

# Exploration of the Formation Characteristics of Biofilm During Invisible Orthodontic Treatment and Its Correlation with Orthodontic Pain

Bojie Chen

School of stomatology, Sechenov University, Moscow, Russia

## ABSTRACT

Invisible orthodontics has become one of the mainstream methods of orthodontics due to its advantages such as aesthetics, comfort, and removal. However, orthodontic pain still seriously affects patients' treatment compliance. Oral biofilm is the core of oral microbiota, and its formation and evolution are regulated by factors such as appliance wearing and changes in oral environment. This article reviews the research on changes in oral microbiota during invisible orthodontic treatment, clarifies the formation stages, components, and structural characteristics of biofilms, analyzes their association mechanism with orthodontic pain, and explores the impact pathway of biofilm inflammation on periodontal pain perception. Research has found that wearing invisible orthodontic appliances can alter the local physical barriers and microenvironment of the oral cavity, promoting the formation of pathogenic bacterial biofilms mainly composed of streptococci and actinomycetes; Biofilm releases inflammatory factors and metabolic products that stimulate periodontal nerves, or worsen periodontitis, thereby exacerbating orthodontic pain. This article aims to provide theoretical basis for clinical regulation of oral biofilm to prevent and alleviate pain related to invisible orthodontic treatment, and to provide reference for optimizing treatment plans and improving patient experience.

## KEYWORDS

Invisible orthodontic treatment; Biofilm; Orthodontic pain; Inflammatory response; Oral microbiota

## 1. INTRODUCTION

The core of orthodontics is to guide teeth to the ideal position through external force. Pain, as the most common adverse reaction, has a high incidence rate and often occurs in the early stages of treatment and 1-3 days after follow-up visits. Periodontal swelling and pain, chewing discomfort, and in severe cases, it can affect eating and sleep, and even reduce treatment compliance. Invisible orthodontic treatment utilizes computer-aided design to achieve precise tooth movement, and the appliance is removable and has minimal stimulation to oral soft tissues, reducing the risk of pain to a certain extent. However, some patients still experience significant pain, and the specific mechanism is not yet clear [1].

The oral cavity is a complex microecological system in the human body, where hundreds of microorganisms interact to form biofilms that attach to the tooth surface, orthodontic appliances, and mucous membranes, making it an important part of the oral microecology. Biofilm is a community formed by microorganisms adapting to the environment, wrapped in extracellular polymers, with stable structure and strong stress resistance. Its pathological impact on the host is more significant than that of planktonic microorganisms. Wearing invisible orthodontic appliances can alter the local physical environment of the oral cavity (such as bite and difficulty in cleaning), chemical environment

(such as pH value and nutrient distribution), and microbial community structure, providing favorable conditions for biofilm formation and proliferation.

The current academic research on invisible orthodontic treatment mainly focuses on the mechanical mechanisms, pain relief pathways, or the correlation between biofilms and periodontal disease. However, there is still a lack of exploration on the direct correlation between the formation characteristics of biofilms during invisible orthodontic treatment and orthodontic pain.

Clarifying this association is not only the key to elucidating the essence of pain occurrence, but also an important prerequisite for developing precise intervention plans. This article reviews recent research trends, analyzes the formation patterns of biofilms in invisible orthodontic environments, explores their intrinsic relationship with orthodontic pain, and provides useful references for optimizing clinical treatment processes and improving patient comfort during orthodontic treatment.

## **2. THE IMPACT OF INVISIBLE ORTHODONTIC TREATMENT ON THE ORAL MICROECOLOGICAL ENVIRONMENT**

Oral microbiota is an organic system composed of microorganisms, host tissues, and the environment, which balance and dynamically coexist with each other. Invisible orthodontic appliances, as exogenous foreign objects entering the oral cavity, disrupt this balance from physical, chemical, and biological perspectives, providing a breeding ground for biofilm growth.

On a physical level, orthodontic appliances cover the dental crown, altering the anatomical shape and oral spatial structure of the teeth, creating a narrow gap between the tooth surface and the appliance [2]. This gap will hinder saliva flow and erosion, weaken oral self-cleaning ability, and provide a stable attachment carrier for microbial colonization. Clinical studies have shown that the saliva flow rate in the contact area between the tooth surface and the orthodontic appliance is significantly reduced after wearing, and the mechanical clearance effect of saliva on microorganisms is weakened, making it easier for them to accumulate on the tooth surface and orthodontic appliance surface. At the same time, orthodontic appliances alter the distribution of occlusal pressure, and local periodontal mechanical stimulation affects the oral mucosal barrier function, creating conditions for microbial invasion.

At the chemical level, orthodontic materials such as polycarbonate and polyurethane undergo slight degradation in the oral cavity, releasing small amounts of chemicals; After wearing it, the difficulty of cleaning increases, and food residues accumulate and ferment in the gaps, leading to a decrease in the local pH value of the oral cavity. Tests have shown that the overall pH value of the patient's oral cavity is lower than the normal neutral range. The acidic environment not only affects enamel mineralization, but also alters microbial metabolic activity, promoting the proliferation of acid producing pathogenic bacteria. In addition, proteins and polysaccharides in saliva can form a conditioned film on the surface of orthodontic appliances, providing a basis for initial microbial adsorption and accelerating biofilm formation.

At the biological level, wearing orthodontic appliances can disrupt the balance of the oral microbiota. The normal oral cavity is mainly composed of beneficial bacteria such as lactobacilli and bifidobacteria [3], which compete for nutrients and produce antibacterial substances to inhibit pathogenic bacteria; However, in the invisible orthodontic environment, the cleaning efficiency decreases, the microenvironment changes, pathogenic bacteria such as streptococcus and actinomycetes increase, beneficial bacteria decrease, and microbial imbalance occurs. The changes in the microbiota not only provide abundant microbial populations for the formation of biofilms, but also further enhance their pathogenic potential and lay potential risks for subsequent inflammation and pain.

### **3. CHARACTERISTICS OF BIOFILM FORMATION DURING INVISIBLE ORTHODONTIC TREATMENT**

The four stages of initial adsorption, proliferation, maturation, and detachment of biofilm like components are characterized by unique patterns in the formation of invisible orthodontic environments, mainly reflected in the formation rate, component composition, and structural characteristics.

In terms of formation speed, biofilm formation in invisible orthodontic environments is significantly faster than in natural oral cavities. The condition film on the surface of the orthodontic appliance provides a rapid attachment carrier for microorganisms, and the oral self-cleaning ability decreases, reducing the removal of microorganisms and significantly shortening their initial adsorption time on the tooth surface and orthodontic appliance surface. Experimental observations show that after wearing orthodontic appliances for several hours, there is a significant accumulation of microorganisms on the tooth surface. After 24 hours, a certain thickness of biofilm has formed, while the accumulation of biofilm in normal oral cavity is significantly less within the same period of time. In addition, periodic force application during invisible orthodontic treatment can cause slight periodontal damage, and the inflammatory factors released by tissue repair can also attract microbial aggregation, accelerating the formation and proliferation of biofilms.

In terms of composition, the biofilm during invisible orthodontic treatment is dominated by bacteria, and the microbial community is more complex. The proportion of beneficial bacteria in normal oral biofilms is high, while in invisible orthodontic environments, the proportion of streptococcus genera (such as *Streptococcus mutans* and *Streptococcus pyogenes*) significantly increases, and the proportion of actinomycetes is also large, including a small amount of periodontal pathogenic bacteria such as *Porphyromonas gingivalis*. These pathogenic bacteria form stable communities by producing extracellular polysaccharides, proteins, and other interconnected substances. At the same time, biofilms contain a large amount of extracellular matrix, mainly composed of polysaccharides, proteins, and nucleic acids, which account for the majority of the dry weight of biofilms. They can provide protection for microorganisms, regulate metabolism, and enhance stress resistance [4].

From a structural perspective, the biofilm related to invisible orthodontic treatment presents a unique multi-layered and three-dimensional structure, with higher adhesion to the tooth surface and orthodontic appliances. By observing with a scanning electron microscope, it can be seen that mature biofilms are clearly divided into three layers: the surface layer is a loose microbial community formed by the adsorption and aggregation of planktonic microorganisms; The middle layer is a dense core where microorganisms rely on the tightly interwoven extracellular matrix; The bottom layer is a substrate structure rich in adsorbed proteins and polysaccharides, which makes it difficult for the biofilm to be completely removed by mechanical mean [5].

What is more noteworthy is that there are many subtle channels hidden inside the biofilm, which undertake the functions of substance exchange and metabolic product transport. This not only provides convenience for the survival and proliferation of microorganisms, but also helps them avoid the effects of antibiotics and mouthwash, thereby significantly enhancing their pathogenic efficacy.

### **4. THE MECHANISM OF ORTHODONTIC PAIN OCCURRENCE**

Orthodontic pain is a physiological and pathological response of the host to mechanical stimulation during tooth movement. The mechanism is complex and is related to periodontal mechanical damage, inflammatory response, and nerve endings stimulation. It is caused by multiple factors working together [6].

Mechanical stimulation is the initiating factor of pain. Invisible orthodontic treatment relies on the appliance to continuously guide the movement of teeth with light force, and the external force is

transmitted to the periodontal membrane, causing the periodontal membrane fibers to stretch and compress, resulting in damage to periodontal membrane cells. These cells undergo morphological changes and increase membrane permeability upon stimulation, releasing cytokines such as interleukin-1  $\beta$  (IL-1  $\beta$ ), tumor necrosis factor -  $\alpha$  (TNF -  $\alpha$ ), and prostaglandin E2 (PGE2), which act as pain signal initiators to activate periodontal nociceptors and trigger pain. At the same time, when teeth move, alveolar bone remodeling occurs, and osteoclasts absorb and release acidic substances from the alveolar bone, further stimulating periodontal nerve endings and exacerbating pain [7].

Inflammatory response is the core mediating factor of pain. The cytokines released by damaged periodontal ligament cells can recruit inflammatory cells such as neutrophils and macrophages to the site of injury, leading to local inflammation. Inflammatory cells release more inflammatory and chemokines, forming an inflammatory cascade that amplifies pain signals. Clinical tests have shown that the concentrations of IL-1  $\beta$ , TNF -  $\alpha$ , and PGE2 in the saliva of patients during the initial stage of treatment are significantly higher than before treatment, and the concentrations are positively correlated with the degree of pain. These inflammatory factors can also increase vascular permeability, leading to periodontal congestion and edema, further exacerbating pain perception.

Changes in sensitivity of nerve endings are also involved in the occurrence of pain. Normally, periodontal nerve endings have a certain threshold for mechanical and chemical stimuli, while mechanical damage and inflammation in orthodontics can lower the pain threshold, causing strong pain even in mild stimuli. In addition, inflammatory factors can also affect the release of neurotransmitters, such as increasing the expression of pain related neurotransmitters such as calcitonin gene-related peptide (CGRP) and substance P, and prolonging pain duration. Clinical observations have found that pain related to invisible orthodontic treatment often peaks 24-48 hours after applying force, and then subsides with the disappearance of inflammation and recovery of nerve sensitivity, consistent with the trend of changes in inflammatory cytokine concentration.

## **5. THE CORRELATION MECHANISM BETWEEN BIOFILM FORMATION AND ORTHODONTIC PAIN**

The abnormal formation of biofilms in invisible orthodontic treatment is closely related to orthodontic pain. The core is to amplify pain signals by triggering and exacerbating periodontitis, which can be achieved through three aspects:

Firstly, pathogenic bacteria in the biofilm directly induce periodontal inflammation, exacerbating pain. Invisible orthodontic biofilms are mainly caused by pathogenic bacteria such as streptococcus and actinomycetes, which proliferate extensively under the protection of the biofilm and release toxic products such as lipopolysaccharides and enzymes. Lipopolysaccharide, as a potent inflammatory inducer, can bind to Toll like receptors on the surface of periodontal ligament cells, activate inflammatory signaling pathways, and promote the release of inflammatory factors such as IL-1  $\beta$  and TNF -  $\alpha$ ; Proteases and collagenases produced by pathogenic bacteria can degrade periodontal tissue components, disrupt periodontal membrane structure, exacerbate tissue damage, and activate nociceptors. Clinical research shows that patients with significant biofilm accumulation have a higher incidence and longer duration of pain.

Secondly, biofilm disrupts the balance of oral microbiota, indirectly amplifying inflammation and pain. The normal balance of oral microbiota is crucial for periodontal health, and the formation of biofilms can disrupt this balance, leading to excessive proliferation of pathogenic bacteria and a decrease in beneficial bacteria [8]. The antibacterial substances secreted by beneficial bacteria gradually decrease, while the toxicity of pathogenic bacteria increases accordingly; Imbalance in the microbiota can lead to metabolic disorder, and the organic acids, ammonia, and other substances produced by pathogenic bacteria can reshape the periodontal microenvironment, weaken tissue defense barriers, and create conditions for inflammation to occur. This microecological imbalance

overlaps with inflammation induced by mechanical stimulation, forming an "inflammation amplification effect" that significantly increases the pain sensitivity of periodontal tissue.

Thirdly, the structural characteristics of biofilms can also prolong the duration of pain. Its multi-layered three-dimensional structure combined with the protection of extracellular matrix makes it difficult for pathogenic bacteria to be washed away by saliva and removed by conventional cleaning methods, becoming a persistent source of infection. Even if patients clean up regularly, it is difficult to completely eradicate mature biofilms, and pathogenic bacteria continue to release toxic products, ultimately leading to chronic inflammation. Chronic inflammation stimulates the periodontal nerve endings for a long time, causing them to be in a highly sensitive state, which not only exacerbates acute pain but also prolongs the duration of pain; And the internal channels of the biofilm facilitate substance exchange and signal transmission, promoting the continuous production of inflammatory factors, forming a vicious cycle of "pain inflammation biofilm formation", further exacerbating the pain experience.

## **6. ORTHODONTIC PAIN RELIEF STRATEGY BASED ON BIOFILM REGULATION**

By combining the correlation mechanism between biofilms and orthodontic pain in invisible orthodontic treatment, clinical strategies such as optimizing oral hygiene, regulating microecology, and targeting inhibition of biofilm formation can be used to reduce inflammation induced by biofilms from the source, alleviate pain, and improve patient treatment compliance.

Optimizing oral hygiene is the foundation for controlling biofilms. Orthodontics increase the difficulty of cleaning, and patients need to be guided to perform targeted cleaning: after daily removal, use a soft bristled toothbrush dipped in specialized cleaning agents to scrub the orthodontic appliance, removing biofilms and food residues; Soak in specialized disinfectant tablets 1-2 times a week to kill residual pathogenic bacteria [9]. Tooth surface cleaning requires the Babbitt brushing method, combined with dental floss, flossers, and other tools, with a focus on cleaning the contact gap between the tooth surface and the orthodontic appliance to reduce microbial colonization. Research has shown that dental irrigators can reduce the accumulation of dental biofilms, lower the concentration of salivary inflammatory factors, and effectively alleviate pain.

Regulating the oral microbiota is the core of inhibiting biofilms. By supplementing probiotics such as Lactobacillus and Bifidobacterium to expand the beneficial bacterial population, the balance of the microbiota can be reshaped. Probiotics compete for nutrients and produce antibacterial substances to inhibit the proliferation of pathogenic bacteria and biofilm formation. Clinical observations have shown that regular consumption of probiotics can reduce the number of pathogenic bacteria such as streptococcus, slow down biofilm formation, and lower the concentration of inflammatory factors and the incidence of pain. Dietary regulation can also assist in improving the microbiota. Patients need to reduce their intake of high sugar and high starch foods to avoid providing nutrients for pathogenic bacteria, while increasing their intake of vitamin C and D to enhance periodontal tissue resistance.

For patients who still experience significant pain after basic oral hygiene and microbiota regulation, targeted interventions such as mouthwash containing chlorhexidine and clopidogrel chloride can be used under the guidance of a doctor to destroy the extracellular matrix of biofilms and inhibit microbial adsorption and proliferation. However, the duration and concentration of use should be controlled to avoid adverse reactions such as mucosal dryness and taste changes [10]. Topical application of gel containing PGE2 inhibitor can directly inhibit the release of inflammatory factors and relieve pain. Combined with mouthwash, it can shorten the duration of pain and relieve pain. Regular oral examinations and professional cleaning are indispensable. Doctors use ultrasound cleaning to remove stubborn biofilms, timely treat early inflammation, and avoid worsening pain.

## 7. CONCLUSION

In the clinical practice of invisible orthodontic treatment, orthodontic pain has always been an important issue affecting patients' treatment experience, and the abnormal formation of oral biofilm is a key influencing factor. Through research, it can be seen that invisible orthodontic treatment changes the original microecological balance of the oral cavity, promoting the rapid formation of biofilms, a high proportion of pathogenic bacteria, and a more stable structure; Orthodontic pain is mainly caused by mechanical stimulation induced periodontitis and nerve endings sensitivity, while biofilms can directly induce inflammatory reactions, amplify microecological imbalances, maintain chronic infection status, and further exacerbate the occurrence and duration of pain.

Based on this intrinsic correlation, the intervention strategies implemented in clinical practice, such as optimizing oral hygiene, regulating oral microbiota, and targeting the inhibition of biofilms, can effectively curb the pathogenic activity of biofilms, reduce inflammatory reactions, break the vicious cycle of "biofilm inflammation pain", and truly enhance patients' comfortable treatment experience.

However, current related research still has limitations, such as the quantitative correlation between specific components of biofilms and pain levels, and the dynamic changes in biofilms during different orthodontic stages, which are not yet clear. In the future, we may rely on cutting-edge technologies such as microbiology and molecular biology to deeply analyze the regulatory mechanisms of specific pathogenic bacteria and pain related inflammatory factors in biofilms, laying a scientific foundation for precise intervention; At the same time, in clinical practice, more attention should be paid to tailoring personalized plans, combining the individual characteristics of the patient's oral microbiota and pain tolerance threshold, selecting appropriate regulatory pathways, and further improving the clinical efficacy and patient satisfaction of invisible orthodontic treatment.

This research perspective from the perspective of biofilm regulation provides new ideas for optimizing the invisible orthodontic process, and has practical significance for promoting the development of precision and comfort treatment in orthodontics.

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