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**EVALUATION OF ANTI-INFLAMMATORY THERAPY EFFICACY IN PARODENTIUM DISEASE
TREATMENT WITH MOLECULAR-GENETIC DIAGNOSTIC TEST-SYSTEMS**

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Abstract

A number of highly specialized and extremely sensitive methods, including the widely popular polymerase chain reaction, have recently been introduced into the clinical diagnostics of the oral cavity microflora. The paper presents the results of a study to illustrate the application of modern DNA-STRIP based diagnostic systems in the professional activity of a dental practitioner and evaluates the efficacy of the subsequent anti-inflammatory treatment for parodontium diseases. The studied 90 patients with periodontitis of moderate severity was divided into 2 groups. To diagnose parodontium pathogenic strains of bacteria in both groups, Micro-IDent diagnostic test (HAIN Lifescience, Germany) was used, which is based on inverse hybridization. After diagnostics, one group was treated traditionally; the other group took antibiotics. The specific therapy with antibiotics directed against the bacteria, which were revealed by the Micro-IDent test, was proved to be more effective than the traditional treatment. Accurate diagnostics of a disease at its early stage and constant monitoring of a therapeutic success could be considered to be the advantages of the data from molecular-genetic systems.

Key words: parodontium disease, molecular-genetic systems, early diagnosis, parodontium pathogenic microorganisms.

Introduction

The issue of parodontium diseases remains one of the most burning issues in modern dentistry. Today, most authors believe the main reason of inflammatory parodontium diseases to be the disorder of the dynamic balance between the pathogenic microbes of dental plaque and the protective forces of the parodontium and the body on the whole [1,2]. While developing in the periodontal pocket, complexes of pathogenic bacteria destroy dentogingival apparatus and increase bone resorption [3-6]. Traditionally, the diagnostics of parodontium pathogens has been made exclusively by means of cultural research methods and involved work only with “live” bacteria.

However, there are more advanced methods to diagnose the parodontium pathogenic bacteria in the oral cavity - the molecular-genetic ones, which are of great interest [7, 8]. The sensitivity of these methods are close to 98%.

The long-term success of any conducted therapy undoubtedly requires targeted elimination of parodontium pathogenic complexes, that finally reduces the bacterial load on parodontium tissues [9-11].

In order to detect parodontium pathogenic microorganisms with molecular-genetic systems and evaluate the subsequent anti-inflammatory treatment efficacy of parodontium diseases, there have been a study, which is described in this paper.

Materials and Methods

The study was approved by the Expert Council of the Medical Institute of Belgorod State University. The studied group included 90 people aged 43-52 years. The participants were patients with chronic generalized periodontitis of moderate severity. The dental health of all patients was thoroughly examined. In our studies, we particularly focused on the patients' complaints, patients' life and disease history, examination of organs and oral cavity tissues. The patient hygiene performance (PHP) index of Podshadley and Haley (1968) and A. Russel's periodontal index (PI) were used to evaluate the parodontium tissues and oral cavity hygiene objectively. All patients were divided into 2 groups with 45 people each in accordance with the purposes of the research. The patients of the group I (45 people) were treated in line with the traditional scheme, including instructions on oral cavity hygiene, dental sanitation, scaling, local anti-inflammatory therapy with a compound medication "Gialudent Gel 2" (LLC "NKF Omega-Dent", Russia). This is a compound medication with metronidazole and chlorhexidine is based on hyaluronic acid.

Antibiotics (mainly broad-spectrum ones) affecting the targeted strains of periodontal pocket anaerobes were prescribed as anti-inflammatory therapy for the patients of the group II (45 people). The medications were recommended by HAIN Lifescience, Germany.

To identify the microbiocenosis of the periodontal pockets, we used the Micro-IDent test system (HAIN Lifescience, Germany) based on inverse hybridization. Its diagnostic kit included sterile tubes with paper points and an accompanying form.

The analysis of the inverse hybridization included DNA amplicon denaturation and application of the obtained sample on the nitrocellulose strip containing specific probes of 11 parodontium genes, a control probe with extracted DNA and conjugate control. The amplicons related the complementary probe were visualized, when streptavidin conjugate with alkaline phosphatase was added. The results were read with the help of the available sample.

Research methods: examined gingival sulcus or pocket was isolated from the saliva, with a paper point being submerged into it for 20 seconds, which was taken out and put into a sterile container and sent to the molecular and genetics laboratory of the Interregional Center of dental innovations at Belgorod National University. The method allowed detecting particular “marking” sequence of bacteria’s nucleic acids rather than the bacteria themselves (the method is sensitive to detect 5-12 main parodontium pathogenic bacteria).

Due to its high specificity and sensitivity, the **Micro-IDent** test, which is aimed to detect parodontium pathogenic bacteria in the exudate of periodontal pockets, quickly identifies the patients from risk groups and gives valuable information to choose the most effective anti-inflammatory treatment.

Results

As it was expected, the clinical study revealed that the PHP average value (1968) was 2.2 ± 0.46 for the patients of both groups that showed rather unsatisfactory hygiene of the oral cavity. The average values of the Russel periodontal index were 3.3 ± 0.24 for the patients of the group I and 3.7 ± 0.12 for the patients of the group II that indicated moderate periodontitis. The patients complained about bleeding in the gum, swelling, painfulness, unpleasant odor from the mouth. Pathologic gingival pockets of about 5 mm could be observed at 79% (71 people).

The microbial landscape of periodontal pockets studied with the Micro-IDent test-system is characterized by the strain markers of pathogenic red complex (*Porphyromonas gingivalis*, *Tannerella forsythia*) for 82% of cases (72 people), of orange one (*Fusobacterium nucleatum*) for 27 % of cases (24 people), of pink one for 51 % of cases (46 people). The markers of pathogenic strain complexes are given is Table 1 and Figure 1.

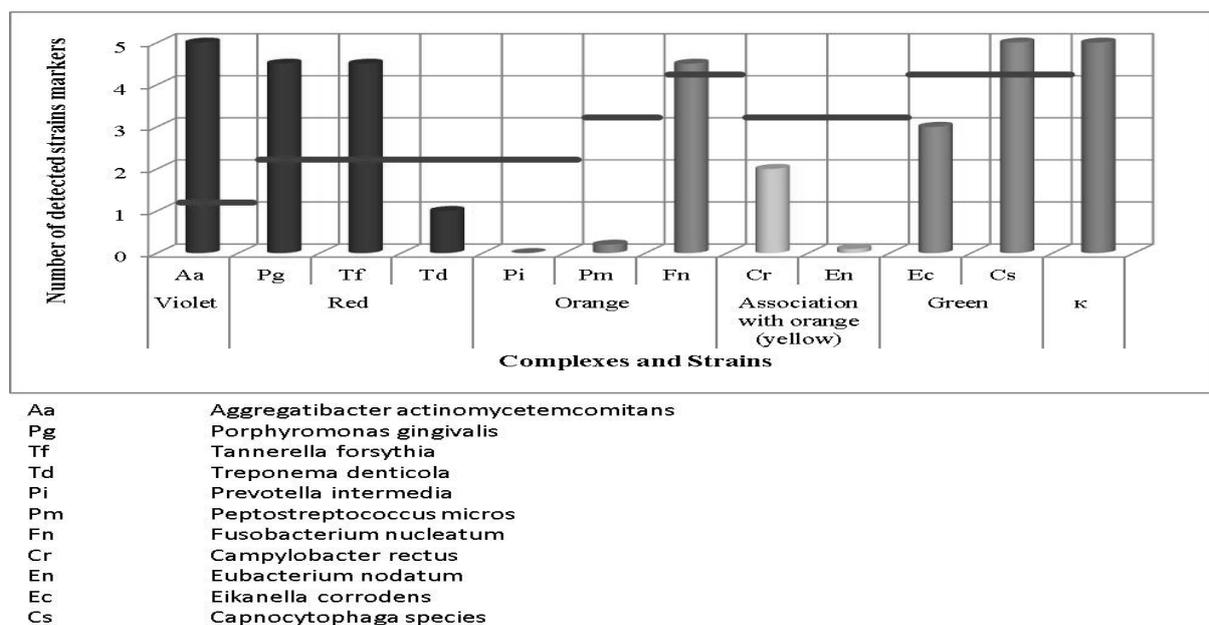


Figure 1. Microbial landscape of periodontal pockets based on the Micro-IDent test results.

The patients of the group I underwent 10 anti-inflammatory therapeutic procedures with Gialugent Gel 2. The gel was applied into the periodontal pockets. The study identified that the patients' condition improved on the second day: bleeding in the gum and the swelling decreased. 14 days after the therapy all patients from the group I were diagnosed with molecular-genetic systems again - to evaluate the efficacy of the treatment. The Micro-IDent analysis revealed that the periodontal pockets still had parodontium pathogenic strains but in significantly lower concentrations, below the threshold values. Figures 2 and 3 illustrate the results of the study before and after the treatment.

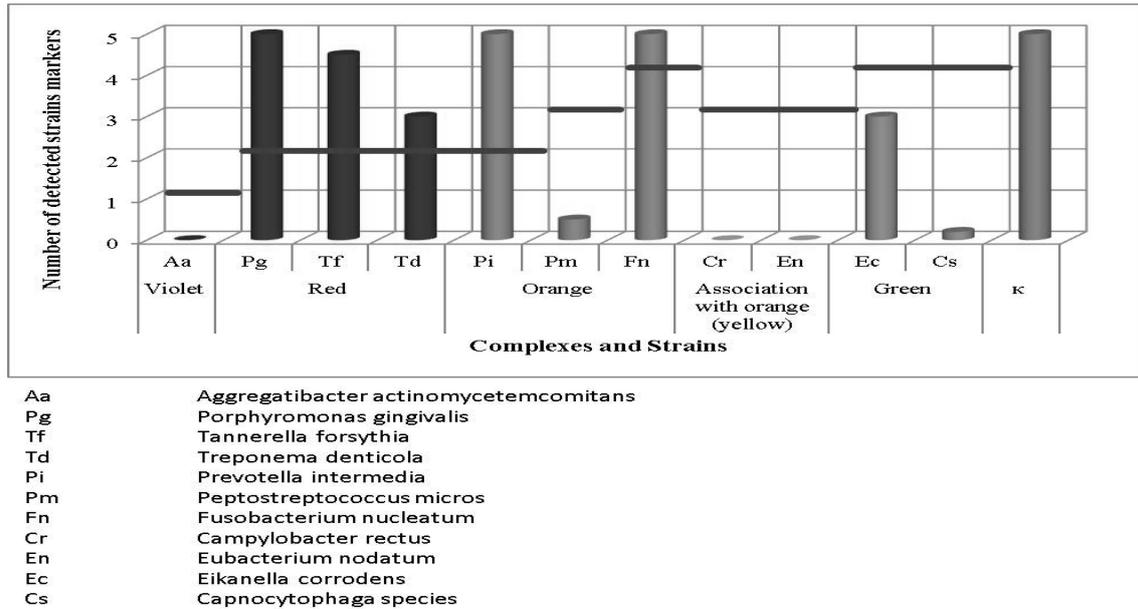


Figure 2. Microbial landscape of periodontal pocket before the combination treatment based on Micro-IDent test results.

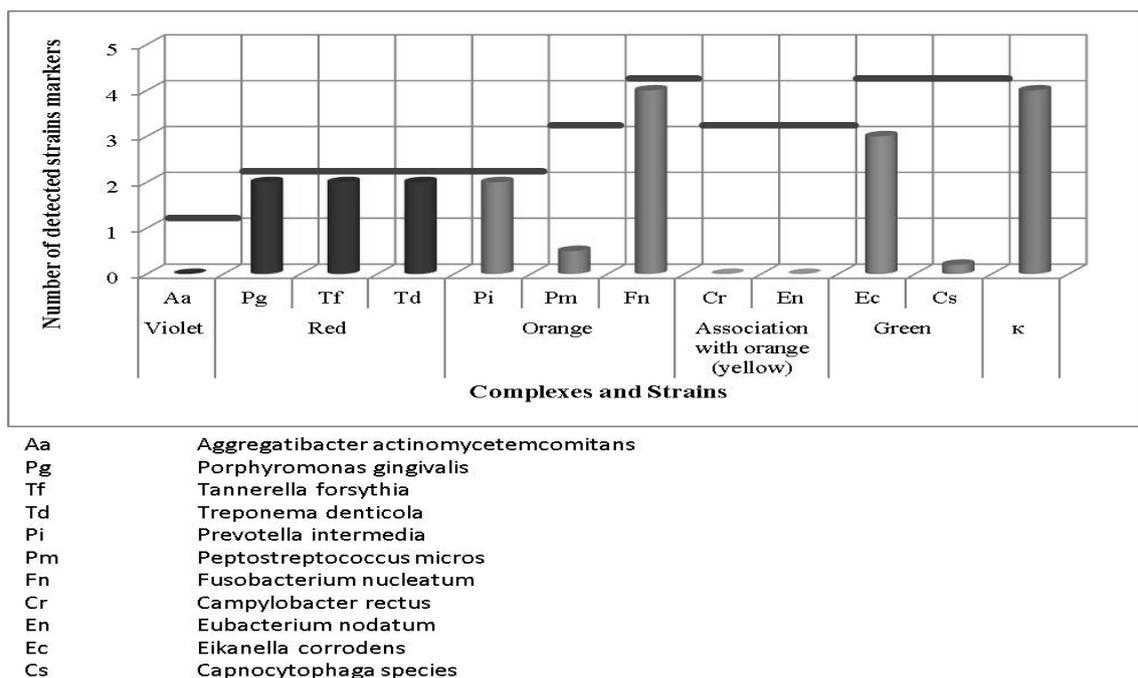


Figure 3. Micro-IDent test results after Gialugent Gel 2 treatment.

As for the patients of the group II, no markers of pathogenic strains were revealed after antibiotic therapy (metronidazole 400 mg 3 times a day, amoxicillin 500g 3 times a day, clindamycin 300 mg 4 times a day, a course of 7 days) based on Micro-IDent test results done after 14 days after a course of anti-inflammatory therapy. Figures 4, 5 give the results for Group II.

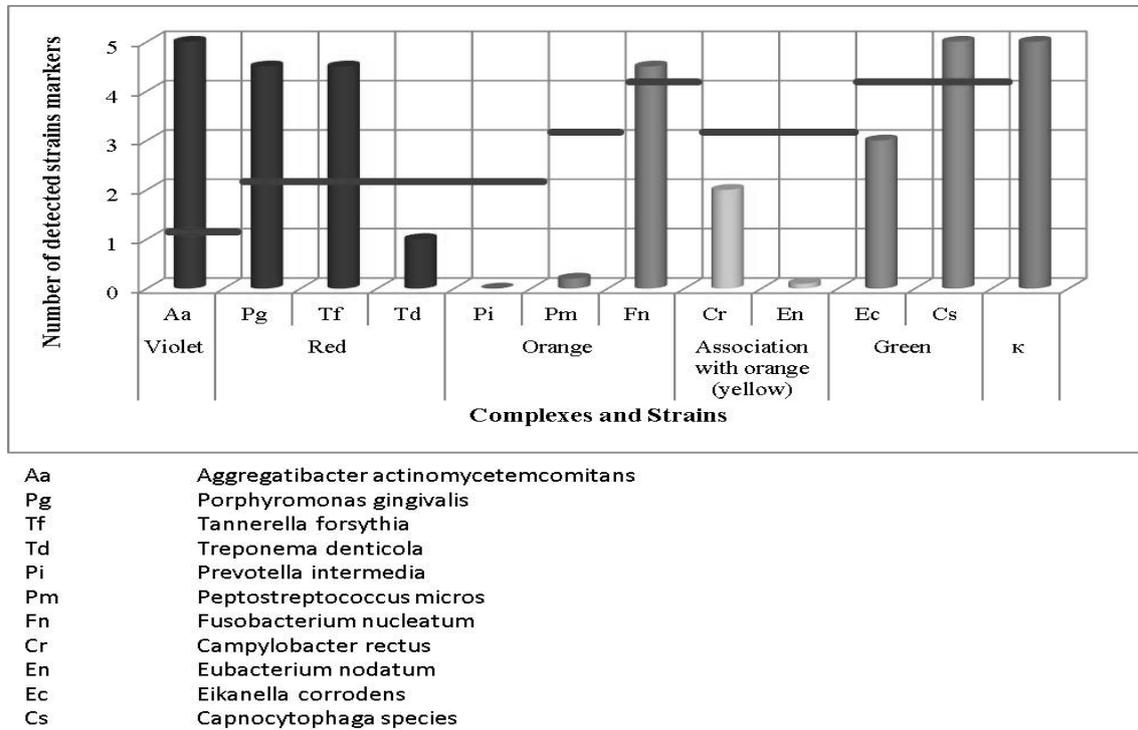


Figure 4. Microbial landscape of periodontal pocket before a combination treatment based on Micro-IDent test results.

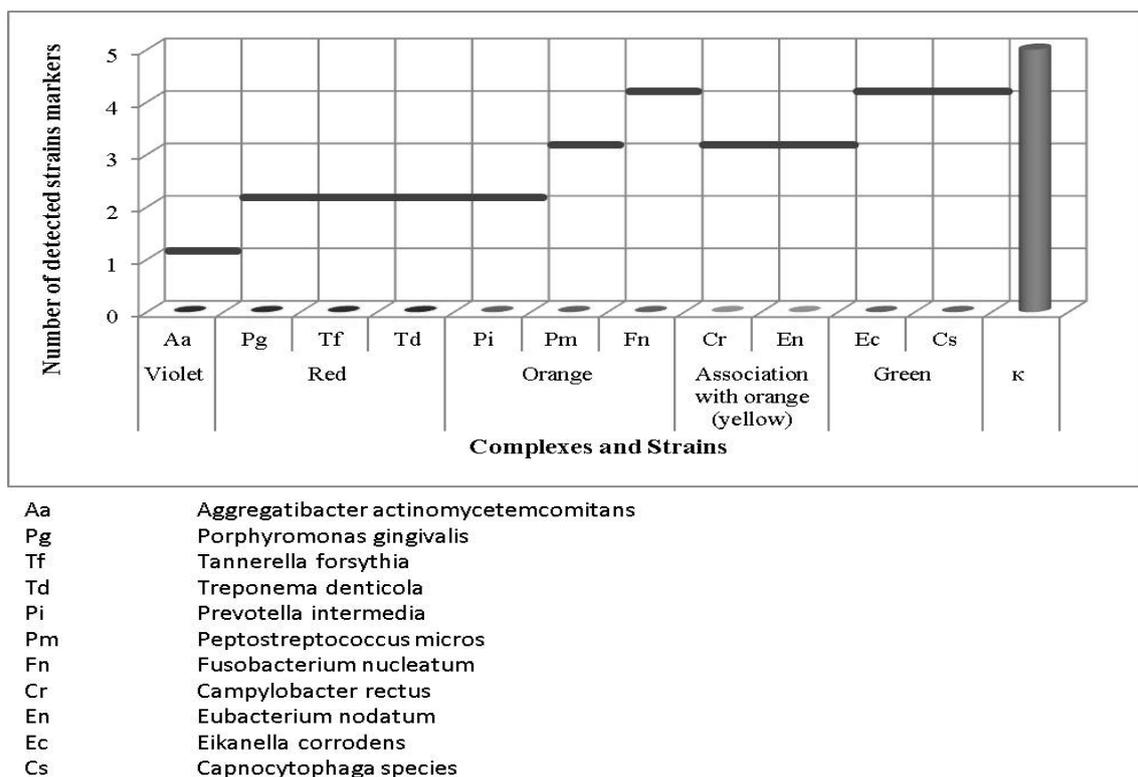


Figure 5. Micro-IDent test results after the antibiotic therapy course.

Discussion

Summing up the results, it should be noted that the diagnostic methods based on periodontal index application are considered to be indirect, despite their wide usage in clinical practices, because they are likely to reveal some local aspects of parodontium disease pathogenesis. Besides, these methods are not very informative in the evaluation of the treatment efficacy. Interpreting such indicators as PHP, PI (Russell index) does not allow us to say that there are no periodontal strain markers in periodontal pockets.

However, the molecular genetic test Micro-IDent (Hain Lifescience), which has detected 11 parodontium pathogens, gives an opportunity for a practicing dentist to assess the flora microbiocenosis before treatment and to evaluate the effectiveness of antimicrobial therapy.

The antibiotic therapy in line with the treatment regimen suggested by HAIN Lifescience (Germany) completely eliminates parodontopathogenic microbiocenosis from the periodontal pocket.

Thus, the usage of modern molecular-genetic systems Micro-IDent (HAIN Lifescience, Germany) in the practices of a periodontist evaluates the risk for the inflammatory parodontium disease development, justifies the etiological diagnosis of a disease, provides early detection of a disease at the stage of minor clinical signs, helps to assign the appropriate antibacterial therapy for each patient.

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