# MICROBIOLOGICAL STUDY OF THE EFFICIENCY OF ROOT CANAL TREATMENT WITH ER:YAG LASER

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## Abstract

Mechanical and drug treatment of the root canal are the most important components of the success of endodontic treatment. This work presents the results on the effectiveness of the root canal disinfection using an erbium laser in combination with a 17% solution of ethylenediaminetetraacetic acid (EDTA) in vitro and in clinical studies. An in vitro study was carried out on removed intact teeth infected with strains of Enterococcus faecalis, Streptococcus sanguinis, and Candida albicans. An experimental group of teeth was treated medically according to a standard protocol using 3% solution of sodium hypochlorite and 17% solution of EDTA followed by irradiation with erbium laser. The control group was treated similarly but without laser irradiation. The treatment effectiveness was evaluated by the reduction of CFU. After the treatment, in the control group after the mechanical and drug treatment, the CFU amount lowered by 4 times, while the complete sterilization was observed in the experimental group. The clinical studies included two groups of 35 patients each diagnosed with chronic periodontitis. In the experimental group of patients, at the final stage, treatment with erbium laser for 1 min using endodontic piece (40 mJ power, 2940 nm wavelength, 10 Hz pulse rate) and a 17% EDTA solution was performed before filling. Periodontitis treatment in the control group was carried out without the laser treatment. The control group of patients saw the reduction on CFU after the mechanical and drug treatment by 3–6 times, while the experimental group achieved the complete sterilization of the root canals. The obtained results prove that the modification of the root canal treatment by the inclusion of erbium laser irradiation is a promising direction in endodontics.

**Keywords:** erbium laser (Er:YAG), laser-activated irrigation, endodontic treatment, microbiological study, root canal.

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# МИКРОБИОЛОГИЧЕСКОЕ ИССЛЕДОВАНИЕ ЭФФЕКТИВНОСТИ ОБРАБОТКИ КОРНЕВОГО КАНАЛА ЭРБИЕВЫМ ЛАЗЕРОМ

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## Резюме

Механическая и медикаментозная обработка корневого канала – важнейшие составляющие успеха эндодонтического лечения. В статье представлены результаты исследования эффективности дезинфекционной обработки корневого канала 17%-ым раствором этилендиаминтетрауксусной кислоты (ЭДТА) в сочетании с эрбиевым лазером (Er:YAG) *in vitro* и в клинических исследованиях. Исследование *in vitro* выполнено на интактных удаленных зубах, инфицированных штаммами *Enterococcus faecalis*, *Streptococcus sanguinis* и *Candida albicans*. Опытную группу зубов обрабатывали медикаментозно по стандартному протоколу с использованием 3%-го раствора гипохлорита натрия и 17%-го раствора ЭДТА с последующим облучением эрбиевым лазером. Обработку контрольной группы зубов проводили аналогичным образом, но без облучения лазером. Эффективность обработки оценивали по уменьшению титра КОЕ. В контрольной группе титр КОЕ после механической и медикаментозной обработки снизился в 4 раза, а в опытной группе была достигнута полная стерилизация корневых каналов. В клинические исследования были включены две группы пациентов по 35 человек с диагнозом хронический пародонтит. В опытной группе пациентов на последнем этапе лечения проводили обработку корневых каналов эрбиевым лазером в течение одной минуты эндодонтической насадкой с энергией 40 мДж с длиной волны 2940 нм, при частоте импульса 10 Гц с 17%-ым раствором ЭДТА и обтурировали. В контрольной группе проводили лечение периодонтита без обработки лазером. В контрольной группе пациентов титр КОЕ после механической и медикаментозной обработки снизился в 3–6 раз, а в опытной группе была достигнута полная стерилизация корневых каналов. Полученные результаты доказывают, что модификация протокола обработки корневого канала излучением эрбиевого лазера является перспективным направлением в эндодонтии.

**Ключевые слова:** эрбиевый лазер (Er:YAG), лазерная обработка, эндодонтическое лечение, микробиологическое исследование, корневой канал.

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## Introduction

The main tasks of mechanical, medical treatment and shaping of the root canal which is convenient for the chosen technique of obturation are the removal of infected dentin and disinfection of the root canal.

The existing variety of root canal treatment techniques demonstrated the lack of a universal approach to these tasks.

The main antiseptic and lubricant is a 3-5% of sodium hypochlorite. Its disadvantage is the rapid inactivation during processing and heating, which requires constant irrigation of the treated area with a new portion of the solution. In addition, hypochlorite is not active against certain forms of microorganisms, for example, *Enterococcus faecalis*. To expand the antibacterial spectrum in modern protocols, it is often recommended to use chlorhexidine 2% or calcium hydroxide in the form of pastes for temporary filling for 10-14 days. After using of calcium hydroxide, repeated treatment of the root canal with a 3% sodium hypochlorite solution is recommended, followed by washing. In addition to that, before obturation, the root canal must be treated with a 17% solution of ethylenediaminetetraacetic acid (EDTA) to remove the smear layer. Then it is needed to disinfect the canal, dry and obturate. According to various authors, the antibacterial effectiveness of this technique is 50-70% [1-3]. An increase in the effectiveness of drug treatment can be achieved by activating solutions to penetrate deeper into the infected areas of the root canal system. For this, physical methods of solution activation are used: sound, ultrasonic, hydrodynamic and laser techniques.

Experimental and clinical developments on the use of laser technologies in dentistry and in particular in endodontics have been conducted since 1988 and to date, extensive clinical experience has been gained on the use of diode lasers in the protocol for endodontic root canal treatment [4-6]. Erbium laser radiation has been used in clinical dentistry as an alternative to the mechanical method of preparing hard tooth tissues since 1997 [7, 8]. The possibilities of using Er:YAG laser radiation (2.94 μm) including the treatment of caries and its complications, sealing fissures, resection of the apex of the roots in chronic periodontitis, as well as patchwork operations in surgical periodontics [8].

The effects of traditional using of laser in endodontics are ablation of tissue remains, destruction of bacteria, and removal of the smear layer [7, 8].

For that, the aim of this research was to study the effectiveness of disinfection of the root canal with an erbium laser (Er:YAG) after standard mechanical and drug treatment of the canal.

## Materials and methods

To study the effectiveness of mechanical and drug treatment of the root canal with an erbium laser, 20 intact single-rooted extracted teeth due to periodontal disease were selected for the study.

The teeth were disinfected and processed by the standard mechanical protocol (hand tools K-files, K-Reamer, H-files, machine tools profiles, M2) and drug treatment, using 3% sodium hypochlorite and 17% EDTA solution with passive ultrasonic for solutions activation. Then the teeth were sanitized in a 75% alcohol solution, washed with sterile distilled water and infected with strains of *Enterococcus faecalis, Streptococcus sanguinis* and *Candida albicans*. After incubation for 7 days, dentin scrapings were taken from the walls of the root canal. then, the teeth were prepared mechanically using hand and machine tools and medically using 3% sodium hypochlorite solution and 17% EDTA solution with passive ultrasonic activation of irrigants. After EDTA treatment, all teeth were randomly divided into two groups. In the first group, the root canals were treated with a 17% EDTA solution in combination with an erbium laser for 1 minute with an energy of 40 mJ, a wavelength of 2940 nm, and a pulse frequency of 10 Hz. The teeth of the second group were not treated with laser. Before medical treatment of the root canal and immediately after its completion, dentin scrapings were taken for microbiological examination. Dentin was taken from the walls of the root canal with a sterile H-file. Dentin samples from the root canal were placed in a transport medium. Sowing the contents of the root canal was carried out on solid nutrient media. Cultivated in an aerobic incubator (Binder, Germany) and a $CO_2$ incubator (Lamsystems, Russia) in accordance with the requirements for incubation conditions for various microorganisms. Quantitative assessment of the results of sowing was carried out according to the Gould method. Identification results were taken into account by MASS spectrometry (MALDI-TOF), Mylа-MC (BioMerieux, France).

To study the antibacterial properties of an erbium laser *in vivo*, 70 patients with a diagnosis of chronic peri-

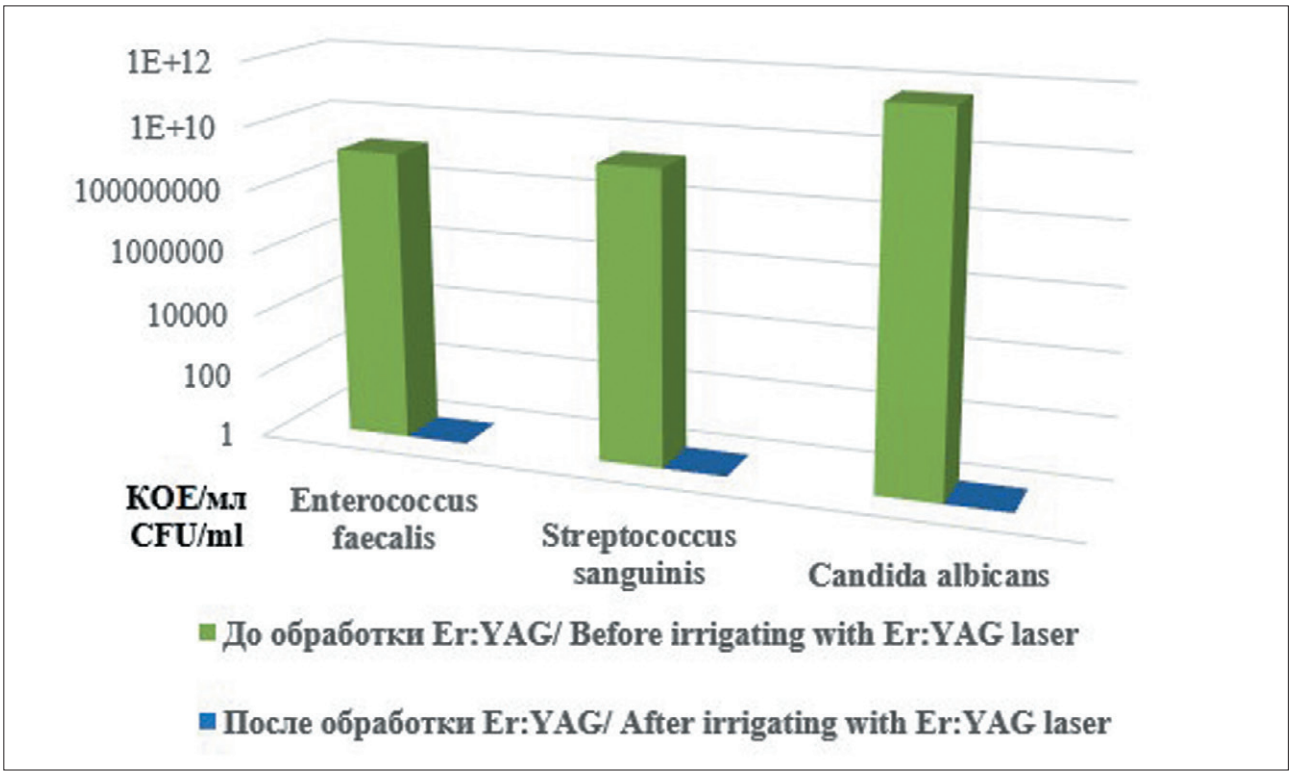


**Рис. 1.** Микробиологическое исследование в опытной группе на удаленных зубах с применением эрбиевого лазера в сочетании с 17%-ым раствором ЭДТА
**Fig. 1.** Microbiological study on group 1 extracted teeth using Er:YAG laser

odontitis (K04.5 chronic apical periodontitis. Apical granuloma) aged 35-60 years were selected. The test group consisted of 35 patients (19 women and 16 men). Endodontic treatment was performed according to standard methods. The operation field was cleaned with brushes with paste and 2% chlorhexidine solution and isolated with cofferdam. The old restoration was removed with sterile boron. Then the boron was replaced, the tooth cavity was opened, the root canals were mechanically and medically treated with manual and machine tools. Root canal irrigation was performed with 3% sodium hypochlorite with passive ultrasonic activation of the solutions. At the final stage, the canal was treated with a 17% solution of EDTA with an erbium laser for one minute using an endodontic tip, with an energy of 40 mJ, a wavelength of 2940 nm, and a pulse frequency of 10 Hz. A laser fiber was inserted into the orifice of the root canal without touching its walls. Root canal filling was performed at the same visit. The comparison group consisted of 35 (20 women and 15 men) patients who received the same root canal treatment without using an erbium laser.

## Results

In the test group of extracted teeth (the group treated with erbium laser), high titers of *Enterococcus faecalis, Streptococcus sanguinis* and *Candida albicans* strains were shown before drug treatment of the root canal. After mechanical and drug treatment of the root canals with 17% EDTA in combination with erbium laser, no colonies were recorded in all tested samples. This indicates the sterilization of the root canal (Fig. 1).

In the control group of extracted teeth (treated without erbium laser), a significant decrease in the titer of *Enterococcus faecalis, Streptococcus sanguinis and Candida albicans* strains was established. After mechanical and drug treatment of the root canals on the extracted teeth, a significant decrease by 4 times in the titer of colonies of the above microorganisms was recorded ($p < 0.05$) (Fig. 2)

According to that, this *in vitro* study showed that mechanical and drug treatment of the root canal reduced the number of microorganisms to titers $10^2$ and $10^3$ CFU/ml and high efficiency of root canal treatment was observed when applied erbium laser for one minute, with a power of 40 mJ, and a pulse frequency of 10 Hz.

To determine the efficacy of root canal treatment with an erbium laser in clinical trial, two groups of patients with a diagnosis of chronic periodontitis (K04.5 chronic apical periodontitis. Apical granuloma) aged 35 to 60 years were included in the study. At the stages of endodontic treatment, materials were collected for microbiological examination: before and after mechanical, drug treatment of the root canal, as well as after using erbium laser.

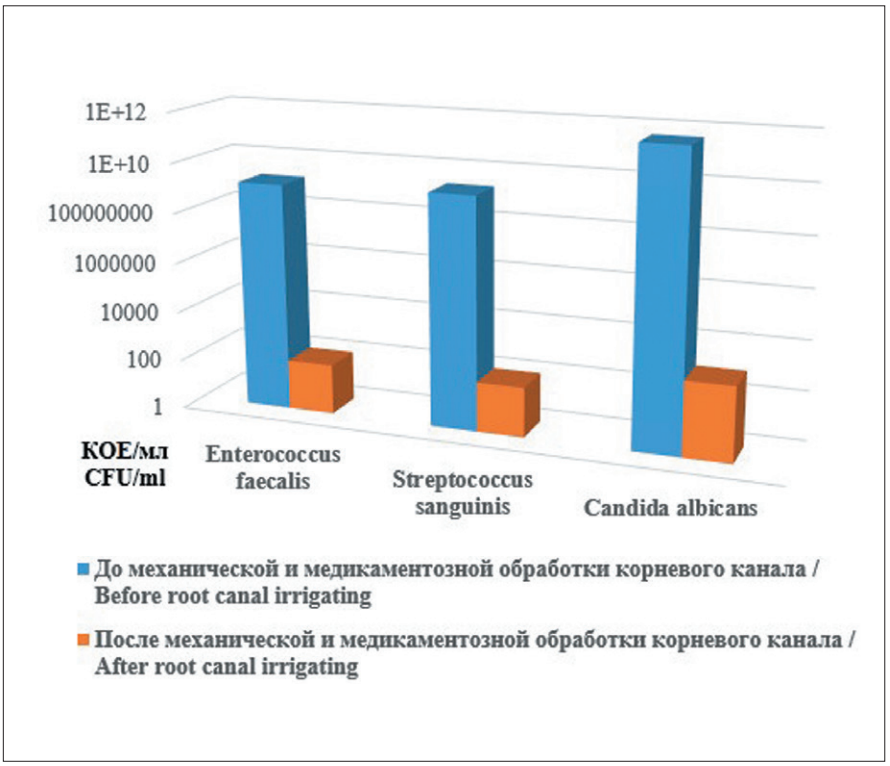


**Рис. 2.** Микробиологическое исследование в контрольной группе на удаленных зубах без применения эрбиевого лазера
**Fig. 2.** Microbiological examination in control group on extracted teeth without using ER:YAG laser

In the test group, high titers of *Enterococcus Haemolyticus, Staphylococcus epidermalis, Streptococcus mitis* and *Streptococcus mutans* from $10^5$ to $10^8$ cells/ml medium were observed immediately after mechanical treatment of the root canals (without drug treatment). After drug treatment of root canals, a significant decrease in the titer of microorganisms by four times to $10^2$ CFU/ml was recorded ($p < 0.05$). The data was presented in Table 1. In root canals, treated with erbium laser in combination with 17% EDTA in all tested samples, colonies growth was not recorded. This indicates sterilization of the root canal.

In the control group, patients received a similar protocol for endodontic treatment and materials were collected before and after drug treatment of the root canal. The data was presented in Table 2. Before root canal treatment, as in the main group, high titers of *Enterococcus Haemolyticus, Staphylococcus epidermalis, Streptococcus mitis* and *Streptococcus mutans* from $10^5$ to $10^8$ CFU / ml of medium were observed. After drug treatment, a significant decrease in the titer of microorganisms to $10^2$-$10^3$ CFU / ml ($p < 0.05$) was detected and only *Streptococcus mitis* growth was not recorded.

## Discussion

The quality of drug treatment of the root canal, carried out according to the endodontic treatment protocols adopted in the Russian Federation, is not sufficient to achieve a long-term clinical effect. The modification of the protocol for treating the root canal by radiation of an erbium laser with a wavelength of 2940 nm, a power of 40 mJ, and a pulse frequency of 10 Hz showed a high quality of sterilization of the root canal. Similar results on the effect of Er, Cr: YSGG laser with a wavelength of 2780 nm were obtained in studies of T.V. Furtseva and others. [9]. Authors, L.U. Orekhova et al. [10], I.I. Malov et al. [11], S.L. Blashkova et al. [12] in their studies, it was demonstrated the high efficiency of using lasers in endodontics. Similar data to our results were published by E. Henninger et al. [13]: the authors studied the radiation efficiency of an erbium laser on strains of *Streptococcus gordonii* in combination with *Actinomyces oris* or *Fusobacterium nucleatum* and showed the effectiveness of this treatment in endodontic infections. Researchers from different countries show high efficiency of root canal treatment with both diode and erbium lasers [14-16].

## Conclusion

Within the limits of this study, the efficiency of root canal treatment with an erbium laser with a wavelength of 2940 nm, energy of 40 mJ, pulse frequency of 10 Hz, and a power of 0.5–8.4 W is quite high. Modification of the irrigation protocol for endodontic treatment, in particular, chronic periodontitis K04.5, by the action of radiation from an erbium laser is an effective method and more further studies are needed in this field.

**Таблица 1**
Результаты микробиологического исследования основной группы пациентов на этапах эндодонтического лечения
**Table 1**
Results of a microbiological study of the main group of patients at various stages of endodontic treatment

| Опытная группа Main group | Enterococcus Faecalis КОЕ/мл CFU/ml | Staphylococcus Haemolyticus КОЕ/мл CFU/ml | Staphylococcus epidermalis КОЕ/мл CFU/ml | Streptococcus mitis КОЕ/мл CFU/ml | Streptococcus mutans КОЕ/мл CFU/ml |
|---|---|---|---|---|---|
| До обработки канала Before irrigation | $10^8$ | $10^8$ | $10^5 – 10^8$ | $10^7$-$10^8$ | $10^7$ |
| После медикамен тозной обработки канала After root canal irrigation | $10^3$ | $10^2$ | $10^2$ | $10^2$ | $10^2$ |
| После медикамен тозной обработки канала и обработки лазером After root canal irrigation and Er:YAG laser | Нет роста No growth | Нет роста No growth | Нет роста No growth | Нет роста No growth | Нет роста No growth |

**Таблица 2**
Результаты микробиологического исследования пациентов группы сравнения на этапах эндодонтического лечения
**Table 2**
The results of a microbiological study of the control group of patients at the stages of endodontic treatment

| Контрольная группа Control group | Enterococcus Faecalis КОЕ/мл CFU, cell/ml | Staphylococcus Haemolyticus КОЕ/мл CFU, cell/ml | Staphylococcus epidermalis КОЕ/мл CFU, cell/ml | Streptococcus mitis КОЕ/мл CFU, cell/ml | Streptococcus mutans КОЕ/мл CFU, cell/ml |
|---|---|---|---|---|---|
| До обработки канала Before root canal irrigation | $10^8$ | $10^8$ | $10^5$ -$10^8$ | $10^7$- $10^8$ | $10^7$ |
| После медикаментоз ной обработки канала After root canal irrigation | $10^3$ | $10^2$ | $10^2$ | Нет роста No growth | $10^2$ |
| Значение критерия p P-value | 0.009 | 0.009 | 0.04 | 0.000 | 0.009 |

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