



## Mouthwash as a factor in controlling the formation of soft dental plaque in patients with orthodontic treatment

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**Abstract.** Patients with dental braces are more likely to experience rapid accumulation of dental plaque and gum inflammation, therefore, antimicrobial mouthwashes that enhance daily oral hygiene can effectively remove and control bacterial plaque, improve gum health, and are safe preventive agents without side effects. The aim of the study was to evaluate the quantitative and qualitative composition of the soft dental plaque microbiocenosis under the influence of an alcohol-free oral hygiene product in patients undergoing orthodontic treatment. The study included patients undergoing orthodontic treatment. Samples were taken from the surface teeth of the maxilla. The obtained swabs from the surface of the upper molars were examined using a bacteriological method with subsequent identification: microscopic, cultural, and biochemical. Comparison of the effectiveness of an alcohol-free mouthwash in terms of changes in the quantitative and qualitative composition of microorganisms and the condition of periodontal tissues in patients undergoing stationary orthodontic therapy carried out after three months of use. The study revealed gram-positive and gram-negative bacteria and *Candida* fungi with a subsequent insignificant decrease in gram-negative bacteria ( $p = 0.999$ ) after using the mouthwash. The results obtained showed changes in the taxonomic composition, namely, a decrease in the number of bacterial genera from 11 to 9. In addition to the mentioned results, changes were found at the population level of the soft dental plaque microbiota in patients who used the alcohol-free mouthwash for three months, namely, a decrease in the number of *Streptococcus* spp.  $\alpha$  ( $p(x \leq T) = 0.9958$ ), *Propionibacterium* spp. ( $p(x \leq T) = 0.9837$ ), while changes in *Streptococcus* spp.  $\beta$  and *Streptococcus* spp.  $\gamma$  were minimal. A comparison of the population level of microorganisms before and after the use of mouthwash revealed significant differences ( $p < 0.05$ ). Determination of the gingival index demonstrated a slight improvement in the periodontal status of the examined subjects. The conducted study is of great importance for the development of new strategies for the prevention and treatment of oral cavity diseases

**Keywords:** microbiocenosis; microorganisms; oral cavity; gingivitis; dental brace

### ★ INTRODUCTION

The placement of orthodontic appliances on teeth often leads to an increase in bacterial colonisation on both the visible and hidden biofilm of the tooth surface. This, in turn, plays a significant role in the inflammation of gum tissues during orthodontic treatment. Given the highly complex ecosystem of the oral cavity, inhabited by a diverse range

of microorganisms, any external intervention, such as fixed orthodontic appliances, can disrupt the microbial balance within the microocenosis of the oral cavity [1, 2].

S.M. Hamdoon *et al.* [3] and D.T. Mahjoub *et al.* [4] state in their research that orthodontic treatment leads to an increase in the number of cariogenic bacteria, specifically

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*Streptococcus* and *Lactobacillus*, as well as fungi and other periodontal pathogens associated with dental plaque. A.K. Shahi *et al.* [5] also demonstrated the effects of various types of antibacterial agents (Ozonated Olive Oil Gel, Chlorhexidine gel, and mouthwash Amflor) on *Streptococcus mutans* and *Lactobacillus* bacteria.

N. Manashchuk *et al.* [6] conducted a review of various oral hygiene products, particularly dental elixirs and mouthwashes, and classified them based on their composition and intended use. They emphasise that any such product should be used in conjunction with toothpaste and a brush, serving as a secondary step in oral care. This is because they are used to clean hard-to-reach areas of the periodontium and interdental spaces.

N. Bila *et al.* [7] noted in their research that the use of antiseptic agents leads to qualitative changes in the composition of the oral microbiome. They emphasised the importance of consulting a healthcare professional regarding the choice of mouthwash for daily oral hygiene, as an excessive amount of sodium fluoride may have adverse effects on the human body, including potential impacts on the thyroid gland, cognitive abilities, and more.

G.M. Tartaglia *et al.* [8] and M. Selvaraj *et al.* [9] demonstrated the importance of conducting research into the side effects of mouthwashes, such as local morphological changes (oral mucosa and dental-crown staining, mucosal lesions) and functional changes (taste modifications, abnormal oral sensation). Given that mouthwashes are commonly used as an adjunct to mechanical tooth brushing, they highlight the need for detailed studies into the combinations of ingredients in these liquids and their impact on reducing dental plaque and improving periodontal tissue health.

S. Ayesha *et al.* [10] highlight the importance of selecting the right mouthwash, especially for patients undergoing orthodontic treatment, as they are prone to dental plaque accumulation, gum inflammation, and mineral loss from tooth enamel. They assert that chlorhexidine and *Aloe vera* mouthwash have shown good results in controlling inflammatory processes in periodontal tissues, gingival bleeding, and plaque accumulation. However, prolonged use of chlorhexidine can have adverse side effects, making plant-based mouthwashes an effective alternative.

However, there is a lack of research investigating the *in vivo* effects of daily oral hygiene products on patients undergoing orthodontic treatment, and analysing their impact on the oral microbiome and gingival tissues. This issue requires further study and a better understanding of the relationship between the substances contained in such mouthwashes and the composition of microorganisms forming soft dental plaque. This study aimed to analyse the quantitative and qualitative composition of the microbiocenosis of soft dental plaque in patients undergoing orthodontic treatment, under the influence of an alcohol-free oral hygiene product.

## ★ MATERIALS AND METHODS

A randomised controlled trial design was developed to assess and compare the effectiveness of an alcohol-free mouthwash in reducing the quantity and quality of microorganisms in 8 patients undergoing orthodontic treatment at a private dental clinic in Ternopil (commenced in 2023 and ongoing in 2024). These changes were assessed

at regular intervals. Inclusion criteria for patients in the study group were: aged 15-40 years; at the initial or intermediate stages of dentition correction; undergoing fixed orthodontic treatment with brackets on the upper teeth. Exclusion criteria for patient selection were: no signs of tooth decalcification; no known hypersensitivity to oral hygiene products; no known medical conditions or medication that could affect oral tissues. Samples were collected from the surface of the upper molars of 8 patients undergoing orthodontic treatment. Samples were collected using a swab moistened with 0.9% saline solution. Samples were transported at a temperature of +18-22°C within 1-2 hours of collection.

For culturing on nutrient agar plates, an inoculum of microorganisms was prepared at a defined concentration (using the McFarland scale (HiMedia, India)). The inoculum, diluted to  $10^{-6}$  in a volume of 100  $\mu$ l, was applied to nutrient agar plates: Endo agar (Farmaktiv, Ukraine) for the isolation of gram-negative microorganisms, blood agar (Farmaktiv, Ukraine) for the isolation of streptococci, mannitol salt agar (Farmaktiv, Ukraine) for the isolation of staphylococci, Sabouraud agar (Farmaktiv, Ukraine) for the isolation of *Candida* species, anaerobic agar (Brewer) (Farmaktiv, Ukraine) for the cultivation of anaerobic bacteria, and Blickfeldt agar (Farmaktiv, Ukraine) for lactobacilli.

Cultivation of facultative anaerobic bacteria was carried out in a thermostat (TS-20, MIS-MA, Ukraine) at a humidity of approximately 70-80% and a temperature of +37°C for 24-48 hours. For the cultivation of *Candida* species, the incubation time was extended to five days at a temperature of +22°C. To create anaerobic conditions, a GENbox with a gas pack (bioMerieux, France) was used, and the incubation time for anaerobic bacteria was 48-72 hours at a humidity of approximately 70-80% and a temperature of +37°C. For identification, the following methods were used: colony morphology, Gram staining (BioGnost, Croatia), and biochemical tests: catalase test (Biolife Italiana S.r.l., Italy), coagulase test (Biolife Italiana S.r.l., Italy), Olkenitskyi medium (Farmaktiv, Ukraine), Voges-Proskauer test (Farmaktiv, Ukraine), and indole test (Farmaktiv, Ukraine) [11, 12]. Quantitative analysis was performed by determining the colony-forming units (CFU/mL) in 1 mL of the sample and calculated using the formula [13]:

$$TCC = \frac{N \times A}{V}, \quad (1)$$

where TCC is the total colony count,  $\lg_{10}$  CFU/mL, N is the number of colonies in a sector, CFU; A is the dilution factor; V is the volume applied to the plate, mL.

To assess the microbial population, a constancy index (C) was determined using the formula:

$$C = \frac{n \times 100}{N}, \quad (2)$$

where C is the constancy index, %; n is the number of samples in which the studied species was detected; N is the total number of samples analysed.

Reference values were taken as follows: dominant >50%, frequent 20-50%, infrequent 1-19%, and rare <1%. Loe and Silness's gingival index was used as a basis for assessment [14]. The gingival index scoring scale was as follows: 0 – indicates no inflammation of the gingiva;

1 – shows mild inflammation (slight changes in gingival condition); 2 – moderate inflammation (characterised by swelling and slight enlargement); 3 – severe inflammation (marked swelling of the gums). To investigate the effect of the mouthwash on the quantitative and qualitative composition of the microcenosis and gingival status, an alcohol-free oral hygiene mouthwash with a plant-based component was selected with the following composition: water, glycerin, propylene glycol, xylitol, disodium EDTA, poloxamer 407, allantoin, methylparaben, cetylpyridinium chloride, sodium fluoride, *Aloe barbadensis* leaf juice, acesulfame potassium, propylparaben, menthyl acetate, glycerin, flavour, CI 19140, C1 42051, limonene.

The initial stage of the examination involved assessing the condition of the soft and hard tissues of the oral cavity to detect any changes during the study and determine their possible association with the use of mouthwash. Patients used the mouthwash twice a day, morning and evening, after brushing their teeth, and were informed not to use any other mouthwash throughout the study. They were also required to return the empty container to monitor product usage. Samples were collected from the tooth surface before the start of use, serving as a control in the study, and again after three months of using the mouthwash. The treatment period of 3 months was chosen because gingivitis is a chronic condition, and to avoid interfering with the hygiene habits of the individuals involved in the study. Regarding the study period, these typically last 6 months with an interim assessment at 3 months to evaluate both the efficacy and safety of the chemicals for patients. However, mouthwashes are also used and prescribed for shorter periods to assess their efficacy over shorter timeframes, which is of scientific interest [15].

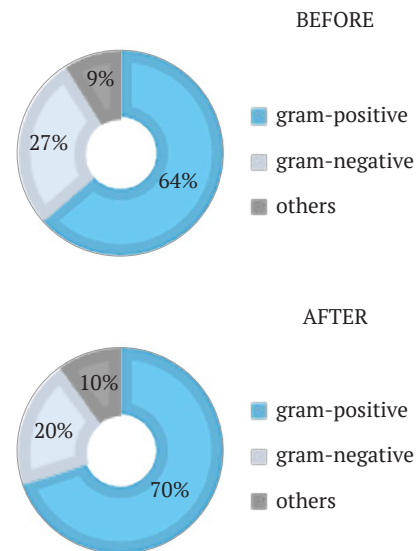
Statistical analysis was performed using the STATISTICA 10 and Microsoft Excel 2016 software packages. Mean values (M) with standard error of the mean ( $\pm m$ ) were calculated. Non-parametric tests were used for datasets with distributions differing from normal: the Mann-Whitney U-test for comparing two independent samples, the Wilcoxon signed-rank test for assessing dynamic changes within groups, and the Student's t-test. The level of significance was set at  $p \leq 0.05$ .

The study was conducted following the recommendations outlined in the "Council of Europe Convention on Human Rights and Biomedicine" [16], taking into account the ethical principles set out in the Declaration of Helsinki of the World Medical Association regarding research involving human subjects, and in compliance with Order No. 690 of the Ministry of Health of Ukraine dated 23.09.2009, as well as the requirements of the bioethics committee of I. Horbachevsky Ternopil National Medical University of the Ministry of Health of Ukraine (protocol No. 75 dated 1.11.2023). All patients provided informed consent to participate in the study.

## ★ RESULTS

A study was conducted on 8 clinical samples from patients undergoing orthodontic treatment. Analysis of the gender and age composition of the patients: women predominated among the patients (6 women – 75.0%), and there were significantly fewer men (2 men – 25%, patient age from 17-37 years (median 20).

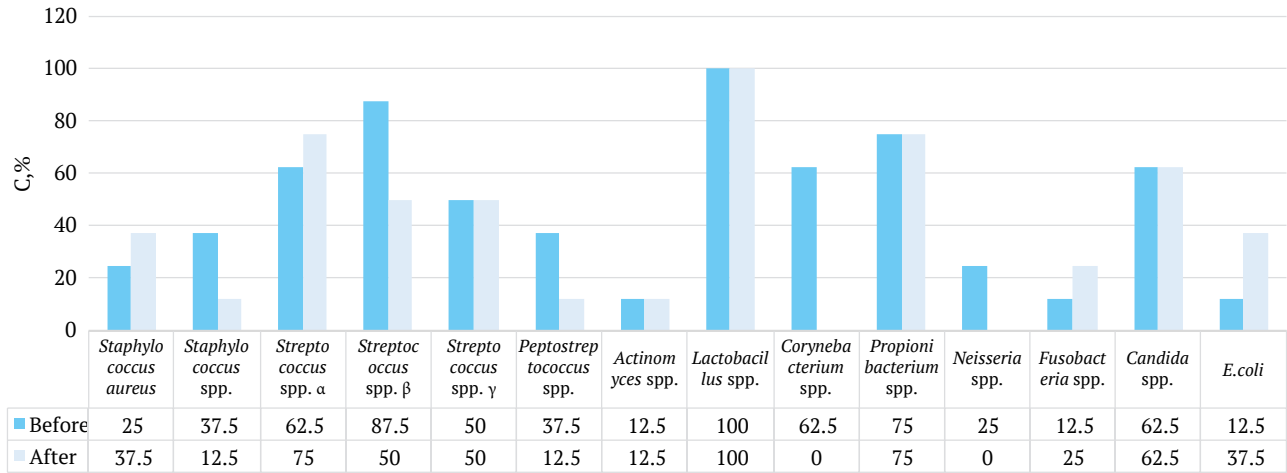
An analysis of soft dental plaque on the surface of the maxillary molar of the examined subjects with dental braces was carried out before and after the use of mouthwash after 90 days of its application. The isolated microorganisms belonged to gram-positive bacteria, the number of which was 63.63% and 70%, gram-negative bacteria 27.27% and 20.0%, and representatives of the other group 9.09% – 10.0% respectively ( $p = 0.999$ ). The results of the paired t-test showed that there was a negligible difference between the two groups and demonstrated a slight decrease in gram-negative bacteria over the group of gram-positive bacteria (Fig. 1).



**Figure 1.** The proportion of microorganism groups before and after using the mouthwash in the examined subjects (n=8)

**Source:** compiled by the authors

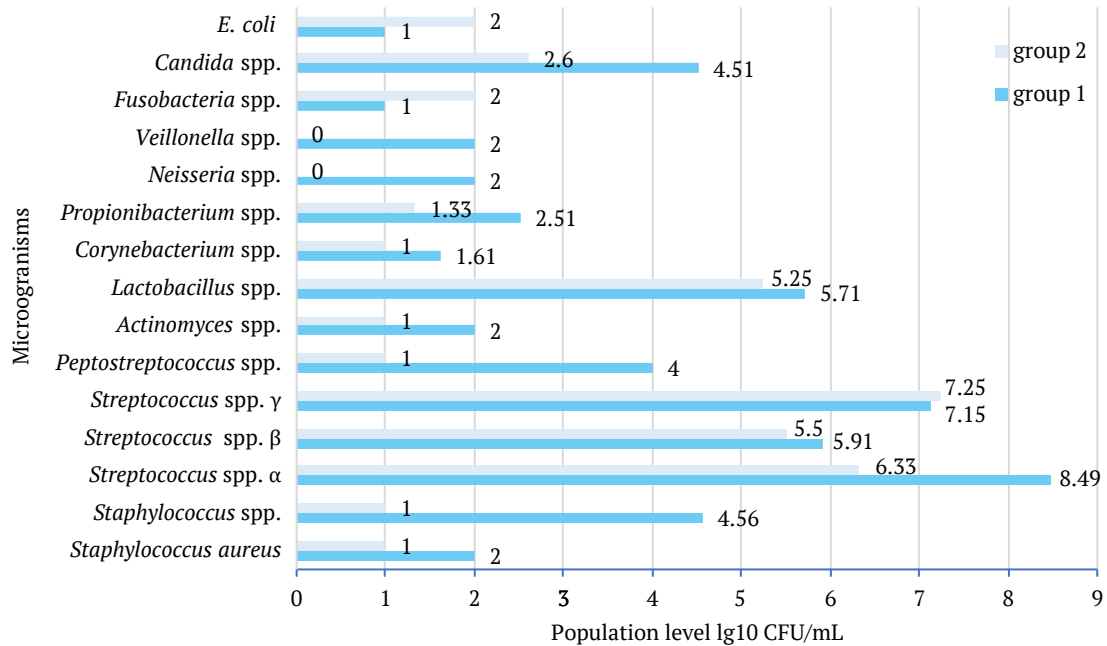
The taxonomic composition of the soft dental plaque microbiocenosis in the examined subjects was represented by microorganisms belonging to 11 genera of bacteria and *Candida* fungi and 9 genera and *Candida* yeasts after the use of mouthwash. Determination of the constancy index indicated the proportion of representatives of different genera in the studied samples before (group 1) and after (group 2) using the mouthwash after 3 months of application. In particular, the dominant ones include *Lactobacillus* spp., *Streptococcus* spp.  $\beta$ , *Streptococcus* spp.  $\alpha$ , *Streptococcus* spp.  $\gamma$ , *Corynebacterium* spp., *Propionibacterium* spp., and *Candida* spp. (from 50.0-100%), which belong to both gram-negative and gram-positive bacteria, however, representatives of *Corynebacterium* spp. were not found after application. Further analysis of the obtained samples demonstrated the presence of *Staphylococcus aureus*, *Staphylococcus* spp., *Peptostreptococcus* spp., *Neisseria* spp., which belong to the frequently occurring ones (from 25.0-37.5%) with the difference between the two groups of bacteria of the genus *Neisseria* spp., which were not found in the second group. Representatives of microorganisms that are not often found were *Actinomyces* spp. and representatives of the allochthonous oral microbiota, bacteria of the genus *Escherichia*, namely *E.coli* – 12.5% respectively (Fig. 2).



**Figure 2.** Constancy index in the examined patients with dental braces

Source: compiled by the authors

Study of the population level of various microorganisms in the microbiocenosis on the surface of the upper molar in patients with dental braces before and after using the mouthwash (Fig. 3).



**Figure 3.** Population level of soft dental plaque microbiota

in patients undergoing orthodontic treatment before and after using the mouthwash

Source: compiled by the authors

The composition of soft dental plaque is formed by autochthonous lactobacilli, *Streptococcus* spp. alpha, gamma, beta, which, on average, reach a significant population level and belong to the group of microorganisms with a high dominance index. The determination of differences between the population composition in patients before and after the use of mouthwash for *Streptococcus* spp. alpha  $p(x \leq T) = 0.9958$ , *Propionibacterium* spp.  $p(x \leq T) = 0.9837$  is statistically significant concerning the mean values  $p < 0.05$ . However, when comparing the mean values for *Streptococcus* spp. beta, it was found that there were no significant differences, and it was  $p(x \leq T) = 0.6544$ , so it can be considered that such differences

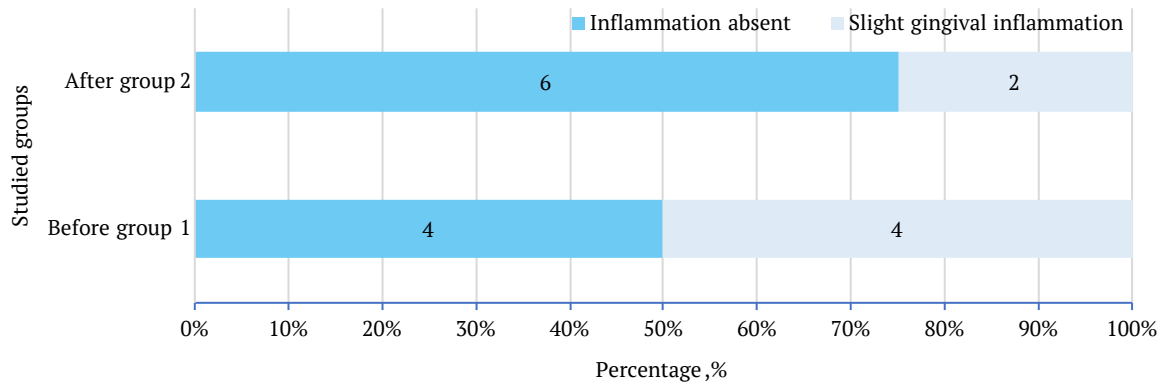
are insignificant. For *Lactobacillus* spp. ( $p(x \leq T) = 0.631$ ), *Streptococcus* spp. gamma ( $p(x \leq T) = 0.5873$ ) the comparison of data between group 1 and group 2 was also insufficient to be statistically significant. The determination of differences at the population level between groups 1 and 2 for the conditionally pathogenic representative of the genus *Candida* spp. by paired t-test ( $p(x \leq T) = 0.04374$ ) indicates insufficient differences between the mean values in both groups.

Other conditionally pathogenic microorganisms such as staphylococci, *E. coli*, *Veillonella*, *Neisseria*, fusobacteria, and *Actinomyces* have low population-level bacterial counts. When comparing the average values of all detected

microorganisms before and after the use of mouthwash in the subjects, it was established that the differences between the groups are statistically significant  $p(x \leq T) = 0.01273$ , and are at the significance level  $p < 0.05$ . When comparing the population levels before and after the use of alcohol-free mouthwash using a paired t-test, it showed that there is

a significant difference between before and after  $p = 0.005$ .

One of the important criteria for choosing a mouthwash is its effect on periodontal tissues. Thus, to determine this effect, the gingival index was chosen, which was determined before the use of mouthwash and after three months of its use (Fig. 4).



**Figure 4.** Comparison of periodontal tissue condition in patients before and after using the mouthwash

Source: compiled by the authors

A review of the subjects before the start of the experiment demonstrated an equal ratio of patients with mild tissue inflammation and those without it was 1:1, after use 1:3. When comparing the gingival index of the two groups based on the results of the paired t-test, a slight difference was demonstrated between before and after,  $p = 0.451$ , the use of alcohol-free mouthwash and was verified using the Wilcoxon signed-rank test, which confirmed that there is a small difference between before and after,  $Z = -0.7$ ,  $p = 0.484$ ,  $r = -0.3$ . As a result, the use of alcohol-free mouthwash for the oral cavity by patients undergoing orthodontic treatment did not reveal a negative impact on the mucous membrane. However, its use led to a decrease in the microbial composition at the population level and an improvement in the condition of the periodontal tissues.

## DISCUSSION

The presence of orthodontic appliances in the oral cavity makes it difficult to maintain proper hygiene. To overcome this problem during the active phase of orthodontic treatment, various hygiene procedures are carried out to reduce the accumulation of bacterial plaque and reduce the risk of periodontitis in patients. The conducted study, regarding the change in the quantitative and qualitative composition of the microbiota of soft dental plaque of the maxillary molars, demonstrated insignificant changes in the taxonomic composition of the identified microorganisms, in particular, after the use of mouthwash, no representatives of the genera *Veillonella* and *Neisseria* were found, before the use of mouthwashes, the taxonomic representation was 11 genera, and after 9 genera, respectively, which was reflected in the decrease in bacteria belonging to the group of gram-negative from 27.0% to 20.0%, and an increase in gram-positive microorganisms from 64.0% to 70.0%, respectively. This is confirmed by the results of a study by a group of scientists, namely R. Bescos *et al.* [17], who indicate an increase in gram-positive bacteria (Firmicutes) and a decrease in representatives of gram-negative

bacteria (Bacteroidetes), which is due to an increase in the acidity of the oral cavity pH, which is one of the factors influencing the physiological processes of microorganisms' development.

The results of the study show that the use of alcohol-free mouthwash with cetylpyridinium chloride significantly alters the oral microbiota. A comparison of the constancy index before and after three months of using the mouthwash revealed a decrease in the number of *Corynebacterium* spp. and *Neisseria* spp., *Veillonella* spp., after application. The predominance of *Lactobacillus* spp., *Streptococcus* spp., *Propionibacterium* spp., and *Candida* spp. remained high (from 50.0 to 100%). It is worth noting that in the study of Z.L.S. Brookes *et al.* [18] the results of research on the microbiome composition under the influence of chlorhexidine were demonstrated, using the genome sequencing method and indicated a decrease in the number of bacteria of the genus *Veillonella*, which led to a slight increase in fungi of the genus *Candida* spp., which is reflected in a decrease in the taxonomic diversity of microorganisms after the use of mouthwash, this is consistent with the obtained results, in particular, a decrease in the number of bacterial genera from 11 to 9, and the presence of *Candida* spp. in the study group of patients undergoing orthodontic treatment according to the constancy index refers them to the dominant group, yeasts are commensals of the oral cavity, however, they have pathogenic potential when the balance in the oral cavity is disturbed. I. Chatzigiannidou *et al.* [19] and A. Al-Kamel *et al.* [20] found that broad-spectrum antimicrobial agents do not always promote healing and can lead to further disruption of the microbiome, as confirmed by studies on the effects of chlorhexidine on oral biofilms, although chlorhexidine reduced the microbial load, however, this led to non-selective destruction of the oral microbiome and contributed to an increase in the number of taxa associated with periodontitis, namely *Fusobacterium*, which is consistent with the obtained results, regarding the increase in bacteria of the genus *Fusobacterium* at the

population level  $p(x \leq T) = 0.01273$  ( $p < 0.05$ ). However, the use of alcohol-free mouthwash in the subjects after three months of use revealed statistical differences ( $p < 0.05$ ) between before and after its use, both in terms of the quantitative and qualitative composition of dental plaque.

In a study conducted by M. Rajendiran *et al.* [21], which compared the composition of different mouthwashes and their effect on plaque formation and gingivitis, it was found that the use of an oral hygiene product containing cetylpyridinium chloride demonstrated a significant reduction in gum inflammation due to disruption of dental plaque maturation. This is consistent with the results obtained during the study, the condition of the periodontal tissues improved in two patients, as confirmed by the gingival index ( $p = 0.451$ ), and the quantitative indicators of the population level of microorganisms in the formation of soft dental plaque also decreased, as confirmed by the obtained statistical indicators.

A study conducted by A. Albert-Kiszely *et al.* [22] demonstrated that after 3 and 6 months of rinsing, there were no significant differences ( $p = 0.05$ ) between the experimental cetylpyridinium chloride and control groups regarding overall gingivitis, gingival bleeding, and plaque accumulation when examining mean values. However, when dichotomous data were examined, there were no statistically significant differences, but they were found over time. For example, 49.0% of samples were positive for *Porphyromonas gingivalis* at baseline. At the 6-month examination, 43.9% were positive ( $p = 0.24$ ) for *P. gingivalis*. After analysing the data on the gingival index, no statistically significant differences were found in gingival bleeding between the two study groups. However, after 3 and 6 months, a statistically significant decrease in gingival bleeding was demonstrated, but between 3 and 6 months these results were not statistically significant. Such an effect was observed in the study in some patients with bleeding gums, in whom the condition of the periodontal tissues improved, leading to a decrease in inflammation.

K. Becker *et al.* [23] suggest that the use of chemical mouthwashes as an adjunct to regular oral hygiene practices was effective in reducing plaque and gingivitis, thereby improving oral health over a 30-day experimental period. To study the effect, a 0.07% solution of cetylpyridinium chloride was used; the results showed a significant difference between pre- and post-observations at significance levels of 0.05% and 0.01% for cetylpyridinium and the control group in reducing gingival bleeding and plaque index. However, for the gingival index, such differences were not significant. This is also confirmed by studies by Z. Brookes *et al.* [24], who published a review of scientific papers on mouthwashes with different chemical compositions. They indicate that the evidence for the effectiveness of these agents on the microbiome has some limitations. In particular, while exerting a bacteriostatic and bactericidal effect on opportunistic or pathogenic microorganisms *in vitro*, acting against plaque and gingivitis *in vivo*, over-the-counter mouthwashes can cause dysbiosis of the oral microbiota. Most research on the clinical efficacy, namely the antimicrobial effect, has been conducted for antiseptic agents containing chlorhexidine. There is much less research on other such agents. In a randomised controlled trial, F.A. Adam *et al.* [25] noted that, regarding

the long-term efficacy of cetylpyridinium chloride for patients without orthodontic treatment, the use of a 0.07% solution of cetylpyridinium chloride for mouth rinsing was effective in reducing plaque levels. Gingival bleeding scores did not differ after 6 months. The tested product was well-tolerated and did not cause any serious clinical adverse effects or negative impact on the microbiota. This is emphasised by studies on the effect of mouthwash on the state of the oral microbiota, as well as the condition of teeth and gums, and does not indicate a negative impact on the composition of the microbiome.

In the study by B.A. Newman *et al.* [26], it was demonstrated that the number of *Porphyromonas*, *Corynebacterium*, *Abiotrophia*, and other known periodontal pathogens did not increase in supra-gingival plaque during 21 days of experimental gingivitis induction in the presence of cetylpyridinium chloride. At the same time, a slight increase in gum inflammation and bleeding was observed compared to the beginning of the study. The authors assert that a mouthwash containing cetylpyridinium chloride, when used as the sole oral hygiene product, provides significant benefits in reducing gingival inflammation by disrupting the maturation process of dental plaque (i.e., gingivitis-associated microbiota) and balancing the diversity and composition of the oral microbiota (i.e., health-associated microbiota). The results show a decrease in the number of *Corynebacterium* spp. and a reduction in periodontal tissue inflammation after the use of alcohol-free mouthwash.

I. Chen *et al.* [27], conducting a study on changes in the microbiota during orthodontic treatment using the sequencing method, argue that there is an increase in microbial richness in such patients compared to the control. Therefore, the use of mouthwashes as an auxiliary hygiene tool is justified and is confirmed in studies by U. Hussain *et al.* [28] regarding the use of chlorhexidine, which helps to control plaque formation and gingivitis but requires careful use due to possible side effects. G.P.J. Langa *et al.* [29] and M.M.T. Oo *et al.* [30] in their scientific papers argue that the use of a mouthwash containing cetylpyridinium chloride compared to provides a reduction in plaque and gingivitis index compared to patients who do not use mouthwashes to improve oral hygiene.

To summarise the results obtained regarding the use of alcohol-free mouthwash, it can be noted that it is insufficient as the sole oral hygiene agent for reducing plaque and improving the condition of periodontal tissues. However, in a comprehensive care regimen, when used in conjunction with a toothbrush, toothpaste, and mouthwash, it can help control plaque formation and maintain oral hygiene.

## ★ CONCLUSIONS

The study demonstrated that the use of an alcohol-free oral hygiene product in patients undergoing orthodontic treatment affects the composition of the microbial census in soft dental plaque. A decrease in gram-negative (from 27.27% to 20.0%) bacteria compared to gram-positive (from 63.63% to 70%) ( $p = 0.999$ ) indicates the effectiveness of this product in reducing pathogenic microbiota. It was also found that at the population level of the microbiocenosis of the molar surface, after the application of the product, the number of representatives of *Streptococcus* spp.  $p(x \leq T) = 0.9958$ , *Propionibacterium* spp.  $p(x \leq T) = 0.9837$

differences were statistically significant  $p < 0.05$ , for bacteria of the genera *Streptococcus* spp.  $p(x \leq T) = 0.6544$ , *Lactobacillus* spp.  $(p(x \leq T) = 0.631)$ , *Streptococcus* spp.  $\gamma$   $(p(x \leq T) = 0.5873)$  and fungi of the genus *Candida* spp.  $(p(x \leq T) = 0.04374)$  differences were insignificant, however, a trend towards their decrease was observed. A decrease in the quantitative composition of microorganisms in dental plaque leads to a decrease in the products released by them, which act as activators of gum inflammation. It has also been demonstrated that the population levels of microorganisms change significantly before and after using the product, confirming its effectiveness in reducing the number of bacteria in the oral cavity. The anti-inflammatory effect of the mouthwash on the condition of the gums of the subjects was better, due to a decrease in bleeding sites, as demonstrated by the gingival index, which was

determined before and after use ( $p = 0.451$ ). Therefore, the studied mouthwash caused insignificant changes in the quantitative and qualitative composition of dental plaque and improved the condition of the gum tissues, compared to the baseline. This study is important for creating new strategies for the prevention and treatment of oral diseases. However, it is necessary to conduct a double-blind, randomised, clinical trial to supplement the data on the impact on the oral microbiome, since even alcohol-free agents can have both positive and negative effects.

#### ✦ ACKNOWLEDGEMENTS

None.

#### ✦ CONFLICT OF INTEREST

None.

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## Ополіскувач як фактор контролю формування м'якого зубного нальоту у пацієнтів з ортодонтчним лікуванням

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**Анотація.** Пацієнти з брекет-системами частіше стикаються з проблемою швидкого накопичення зубного нальоту та запалення ясен, тому антимікробні ополіскувачі для ротової порожнини, які покращують щоденний домашній догляд, можуть забезпечити ефективне видалення і контроль бактеріального нальоту, покращення стану ясен та є безпечними профілактичними засобами без побічної дії. Метою дослідження було оцінити кількісний та якісний склад мікробіоценозу м'якого зубного нальоту під впливом безалкогольного засобу для гігієни ротової порожнини у пацієнтів, що отримували ортодонтчне лікування. У дослідженні брали участь пацієнти, які проходять ортодонтчне лікування, забір матеріалу відбувся з поверхні зубів верхньої щелепи. Отримані мазки з поверхні верхніх молярів досліджували бактеріологічним методом з подальшою ідентифікацією: мікроскопічною, культуральною, біохімічною. Порівняння ефективності безалкогольного ополіскувача, щодо змін кількісного та якісного складу мікроорганізмів та стану тканин пародонту у пацієнтів, які проходять стаціонарну ортодонтчну терапію, здійснювали після трьох місяців застосування. У дослідження було виявлено грампозитивні та грамнегативні бактерії та гриби *Candida* та з подальшим незначним зменшенням грамнегативних бактерій ( $p=0,999$ ) після застосування ополіскувача. Отримані результати показали зміни таксономічного складу, а саме, зменшення кількості родів бактерій від 11 до 9. Крім зазначених результатів було виявлено зміни на популяційному рівні мікробіоти м'якого зубного нальоту пацієнтів, які використовували безалкогольний ополіскувач протягом трьох місяців, виявлено, зменшення кількості *Streptococcus* spp.  $\alpha$   $p(x \leq T) = 0,9958$ , *Propionibacterium* spp.  $p(x \leq T) = 0,9837$ , щодо *Streptococcus* spp.  $\beta$ , з *Streptococcus* spp.  $\gamma$  зміни були незначними. При порівнянні популяційного рівня мікроорганізмів до та після застосування ополіскувача було виявлено достовірні відмінності ( $p < 0,05$ ). Визначення індексу гінгівіту продемонстрував незначне покращення стану пародонту у обстежуваних. Проведене дослідження має важливе значення для розробки нових стратегій профілактики та лікування захворювань порожнини рота

**Ключові слова:** мікробіоценоз; мікроорганізми; ротова порожнина; гінгівіт; брекет-система