones of interest.

PS accumulation in tissues is assessed by visual analysis of the intensity of the fluorescent signal in the zones of interest in the mode of combined black-and-white and color images. The fluorescent intensity of the investigated zone is proportional to the brightness of the pixels composing the image of this zone.

The software for the video-system was developed at the Laboratory of Laser Biospectroscopy of the GPI RAS. It allows registering images simultaneously in black-and-white, color, and combined modes [22]. Studies [23,24] have demonstrated that this video-system is efficient and easy to use. It is the basis for further research.

The pathologically altered zones were determined as a result of fluorescent diagnosis. These zones were sequentially subjected to PDT using a semiconductor laser (λ =635 nm, P_{max} = 1.5 W). Doses of light energy were 100–200 $\frac{J}{cm^2}$. The average power density was 0.4 W/cm^2. Laser radiation was delivered to the pathologically altered zone using a cylindrical diffuser with a length of 15 mm for Case 1 and an end-face

Table 2
Results of spectral- and video-fluorescence diagnosis.

Case	Localization	Fluorescent index, a.u. Square 650–750/Square 625–645		Contrast index, a.u.	
		Before PDT 632.8 nm	After PDT 632.8 nm	Before PDT 635 nm	After PDT 635 nm
1	Laryngeal mucosa	33	7	–	–
2		18	6	–	–
3		15	7	–	–
4		11	6	–	–
5		–	–	90	07
6		–	–	80	09
7		–	–	64	05
8		–	–	65	05
9	Oral mucosa	–	–	77	09
10		–	–	32	05
11		–	–	31	05
12		–	–	50	10
13		–	–	27	08
14		–	–	60	10
15		–	–	27	09
16		–	–	19	09

optical fiber with a numerical aperture NA = 0.22 for Cases 2–16. Local anesthesia with lidocaine was used if patients had pain in the irradiated area.

3. Results

All patients included in the study tolerated the sublingual 5-ALA administration quite well. No side effects or allergic reactions were observed. However, slight pain in the area under investigation was observed during PDT.

Spectral-fluorescent diagnosis was performed for Case 1–4, video-fluorescent diagnosis was carried out for Case 5–16 (Table 2).

Figs. 2 and 3 show the results of spectral diagnosis of laryngeal papillomatosis and oral leukoplakia, respectively. A laser with a wavelength of 632.8 nm was used to diagnose the larynx and oral cavity. The fluorescence spectra of pathologically altered tissues of the larynx and oral cavity before and after PDT in comparison with normal tissues were measured as a result of diagnosis.

Based on Fig. 2, the fluorescence level 2 h after the 5-ALA administration increased almost ten times relative to normal tissue. It indicates a high selectivity of PpIX accumulation in biological tissues of pathologically altered zones. The fluorescence level of the pathologically altered laryngeal tissue decreased sharply by almost four times after PDT due to PpIX photobleaching. The fluorescence spectra of the larynx mucosa were not measured before 5-ALA administration to minimize the number of interventions in the studied zones.

The fluorescence intensity of pathological zones of the oral mucosa (Fig. 3) 2 h after 5-ALA administration increased approximately four times compared with the fluorescence intensity before 5-ALA administration. The fluorescence intensity after PDT decreased by approximately four times.

The fluorescence intensity increased 2 h after the 5-ALA administration by almost eight times, and it decreased five times after PDT on average for all patients.

Fig. 4 shows images of the oral mucosa of a patient with mild dysplasia before and after PDT, obtained using a two-channel fluorescent video-system. Investigated zones are marked with a red dotted line in the image obtained in the combined mode (Fig. 4.1. c). These zones have strongly expressed fluorescence. They were subsequently subjected to PDT. The contrast indices before PDT in the investigated zones were 27 and 19 a.u. The contrast indices decreased after PDT to 10 and 09 a.u., respectively, and photobleaching of the PpIX accumulated in pathologically altered tissues occurred. The contrast index decreased approximately 8 times after PDT on average for all patients.

Fig. 5 shows images of the larynx mucosa of a patient with papillomatosis before PDT and six days after PDT, obtained with an endoscope. Multiple papillomas are visible on the mucosa of the patient's larynx before PDT, associated with obstruction of the airway. Multiple papillomas completely regressed after PDT. The surface of the mucosa is visibly leveled. This indicates successful treatment.

Tissue samples were taken from the investigated zones of each patient before and after PDT for morphological verification in the pathology laboratory. Fig. 6 shows the results of the histological investigation of the tissue obtained from patient 16 with mild oral dysplasia before and after PDT.

According to the results, tissue taken from the investigated zones of the oral mucosa of patient 16 before PDT showed mild dysplasia. The results of the histological investigation after PDT showed necrosis of the surface layers. There were no signs of dysplasia with intact epithelial stratification in the integumentary epithelium after PDT.

Positive dynamics of treatment was noted in patients with papillomatosis of the larynx and oral cavity. Leukoplakia and oral dysplasia were not detected after PDT. Repeated PDT was required for two patients with leukoplakia. As a result of PDT, leukoplakia was eliminated.

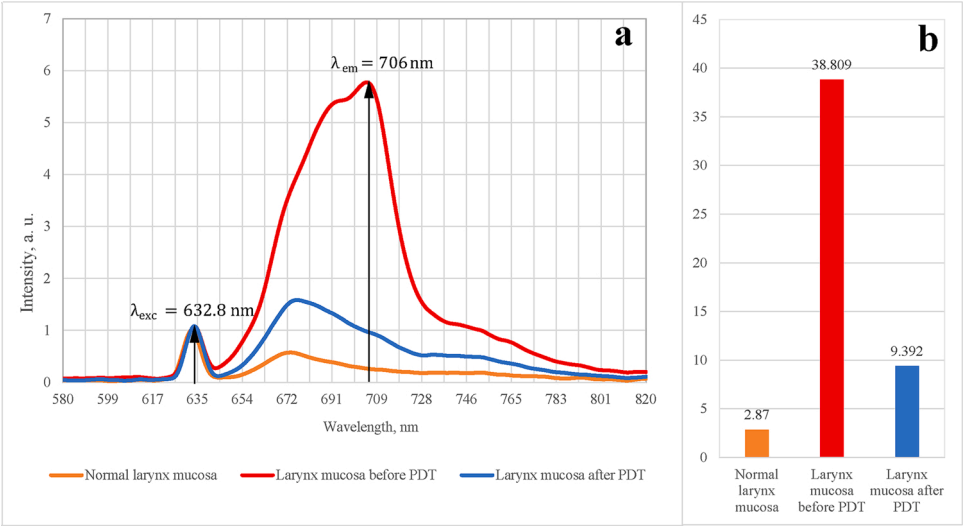

Fig. 2. Results of spectral fluorescent diagnosis of patient 1 with larynx papillomatosis. **a**. Fluorescence spectra of the normal and pathological larynx mucosa before and after PDT; **b**. Fluorescence indices.

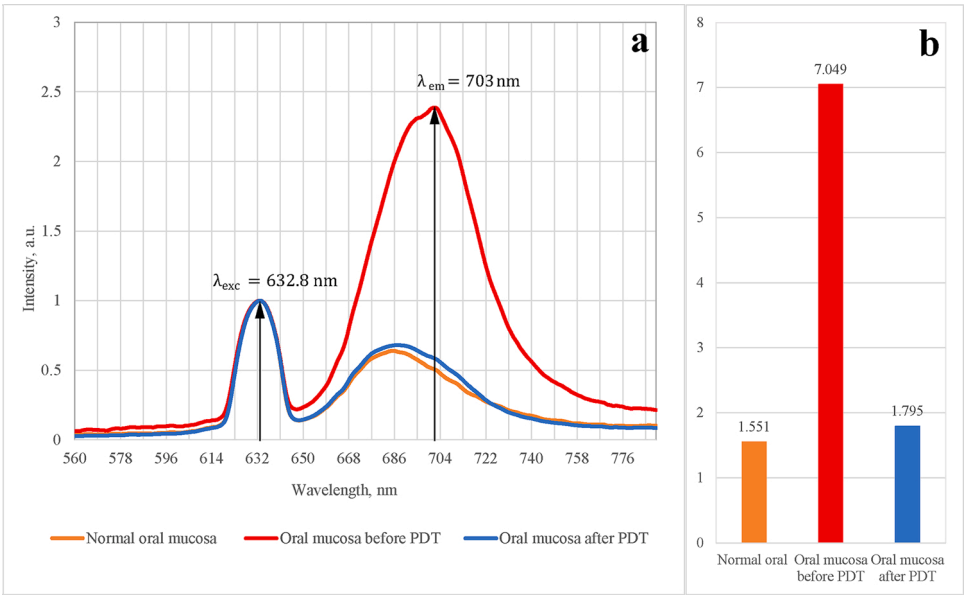

Fig. 3. Results of spectral fluorescent diagnosis of patient 3 with oral leukoplakia. **a**. Fluorescence spectra of the normal and pathological oral mucosa before and after PDT; **b**. Fluorescence indices.

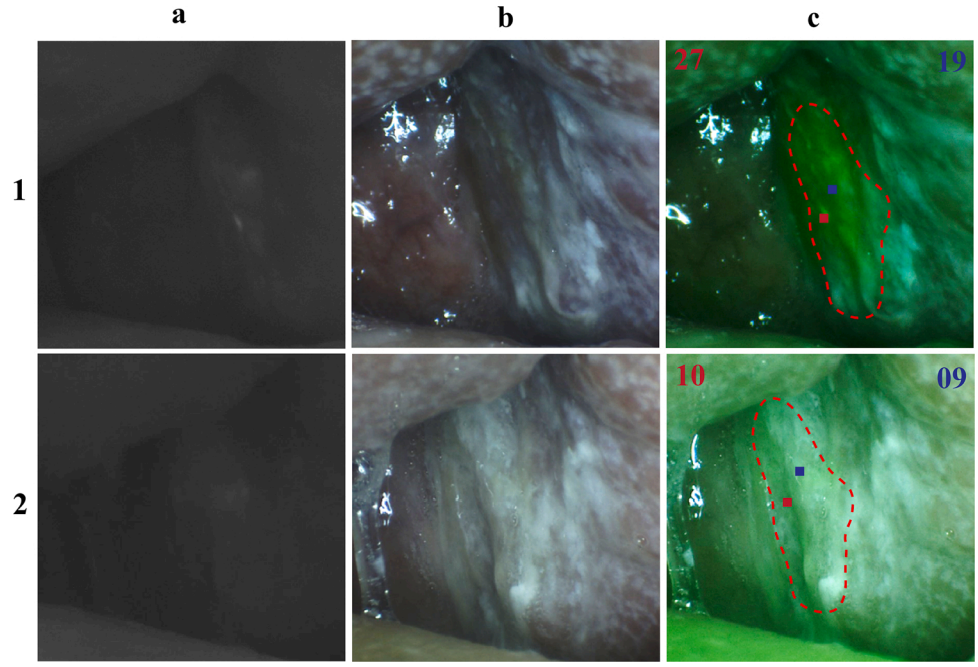

Fig. 4. Images of the oral mucosa of a patient 16 with mild dysplasia. **1**. Before PDT; **2**. After PDT; **a**. Fluorescent mode; **b**. Color mode; **c**. Combined mode (the fluorescence index is displayed in the corners of the image).

The results of the study indicate that PDT is effective for the treatment of premalignant lesions of the oral cavity and larynx.

4. Discussion

PDT with topical application of 5-ALA is a widely approved therapy for the treatment of superficial lesions, such as actinic keratoses, squamous cell carcinoma in situ, basal cell carcinomas [25] and neoplasms of the oral cavity [26]. PDT has been reported to be effective for precancerous diseases and cancer of the oral cavity at a depth of 0.1–1.3 mm from the surface layer of the lesion [27]. This study showed that 5-ALA-induced PpIX was localized to the basal layer (0.5–0.8 mm thick) after topical application. The effect of ALA-PDT was observed throughout the entire depth of the epithelium of the oral mucosa. It is suggested that ALA-PDT may be more effective for lesions confined to the epithelial layer of the mucosa. Tissue permeability for 5-ALA can result in a high PpIX yield in the basal layer. However, the metabolic activity of the basal layer can be involved. Disadvantages of topical administration of 5-ALA include pain that occurs when light is applied to the sensitized lesion.

The main advantages of oral administration of 5-ALA are ease-of-use and low light sensitivity for no more than 24 h [28]. Oral administration of 5-ALA is widely used to treat bladder cancer [17,29], as well as brain and spinal cord tumors [30–32]. Fluorescent diagnosis with oral

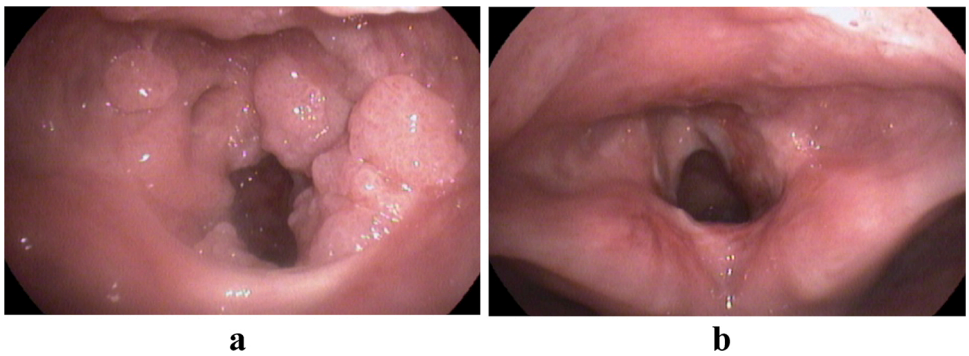

Fig. 5. Endoscopic image of the larynx mucosa of a patient 1 with papillomatosis. **a**. Before PDT; **b**. 6 days after PDT.

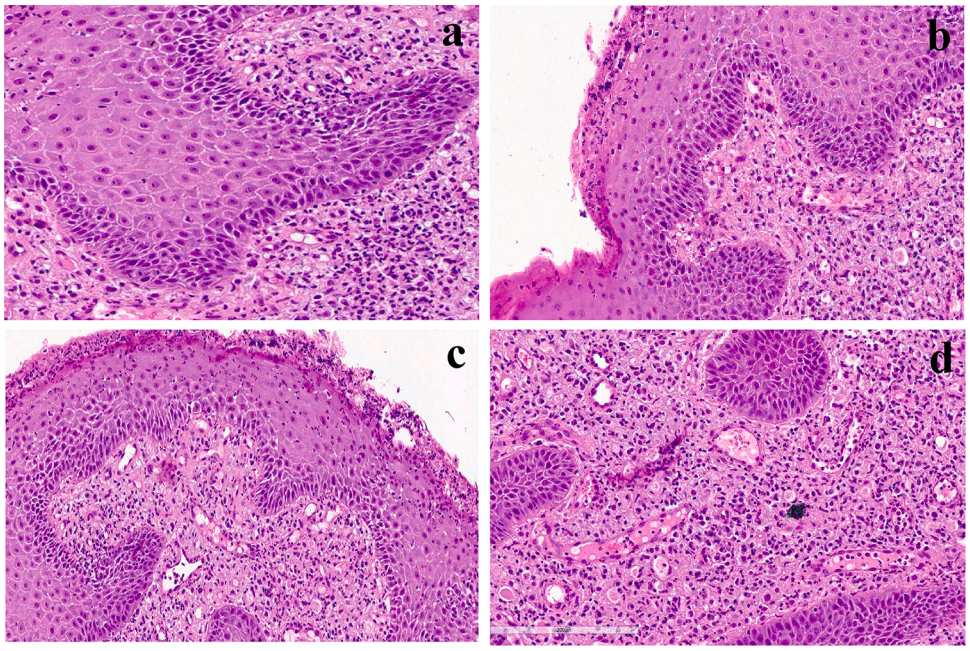

Fig. 6. Results of histological investigation. **a**. Before PDT; fragment of the mucous membrane of patient 16 with mild dysplasia (mild flat epithelial neoplasia); **b**. After PDT; acanthosis, necrosis of the superficial layers of the stratified squamous epithelium; **c**. After PDT; epithelial layer with preserved stratification, necrosis of the superficial layers of the stratified squamous epithelium; **d**. After PDT; angiomatosis of the subepithelial base with fibrin thrombi in the lumen of the capillaries. Hematoxylin-eosin staining.

administration of 5-ALA has shown successful results in detecting peritoneal metastases of ovarian cancer [33], and lymph node metastases in gastrointestinal malignant neoplasms [34]. However, this method of 5-ALA administration has several disadvantages. During surgery or laparoscopy, reducing the interval (almost twice) from the 5-ALA administration to the anesthesia administration has a potential risk of aspiration complications, especially in patients with impaired gastric evacuation. Oral administration of the solution increases the acidity of the already acidic environment of the stomach. It can increase chronic inflammation. Administration of antacids and antisecretory drugs, as well as complete or incomplete stomach tumor lesions, may interfere with the 5-ALA absorption. Unstable hemodynamics are possible after the oral administration of 5-ALA in patients with cardiovascular pathologies [28].

PDT with intravesical administration of 5-ALA is an effective treatment for bladder cancer with minimal side effects such as dysuria and irritation during urination. Intravesical instillations of 5-ALA are not associated with cutaneous photosensitivity compared to oral administration. However, the results obtained with intravesical instillations of 5-ALA are worse than with oral administration [35].

The advantages of the inhalation method are the absence of skin phototoxicity and a reduction in treatment costs. In addition, photodynamic therapy and inhaled bronchoscopy are relatively safe methods of diagnosis and treatment. The main disadvantage of this method is the high percentage of false positives results [36]. It is because false-positive inflammatory lesions often show increased PpIX fluorescence [37]. Nevertheless, this method is an excellent addition to the usual diagnostic approaches, especially for detecting early lesions in operated patients [36].

The described methods of 5-ALA administration for PDT of laryngeal and oral neoplasms are widely accepted. However, there are significant disadvantages, such as pain and irritation of the mucosa, low PS penetration, false-positive results, etc. Sublingual administration of 5-ALA excludes these side effects without compromising the accuracy of diagnosis and effectiveness of treatment. This method of 5-ALA administration can make it possible to carry out the necessary therapeutic and diagnostic procedures in patients, independent of tumor localization. At the same time, the possibility of aspiration during anesthesia administration, which exists during oral administration, is completely negated. According to the results of spectral- and video-fluorescent diagnosis, a high level of fluorescence intensity before PDT indicates this. The authors of the reference [19] claim the presence of glucose in the photosensitizing dose supported the active transport of 5-ALA into the cell. 5-ALA is converted to PpIX in the metabolic pathway. Besides, glucose has been described to inhibit the induction of aminolevulinic acid synthase and ferrochelatase [19]. Inhibition of ferrochelatase can reduce the conversion of PpIX to heme, which would lead to increased accumulation of PpIX in the cell [19,38,39]. Sufficient

production of PpIX in abnormal cells can increase the effectiveness of 5-ALA-PDT.

5. Conclusion

A sublingual method of 5-ALA administration to patients with precancerous diseases of the oral cavity and larynx was developed. The sublingual method of administration has significant advantages compared with other methods. They include low invasiveness, low skin phototoxicity, exclusion of aspiration during anesthesia administration. Therapeutic manipulations can be performed independently of the tumor location. Glucose contained in the sublingual solution supports active transport of 5-ALA into the cells. It increases the PpIX accumulation in the cells, and therefore improves the PD and PDT efficacy. However, the sublingual administration of 5-ALA to patients with diabetes mellitus requires caution since the solution contains glucose.

The results of spectral-fluorescent diagnosis correlate well with the results of video-fluorescent diagnosis. Both diagnostic methods suggest that endogenous PpIX is induced in cells of human biological tissues after the sublingual administration of 5-ALA solution. The PpIX induction in the pathologically altered tissues was several times higher than in normal ones.

The study and the initially obtained results demonstrated the possibility and effectiveness of laser-induced PD and PDT with sublingual administration of 5-ALA to patients with premalignant lesions of the oral cavity and larynx. It can eliminate the threat of the transformation of these diseases into malignant tumors and save the patient from surgical treatment.

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