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# Alterations in Oral Microbiota among Orthodontic Patients and Their Impact on Oral Health: A Review

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## Abstract

Currently, there is growing attention toward understanding the biological and microbiological alterations associated with orthodontic therapy. Advances in knowledge of the oral microbiome increasingly enable the identification and characterization of microbial profiles linked to both oral and systemic conditions. This study aims to explore the associations between orthodontic appliances and changes in the composition and quantity of oral microbiota, particularly regarding factors that may predispose patients to caries, periodontal disease, and other infections affecting overall oral and systemic health. Compared with individuals without orthodontic devices, patients undergoing orthodontic treatment exhibited notable qualitative and quantitative differences in both supra- and subgingival plaque throughout the treatment period. Specific elements of fixed appliances, including bonded molar brackets, ceramic brackets, and elastomeric ligatures, were associated with elevated risks of dental caries and periodontal issues. The prevalence of Candida species remains unclear, and research on viral and protozoal components of the oral microbiome in orthodontic patients is limited, warranting further investigation. The findings of this work may assist clinicians in optimizing follow-up schedules and motivating patients to control plaque accumulation, ultimately reducing the incidence of plaque-related oral diseases.

**Keywords:** PCR, Orthodontics, Brackets, Clear aligners, Microbiome, Microbiota, Candida albicans, Oral health, Caries, Periodontitis, Materials, Dental plaque

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## Introduction: Oral Dysbiosis and Associated Oral Conditions in the General Population

In recent decades, the demand for orthodontic treatment has grown, particularly in developed nations [1], driven by both therapeutic and aesthetic considerations, alongside the availability of diverse orthodontic devices and treatment protocols [2-5] tailored to meet individual patient needs. Orthodontic interventions are employed to correct dental malocclusions and craniofacial skeletal discrepancies arising from genetic, familial, or environmental factors, using either fixed or removable appliances.

Recent studies have focused on identifying biomarkers to monitor biological changes during tooth movement before and throughout treatment, highlighting the influence of multiple factors, including oral hygiene practices and dietary habits, on the development of dental and periodontal disorders during orthodontic therapy [6,7].

The primary plaque-related periodontal conditions are gingivitis and periodontitis. Gingivitis represents a non-

destructive, usually reversible inflammation controlled through plaque management, whereas periodontitis—triggered by a combination of genetic and environmental predispositions—persists even after restoring oral hygiene [8], leading to irreversible attachment loss and tooth loss due to sustained local inflammation initiated by periodontopathogenic bacteria [9].

Systematic reviews have demonstrated that clinical indicators of periodontal disease, such as plaque index, bleeding on probing, attachment loss, pocket formation, and gingival recession, often worsen depending on the duration and type of orthodontic treatment, though the extent of reversibility varies post-treatment. Factors contributing to periodontal disease in orthodontic patients include:

- increased difficulty in maintaining oral hygiene,
- plaque accumulation around orthodontic devices,
- bone and periodontal remodeling under orthodontic forces, which can facilitate subgingival plaque buildup and elevate the pathogenic potential of the periodontium [10,11].

Beyond periodontal concerns, plaque retention associated with orthodontic appliances has been linked to a higher risk of dental caries, mediated by transient or persistent shifts in the composition and abundance of oral microbiota, which can lead to oral infections with potential systemic consequences [12,13], thereby making orthodontic patients more susceptible to various pathologies than non-orthodontic individuals [14].

## Characterization of Dental Plaque

Dental plaque exhibits dynamic variations over time and across oral sites, with early versus mature, and supragingival versus subgingival plaques displaying distinct microbial profiles responsible for different diseases, notably caries and periodontal disorders. Dental plaque is, by definition, a polymicrobial biofilm composed of diverse bacterial complexes that benefit from mutual adhesion, coaggregation, and metabolic interactions [15].

Supragingival plaque primarily contributes to enamel and dentin demineralization and the development of caries due to Streptococcus mutans and other cariogenic species, such as Lactobacilli and Actinomycetes as secondary colonizers [14]. Moreover, supragingival plaque facilitates the late-stage colonization of subgingival niches by periodontopathogenic bacteria, which are predominantly gram-negative anaerobes closely associated with periodontal disease [16].

The advent of culture-independent, high-throughput techniques, including reverse transcriptase-polymerase chain reaction (RT-PCR) [17], has significantly expanded knowledge of the oral microbiota. Unlike traditional culture-based methods, these approaches allow simultaneous assessment of a large array of microbial species from multiple sample types—saliva, supra- and subgingival plaque—across diverse populations, providing insights into microbiota changes beyond baseline conditions. The oral microbiota comprises over 600 species, including bacteria, archaea, fungi, viruses, and eukaryotes [18, 19], with 169 species forming the indigenous "core oral microbiome," which is distributed across three main ecological niches: Group 1—buccal mucosa, keratinized gingiva, and hard palate; Group 2—saliva, tongue, tonsils, and oropharyngeal walls; and Group 3—subgingival and supragingival plaque [20].

## Subgingival Plaque and Periodontopathogenic Bacteria

The characterization of subgingival plaque was first detailed in 1998 through checkerboard DNA–DNA hybridization (CDDH) by Socransky *et al.*, who analyzed the subgingival microbiota in adults with and without periodontitis and organized the forty most frequently detected species into distinct complexes [21]. These complexes were classified based on their strong or weak associations with periodontitis and its clinical manifestations and were assigned color labels—red, orange, yellow, green, and purple—a system still widely used today to indicate the pathogenic potential of each bacterial species in periodontal disease.

Periodontal tissue damage is initiated by the "early colonizers," which include bacteria from the green and yellow complexes. These microbes attach to the dental pellicle via fimbriae, facilitating the adhesion and co-aggregation of orange-complex bacteria. The orange complex acts as a "bridge," linking the early colonizers to the red-complex bacteria, while producing toxins and enzymes that promote attachment loss and increase pocket depth, thus creating an environment conducive to colonization by the red-complex species. Red-complex bacteria are "late colonizers" that reside in the deepest periodontal pockets and are strongly correlated with bleeding in advanced periodontitis [21].

Thus, periodontal destruction by red-complex bacteria represents the culmination of a sequential process in which early and bridge species accumulate and co-aggregate, transforming the subgingival niche into a suitable habitat for the pathogenic red bacteria.

## Supragingival Plaque and Periodontopathogenic Bacteria

A decade later, Haffajee, Socransky, and colleagues conducted a similar analysis to explore microbial interactions in supragingival plaque from a control group of periodontally healthy individuals and a test group of

patients with attachment loss [22]. They observed that supragingival plaque in affected patients contained microbial complexes largely resembling those found in subgingival plaque, with minor variations. The colonization sequence begins with Streptococci (yellow complex), followed by Actinomyces species, which are eventually joined by orange- and red-complex bacteria at sites of gingival bleeding and in periodontal pockets. These complexes are present in both supragingival and subgingival plaques and in inflamed as well as non-inflamed sites.

## The Role of Other Microscopic Agents in Gingival and Oral Health

Beyond bacteria, other microorganisms such as viruses, fungi, and protozoa can contribute to gingival disease. Examples include linear gingival erythema linked to Candida albicans in children with AIDS [23] and Entamoeba gingivalis in periodontal tissue destruction [24].

Candida species are known not only for causing various oral and systemic opportunistic infections [25, 26], but they also play a role in caries development due to their synergistic interactions with Streptococcus mutans, frequently observed in mature dental plaque, particularly in children [27].

Viruses, forming the "oral virobiome," are markers of potential infections, as they infect eukaryotic cells and can induce persistent or recurrent effects [28, 29]. Common oral viruses, including Epstein–Barr virus (EBV), Human Cytomegalovirus (CMV), and Herpes Simplex Virus (HSV), are detected at higher levels in patients with severe periodontitis compared to healthy individuals [30, 31]. Several studies have reported viral–bacterial co-occurrences in aggressive periodontitis, such as EBV or CMV with Porphyromonas gingivalis and Aggregatibacter actinomycetemcomitans [32, 33]. These viral infections may induce local immunosuppression, facilitating subgingival colonization and proliferation of periodontopathogenic bacteria, thereby indirectly contributing to periodontal tissue destruction [32, 33].

## Aims

Building on evidence observed in the general population, this study aims to explore the associations between orthodontic appliances and the corresponding qualitative and quantitative shifts in oral microbiota, with particular attention to factors that may predispose patients to caries, periodontal disease, and other infections, ultimately influencing both oral and systemic health in individuals undergoing orthodontic treatment.

## Microbiota Changes in Orthodontic Patients with Fixed Appliances

Multiple systematic reviews have summarized the clinical and microbiological differences observed in saliva and plaque samples when comparing orthodontic patients (cases) with non-orthodontic individuals (controls). Pan *et al.* reported notable disparities in microbial counts between 61 orthodontic patients and 56 non-orthodontic subjects aged 11–17 years [34], identifying the greatest increase in microbial load three months after appliance placement. They also observed a significantly higher prevalence of highly pathogenic Porphyromonas gingivalis fimA genotypes in the orthodontic group, linking this variant specifically to orthodontic gingivitis.

These substantial quantitative and qualitative modifications in plaque, evident as early as one month into treatment, were further corroborated by a systematic review conducted by Lucchese *et al.* [11]. Their findings indicated that while all types of orthodontic appliances can alter oral microbiota during treatment, fixed appliances were associated with more pronounced cariogenic and periodontopathogenic effects compared to removable devices.

## Fixed Appliances and Periodontopathogenic Bacteria

Guo *et al.* [35], in 2017, conducted a meta-analysis to examine subgingival microbial shifts associated with metal fixed orthodontic appliances, comparing patients before, during, and after treatment to controls. They specifically monitored red-complex bacteria (P. gingivalis, Prevotella intermedia, Tannerella forsythia) and A. actinomycetemcomitans. Results showed a significant rise in T. forsythia three months after appliance placement, while a temporary increase in all monitored species was noted at six months. Interestingly, P. intermedia exhibited greater proliferation at the incisors than at molars.

In 2018, Sun *et al.* [36] investigated salivary microbial diversity in 30 patients undergoing full fixed orthodontic therapy and 20 healthy individuals during the mid-phase of treatment (10–12 months). Using real-time PCR with gene-specific primers, they identified two distinctly different microbial profiles. The orthodontic group displayed higher overall microbial diversity, with notable elevations in Pseudomonas spp., P. synxantha, Burkholderia spp., and Veillonella parvula, while levels of S. oralis and Neisseria lactamica remained similar to the control group.

The relationship between clinical periodontal changes and subgingival microbiota was further confirmed by Naranjo *et al.* [37], who analyzed subgingival cultures from 30

patients pre- and post-bracket placement, compared with 30 non-orthodontic controls. Three months after bracket installation, elevated levels of P. gingivalis, P. intermedia/Prevotella nigrescens, T. forsythia, and Fusobacterium spp. were observed in orthodontic patients. Although various gram-negative superinfecting bacteria and enteric rods—including Klebsiella oxytoca, Enterobacter cloacae, Klebsiella pneumoniae, Serratia marcescens, Pseudomonas spp., Enterobacter aerogenes, and Enterobacter gergoviae—were detected in all groups, their presence did not differ significantly between treated and untreated individuals.

Kim *et al.* [38] explored temporal changes in subgingival microbiota in 30 adolescents during the first six months of fixed-appliance therapy (metal brackets, stainless steel ligatures, molar bands) using PCR. From three months onward, at least one periodontopathogen—among A. actinomycetemcomitans, T. forsythia, C. rectus, Eikenella corrodens, P. gingivalis, P. intermedia, P. nigrescens, and Treponema denticola—was detected in all patients. C. rectus and P. nigrescens increased within the first week, whereas T. forsythia appeared later, between three and six months. A. actinomycetemcomitans remained scarce throughout the observation period. Pathogen prevalence was consistently higher in molars compared with incisors.

Lemos *et al.* [39] reported that in adults wearing full fixed appliances, Actinomyces spp. declined after one year, while orange-complex species such as P. intermedia increased; in contrast, red-complex bacteria proportions remained largely stable, as determined via CDDH analysis of subgingival biofilms.

After orthodontic therapy concludes, some researchers have examined whether the oral microbiota revert to their original state or remain altered. Guo *et al.* [35] reported that several months after the removal of fixed appliances, the microbial composition largely returned to pre-treatment levels. Choi *et al.* [40] confirmed these findings using PCR analysis of subgingival plaque from thirty orthodontic patients three months post-appliance removal, compared to thirty healthy controls. They observed that periodontopathogens, especially T. forsythia, C. rectus, and E. corrodens, had increased during treatment but declined after appliance removal, eventually approximating levels seen in untreated individuals. Notably, T. forsythia and P. nigrescens remained slightly elevated, though the differences were not statistically significant.

Lo *et al.* [41] also described changes in microbial composition using culture-based methods in 10 patients aged 10–17 years, from pre-treatment up to twelve weeks after appliance placement. Before treatment and after one year, the flora was predominantly aerobic, including Streptococcus spp., A. odontolyticus, and A. israelii. During the first 2–4 weeks after appliance insertion, facultative aerobic bacteria such as A. actinomycetemcomitans, Actinomyces viscosus, and Capnocytophaga gingivalis, as well as anaerobic species like E. corrodens, Fusobacterium nucleatum, Micrococcus micros, Peptostreptococcus anaerobius, P. gingivalis, and Pseudomonas spp., became dominant.

## Fixed Appliances and Cariogenic Microorganisms

Klaus *et al.* [42] conducted a cross-sectional study in 2016 on 75 adolescents (mean age 14.4 ± 1.8 years) with fixed orthodontic appliances, examining the prevalence of Candida spp., S. mutans, and Lactobacilli in saliva and plaque via culture methods. Participants were classified based on oral hygiene status: good oral hygiene (GOH), poor oral hygiene (POH), and POH with white spot lesions (POH/WL). High prevalence of Candida spp. was observed across all groups (73% in saliva, 61% in plaque), with significantly greater abundance in POH and POH/WL groups. C. albicans dominated (86% of isolates), followed by C. dubliniensis (15%). Both S. mutans and Lactobacilli were found in all salivary samples and 91 percent of plaque samples, with higher counts in POH and POH/WL patients relative to GOH.

Similarly, Topaloglu-Ak *et al.* [43] analyzed salivary S. mutans, Lactobacillus spp., and C. albicans in thirty five children with fixed appliances and thirty four with removable devices. They found a marked increase in S. mutans and Lactobacilli 6 months after appliance placement, with C. albicans levels being significantly higher in the fixed appliance group compared to the removable appliance group.

Andrucioli *et al.* [44] investigated bacterial colonization after 30 days of premolar band placement in 18 patients (ages 11–29) who had undergone 16 months of fixed orthodontic treatment, using checkerboard DNA–DNA hybridization. Cariogenic species such as S. mutans and Streptococcus sobrinus were more abundant than Lactobacillus acidophilus and Lactobacillus casei. Orange-complex periodontopathogens were detected at higher levels than red-complex bacteria, accounting for about 40 percent of the total microbial load.

## Conventional Brackets with Elastomeric and Steel Ligatures, Self-Ligating Brackets, and Ceramic Brackets

Among fixed orthodontic systems, studies have shown that metal brackets secured with elastomeric ligatures tend to accumulate more dental plaque and lead to higher bleeding on probing and elevated plaque index compared with steel ligatures. In a split-mouth study involving 21 patients, Türkkahraman *et al.* [45] observed that teeth with elastomeric rings exhibited greater bleeding than those

ligated with steel. Alves de Souza *et al.* [46] further confirmed these results, finding increased levels of T. forsythia and P. nigrescens around elastomeric ligatures, while P. gingivalis, A. actinomycetemcomitans, and P. intermedia remained similar regardless of ligature type.

Self-ligating brackets have also been examined for their microbial impact. Some reports associate them with elevated plaque index, increased gingival bleeding, and higher counts of gram-positive and gram-negative bacteria—particularly Streptococci and Lactobacilli—although these differences did not reach statistical significance compared with conventional stainless steel-ligated brackets [47, 48]. Regarding caries risk, Jing *et al.* [49] found that S. mutans levels were significantly higher in patients using conventional brackets compared to those with self-ligating brackets over an 18-month follow-up.

Anhoury *et al.* [50] analyzed 24 metallic and 32 ceramic brackets immediately after debonding to compare total bacterial load and the presence of cariogenic and periodontopathogenic species. Overall, P. gingivalis levels were comparable between metallic and ceramic brackets for both anterior and posterior teeth, as were P. nigrescens, Actinomyces odontolyticus, T. forsythia, Actinomyces naeslundii, Capnocytophaga ochracea, Actinomyces israelii, and cariogenic species like S. mutans and L. acidophilus. Distinct differences were observed for eight periodontopathogens: metallic brackets had higher counts of T. denticola, A. actinomycetemcomitans, F. nucleatum ss vincentii, S. anginosus, and E. nodatum, whereas ceramic brackets showed elevated E. corrodens, Capnocytophaga showae, and Selenomonas noxia. Additionally, anterior ceramic brackets had more Streptococcus sanguis, Actinomyces gerencseriae, and Streptococcus constellatus, while posterior metallic brackets had higher C. rectus counts.

## Molar Bands versus Bonded Molar Tubes

Ireland *et al.* [51] studied microbial communities in 24 orthodontic patients aged 11–14 years to compare banded and bonded molars. Using DGGE and 16S rDNA microarray, they found that within three months of appliance placement, both groups exhibited increases in T. denticola and P. nigrescens and decreases in A. actinomycetemcomitans. After treatment, both molar types showed elevated P. gingivalis, T. forsythia, and E. nodatum, but bonded molars additionally had higher levels of C. rectus, Parvimonas micra, A. odontolyticus, and V. parvula. One year post-treatment, banded molars largely returned to baseline microbiota, while bonded molars maintained altered microbial profiles.

Mártha *et al.* [52] examined 25 patients (ages 11–17) during the first two months of fixed treatment, analyzing eleven periodontopathogenic species in subgingival plaque from banded and bonded molars using DNA-strip technology. Across both groups, F. nucleatum (92%), E. corrodens (76%), and Capnocytophaga spp. (C. gingivalis, C. ochracea, C. sputigena) were most prevalent. The other species—A. actinomycetemcomitans, P. gingivalis, P. intermedia, T. forsythia, T. denticola, P. micra, C. rectus, and E. nodatum—were detected less frequently. Notably, E. nodatum appeared in only two bonded molar cases, while E. corrodens, P. micra, T. denticola, and T. forsythia remained stable in banded molars but increased significantly in bonded molars after two months, along with Capnocytophaga spp.

## Microbial Changes in Children Using Functional Orthodontic Appliances

In pediatric interceptive orthodontics, various functional devices are employed, such as those designed for rapid maxillary expansion. Ortu *et al.* [53] assessed how these appliances influence oral microbiota by measuring S. mutans and Lactobacillus spp. in 30 children aged 6–9 years. The participants were divided into three groups: 10 with a rapid palatal expander (RPE), 10 with a McNamara expander, and 10 untreated controls. The study revealed that all children experienced increased levels of Streptococci and Lactobacilli over time, yet the McNamara group showed a particularly pronounced rise in Lactobacilli at six months, surpassing both the RPE and control groups.

## Comparison of Clear Aligners and Fixed Orthodontic Appliances in Adults

A prospective study by Levrini *et al.* [54] in 2015 examined 77 adult patients categorized into clear aligner, fixed appliance, and control groups to evaluate changes in periodontal health and plaque microbiota. The results indicated that patients using clear aligners maintained superior oral hygiene and had milder periodontal indices compared to those with traditional fixed appliances. This finding aligns with the conclusions of Rossini *et al.* [55], whose meta-analysis confirmed that clear aligner therapy is generally associated with better periodontal outcomes than fixed orthodontics.

Focusing on subgingival microbial composition, Lombardo *et al.* [56] conducted a longitudinal study comparing clear aligners and fixed appliances, targeting the quantification of A. actinomycetemcomitans, P. gingivalis, F. nucleatum, C. rectus, T. denticola, and T. forsythia via real-time PCR. While total bacterial loads increased in both groups during treatment, significant growth of C. rectus and F. nucleatum was observed only in the fixed appliance group. Notably, A. actinomycetemcomitans was absent, and P. gingivalis, T. forsythia, and T. denticola appeared sporadically. In a

similar study, Mummolo *et al.* [57] monitored 80 adults (40 using clear aligners and 40 with fixed appliances) and found that cariogenic species such as Streptococci and Lactobacilli increased significantly in the fixed appliance group.

The advantage of removable appliances in maintaining periodontal health has been further reinforced by two meta-analyses by Lu *et al.* [58] and Wu *et al.* [59], which reviewed RCTs and cohort studies comparing fixed and removable orthodontic treatments. Across these analyses, while gingival index (GI) and sulcus probing depth (SPD) showed no significant differences between groups, patients wearing clear aligners consistently demonstrated lower plaque index (PI) and sulcus bleeding index (SBI), highlighting reduced plaque accumulation and less gingival inflammation throughout treatment.

## Protozoa, Fungi, and Other Microbial Populations in Orthodontic Patients

Beyond the well-studied cariogenic and periodontopathogenic bacteria, research has also examined other microorganisms in orthodontic patients, including opportunistic pathogens capable of affecting oral and systemic health. In 2019, Perkowski *et al.* [60] conducted a culture-based analysis of periodontal swabs from two age groups: 25 children (6–13 years) using removable orthodontic devices and 25 adolescents/young adults (14–23 years) with fixed appliances, each compared to 50 healthy age-matched controls. The results revealed that younger patients exhibited higher caries indices (DMFT), while older participants displayed elevated bleeding on probing, regardless of treatment status. Patients with fixed appliances demonstrated a greater presence of Enterococcus faecalis, Enterococcus faecium, Staphylococcus aureus, and Escherichia coli, along with Gram-negative species such as Enterobacter cloacae, Pantoea agglomerans, and Klebsiella spp., as well as multiple Candida species. Among these, C. albicans was particularly associated with higher bleeding scores and poor oral hygiene. Rare occurrences of protozoa (Trichomonas tenax and Entamoeba gingivalis) were reported in a few older subjects, while cysts of Acanthamoeba were identified exclusively in three patients with fixed appliances.

Previous studies have consistently highlighted the higher prevalence of Candida in patients with fixed appliances compared to those with removable devices or no orthodontic treatment, as noted by Topaloglu *et al.* [43] and other authors [61, 62]. Klaus *et al.* [42] also observed a strong link between poor oral hygiene and elevated Candida presence, corroborated by other literature [63]. Arslan *et al.* [62] reported that in a cohort of adolescents with fixed appliances, C. albicans was the dominant species (73.8 percent), followed by C. tropicalis, C. krusei, and C. kefyr (7.14 percent) and C. parapsilosis (4.76%). Grzegocka *et al.* [64] similarly found that 59% of 17 orthodontic patients carried Candida, with a notable correlation to heavy plaque accumulation.

Conversely, recent controlled trials have questioned the magnitude of these changes. Sanz-Orrio-Soler *et al.* [65] followed 124 patients undergoing fixed appliance therapy, analyzing oral Candida colonization before, during, and after treatment. Their culture-based results showed no significant overall increase in C. albicans, although ceramic brackets were associated with a slightly higher frequency. Similarly, Tapia *et al.* [66] reported that only 6.7% of 90 patients (mean age 20.6 ± 7.1 years) carried C. albicans or C. tropicalis, suggesting limited colonization in this population.

## Discussion

This review analyzed current literature to identify the microbial alterations that occur during and after different orthodontic treatments. Compared with individuals not undergoing orthodontic therapy, patients with orthodontic appliances exhibited notable differences in both the composition and quantity of plaque throughout treatment. Removable devices appeared less detrimental to periodontal health and caries risk, likely because they can be taken out to facilitate proper oral hygiene, despite being worn nearly full-time. These findings align with a recent meta-analysis by Jiang *et al.* [67], which reported that clear aligner users maintain better periodontal parameters and overall oral health compared to those with fixed appliances, suggesting that aligners may be the preferred option for adults or patients at high risk for gingivitis or periodontitis [68]. Additionally, patients with fixed devices showed higher counts of Streptococci and Lactobacilli, indicating an elevated risk of dental caries relative to clear aligner wearers [57].

Within fixed appliance therapy, conventional brackets have been associated with higher levels of S. mutans, placing patients at increased risk for white spot lesions and caries compared to self-ligating systems [49]. Elastomeric ligatures and ceramic brackets were also linked to poorer oral conditions and elevated counts of cariogenic and periodontopathogenic species. Consequently, while self-ligating brackets appear microbiologically safer, elastomeric ligatures and ceramic brackets should be considered higher-risk for periodontal and carious complications.

Microbial changes in fixed appliances begin within the first weeks of therapy. Early colonizers such as Streptococci and Actinomyces spp. dominate initially, alongside species including C. rectus and P. nigrescens (orange complex), A. actinomycetemcomitans, A. viscosus, Capnocytophaga gingivalis, and anaerobes like

Eikenella corrodens, F. nucleatum, Micrococcus micros, Peptostreptococcus anaerobius, P. gingivalis, and Pseudomonas spp. By three months, orange complex species increase to roughly 40% of total bacterial counts, while red complex bacteria—P. gingivalis, P. intermedia, and T. forsythia—as well as Lactobacilli, rise significantly after six months. Most studies indicate that after treatment concludes, the oral microbiota largely reverts to baseline, suggesting that the microbial shifts during orthodontic therapy are temporary [35]. However, some evidence indicates long-term effects on periodontal health in patients with bonded molar tubes compared to traditional molar bands [51], highlighting the need for careful post-treatment follow-up in these cases.

Regarding Candida species, several studies have reported a substantial prevalence in orthodontic patients, while others found minimal colonization. These discrepancies may be due to small sample sizes and reliance on culture-based detection, which can underestimate yeasts that are difficult to grow, particularly when present in low abundance. Therefore, larger-scale studies using high-throughput techniques are recommended to more accurately characterize the oral mycobiome in orthodontic populations. It is important to note that Candida typically exists as a commensal organism; however, under local or systemic disturbances, it can become opportunistically pathogenic, potentially affecting patient health to varying degrees [26].

A study also detected protozoa, including Trichomonas tenax, Entamoeba gingivalis, and Acanthamoeba spp., which are not typically found in the oral cavity; their opportunistic colonization can be linked to severe systemic conditions such as lung abscesses, granulomatous encephalitis, and keratitis, and their proteolytic activity can harm oral mucosa and penetrate periodontal tissues [58]. Regarding the virobiome in orthodontic patients, the literature remains largely unexplored, with no substantial reports on viruses associated with orthodontic treatment; nevertheless, considering the viral–bacterial interactions observed in periodontitis [32], it is plausible that HSV, EBV, and CMV—viruses commonly detected in children and adolescents [69, 70]—could contribute to periodontal damage when present alongside high levels of periodontopathogenic bacteria.

## Conclusions

Changes in the microbial plaque of orthodontic patients, both in composition and abundance, can be observed as early as one week after treatment initiation and become more pronounced after three months, with stable colonization initially by orange-complex species followed by red-complex species. For patients undergoing fixed orthodontic therapy, it is particularly important to reinforce oral hygiene practices and schedule frequent clinical evaluations during the early months to prevent the development, maturation, and organization of cariogenic and periodontopathogenic bacteria in dental plaque.

The findings of this review align with those reported by Müller *et al.*, who highlighted that while periodontal alterations are reversible, enamel demineralization and white spot lesions tend to persist, supporting the adjunctive use of antibacterial orthodontic bonding systems to help maintain optimal dental health in orthodontic patients [71].

This review specifically considered healthy, immunocompetent individuals without additional local or systemic conditions; thus, it can be anticipated that patients with special needs, immune deficiencies, or other oral or systemic comorbidities may be more susceptible to the cariogenic and periodontopathogenic consequences of mature plaque, and a high load of Candida spp. could contribute to fungal stomatitis or systemic candidiasis in vulnerable populations.

Given the growing evidence linking oral dysbiosis with systemic diseases [72,73], further research is warranted on the less studied components of the oral microbiota—such as fungi, viruses, and protozoa—as well as on at-risk populations undergoing orthodontic treatment, in order to prevent both oral and systemic complications and to provide personalized dental care tailored to each patient's unique microbiological and clinical profile [74].

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