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Decoding Oral Microbiome: A Comprehensive Review

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Abstract

The oral microbiome is a dynamic community of microorganisms residing within the oral cavity, crucial for maintaining oral health and influencing systemic well-being. Recent advancements in sequencing technologies and computational biology have unveiled the intricate composition, diversity, and functional dynamics of the oral microbiome, offering unprecedented insights into its role in health and disease. This review synthesizes key findings surrounding the oral microbiome, highlighting methods of studying the oral microbiome. By shedding light on the complexities of the oral microbiome, this review underscores the importance of ongoing research in this field and its implications for advancing oral healthcare practices.

INTRODUCTION

Over three hundred years ago, Antony van Leeuwenhoek achieved a significant milestone as the initial observer of microbes, potentially bacteria, within his own dental plaque using a microscope of his own making. This groundbreaking observation laid the groundwork for the field of microbiology. Subsequent discoveries have emphasized the intricate interactions between these microbes and their human hosts. According to recent estimates, approximately 3.8×10^{13} microorganisms inhabit the human body, constituting roughly half of the total cells present. Collectively, these organisms are referred to as the human microbiome. [1,2,3]

The ubiquitous presence of the human microbiome across diverse body sites is widely acknowledged. According to data from the Human Microbiome Project [4], approximately 34% of primary microbial habitats are attributed to the human skin, 25% to the gastrointestinal tract, and 20% to the cavities of the head and neck region. Within these key human habitats, the oral cavity poses a unique challenge for microbial survival. Subject to substantial daily fluctuations in nutrient availability, temperature, pH levels, as well as mechanical forces from activities like mastication and hygiene practices, and chemical exposure from oral hygiene products, pharmaceuticals, or harmful substances [5], the oral cavity nevertheless maintains a complex and resilient ecosystem. This environment fosters the proliferation of diverse micro-colonizers that thrive amidst its dynamic conditions – collectively comprising the oral microbiome.

The oral microbiome encompasses a diverse community of microorganisms, comprising bacteria, fungi, viruses, archaea, and protozoa, totaling up to approximately 1000 species, as defined by various studies [1, 6]. However, the prevalence of bacteria within the oral microbiome has led to the term "core microbiome" being primarily associated with bacterial species [7]. Consequently, most literature tends to focus exclusively on oral bacteria, often overlooking other components such as fungi, archaea, protozoa, and viruses. Despite the limited research available on these additional biomes, they remain pertinent [1, 8]. Therefore, we will examine each of these biomes within the context of the oral cavity.

Evolution of the Oral Microbiome::

Typically, the fetus resides in a sterile environment within the womb[9]. Upon delivery, the newborn encounters microflora from the mother's uterus and vagina, and subsequently from the surrounding atmosphere. Although there is a high likelihood of contamination, the oral cavity of the newborn remains sterile initially. However, this changes rapidly as the baby begins feeding, introducing microorganisms into the mouth. This marks the initiation of the process of acquiring resident oral microflora [10].

The initial colonizers, referred to as pioneer species, include organisms like Streptococcus salivarius. During the first year of life, the oral cavity is predominantly colonized by aerobes, which may encompass species such as Streptococcus, Lactobacillus, Actinomyces, Neisseria, and Veillonella. As teeth erupt, these microorganisms can adhere to non-shedding surfaces, with further colonization occurring as more teeth emerge. Gingival crevices develop, providing niches for periodontal microbes to inhabit. Plaque accumulation is observed on various tooth surfaces, including smooth surfaces and pit and fissures, facilitating the establishment of different microbial colonies. This process fosters high species diversity and microbial succession over time. As individuals age and lose all their teeth, the oral flora reverts to a composition resembling that of a pre-tooth eruption child[11] .

Bacteria form complex communities by adhering not only to oral surfaces but also to each other. The composition and stability of these communities are influenced by specific partner relationships. Factors such as selective adherence to tooth surfaces or epithelium, as well as cell-to-cell binding, play crucial roles in shaping early community composition. Interactions between organisms lead to changes in the local environment, representing the initial stages of oral disease development [12].

Host-Microbial Interplay in the Oral Environment

The oral microbiome, with its intricate network of interspecies interactions, thrives within the dynamic environment of the oral cavity. Consequently, it not only facilitates microbial interspecies communication but also engages in interactions with the oral cavity itself, establishing a symbiotic relationship with the human host [3]. These interactions significantly influence the microbial composition, thereby impacting the host's health and disease status [5].

The term "Oralome" to encompass all interactions occurring between the oral microbiome and the host. For instance, it is well-established that the oral microbiota play a crucial role in shaping and maturing the oral immune response. The host immune system must defend against Pathogenic microbes while simultaneously maintaining a harmonious relationship with commensal oral microbes [13]. However, it is essential for the immune system to strike a delicate balance between pathogen eradication and preventing an unwarranted immune response against the host's own tissues and commensal organisms. Mounting an overly aggressive immune response against harmless microbes would be energetically costly and potentially harmful to host tissues [14]. In return, certain oral commensals serve as "sensors," "mediators," and "killers," playing coordinated roles in initiating antagonistic actions against pathogens to prevent their colonization—a phenomenon known as colonization resistance [15]. For instance, Streptococcus salivarius antagonizes Streptococcus pyogenes, the primary etiological agent of pharyngitis, thus preventing its colonization and subsequent pharyngitis development . This mutual protection indicates that the host immune system has evolved to tolerate and preserve beneficial bacteria [16]. Additionally, some mucosally-adherent species can shield the host from carcinogenic metabolites.

Furthermore, beyond its positive effects on oral health, the oral microbiome plays a crucial role in influencing the systemic health of the host . For example, certain oral bacteria contribute to an entero-salivary nitrate-nitrite-nitric oxide cycle, where dietary nitrate in saliva is converted into nitrite and nitric oxide [17]. This process has been linked to cytoprotection, vasodilation, antithrombotic effects, immune modulation, and regulation of blood pressure. Additionally, it may have beneficial effects on outcomes related to myocardial infarction, heart failure, pulmonary hypertension, and vascular hypertrophy, including reductions in infarct size and improvements in cardiac function. Interestingly, high levels of Rothia and Neisseria genera have been associated with the maintenance of nitric oxide homeostasis and cardiovascular health, while elevated levels of Prevotella and Veillonella genera have been linked to disruptions in homeostasis.

Moreover, studies utilizing 16S rDNA sequencing have identified specific microbial patterns indicative of "healthy oral microbiota" [17,18]. In this context, a healthy oralome—characterized by symbiotic interactions between humans and these microorganisms—represents an example of eubiosis [4,8].Eubiosis promotes beneficial mutualism or commensalism without causing harm to either the microorganisms or the host. Despite this stability, it's important to note that the oralome (oral microbiome) may not exhibit uniformity across all individuals. Recent studies indicate the presence of up to five distinct clusters in the salivary oralome of various healthy individuals. However, a comprehensive population-based study involving over 2,300 individuals identified only two primary salivary community types: one associated with oral health and the other linked to oral diseases. These findings suggest that the demarcation between health (eubiosis) and disease (dysbiosis) may be intricate, extending beyond natural heterogeneity observed in healthy individuals [18,19].

Common Diseases and Oral Biofilm Dysbiosis:

In 1978, the term "biofilm" gained significance following J. W. Costerton's publication detailing bacterial adhesion, referring to matrix-enclosed bacterial communities pivotal for understanding bacterial interactions with the environment. Subsequent studies have demonstrated that bacteria within biofilms often exhibit distinct phenotypes compared to their planktonic counterparts. Within biofilms, there is extensive exchange of

metabolites, trafficking of signals, and varied interactions among different species. The integration of biofilm theory into oral microbiology spurred researchers to examine dental plaque more closely [20,21].

In the realm of dental caries, the widely accepted 'ecological plaque hypothesis' stands as a prominent theory regarding its origins. According to this hypothesis, frequent consumption of sugars prompts commensal bacteria to adapt to a more acidic micro-environment, fostering the proliferation of aciduric bacterial species, particularly streptococci and lactobacilli. Concurrently, this acidic environment inhibits the growth of beneficial organisms that thrive at neutral pH levels. This adaptation to a dysbiotic microbiome is facilitated by an increase in certain species within the Streptococcus, Lactobacillus, and Actinomycetes genera, which are implicated in the development of dental caries in adults. For example, compared to caries-free children, those with severe early childhood caries exhibit heightened levels of Streptococcus, particularly S. mutans, along with Leptotrichia, Bifidobacterium, Porphyromonas, Leptotrichia, Stomatobaculum, Prevotella, and Selenomas genera. Notably, S. mutans has garnered significant attention for its cariogenic properties and is widely recognized as one of the primary pathogens associated with caries. However, it's important to acknowledge that other species may also contribute to the disease, as S. mutans can persist in the oral cavity without evident demineralization and caries, while caries can manifest even in the apparent absence of this species [22,23].

Zargar et al. identified 18 species in root canals of patients with irreversible pulpitis, including Dialisterinvisus and P. gingivalis, with Lysinibacillus fusiformis detected for the first time. This suggests a potential ecological succession during caries progression. The dysbiotic microbiome, led by S. mutans, produces acids, primarily lactic acid, demineralizing the tooth surface and leading to decay. Pathogenic bacteria, including S. mutans, can invade tooth pulp, causing pulpitis. Inside the pulp, dysbiotic bacteria may enter the circulatory system, potentially causing transient bacteremia. Oral streptococci, particularly S. mutans, S. sanguinis, and S. mitis, are implicated in infective endocarditis, although a direct link between S. mutans from dental caries and infective endocarditis remains uncertain. [24,25]

Unlike caries, periodontitis lacks a single etiological hypothesis, contributing to ongoing uncertainty about its initiation and the specific microbial drivers despite decades of study. Five primary hypotheses have been proposed for periodontitis initiation and pathogenesis. The 'ecological plaque hypothesis,' introduced by Marsh in 1994, combines concepts from the Specific and Non-specific Plaque Hypotheses. It suggests that periodontal disease results from a microbial imbalance induced by ecological stress, leading to an enrichment of periopathogens. This hypothesis posits that a healthy microbiome can only support minimal levels of periodontal pathogens, as they are unable to compete with predominant saccharolytic bacteria. External factors can disrupt this balance, increasing the bacterial load in the subgingival microbiome and triggering a host inflammatory response (gingivitis). This response alters subgingival nutrient status, enriching proteolytic Gram-negative bacteria, leading to host tissue degradation and a continued cycle of destruction, culminating in periodontitis. However, this hypothesis does not address the role of host genetic factors in plaque composition and disease susceptibility [26].

The 'keystone pathogen hypothesis,' proposed by Hajishengallis and colleagues in 2012, suggests that specific low-abundance microbial pathogens can orchestrate a transition to a dysbiotic state in the oral microbiome, compromising host defenses for nutritional purposes. P. gingivalis is proposed as the keystone pathogen for periodontitis due to its ability to evade the immune system through various mechanisms. However, some researchers argue that viral infections, such as CMV and EBV, may also contribute to periodontitis by inhibiting immune responses. While no single pathogen has been linked definitively to gingivitis, disease severity correlates with plaque quantity and bacterial load, indicating a role for overall microbial abundance and ecological succession in disease initiation. Hajishengallis and Lamont later updated the hypothesis to the 'Polymicrobial Synergy and Dysbiosis' hypothesis, emphasizing the role of multiple pathogens acting in concert to sustain a dysbiotic microbial community and elicit a destructive host response. This highlights the importance of community virulence factors in driving periodontal pathogenicity [26].

This model implicates both gram-negative and gram-positive bacteria in disease pathogenesis, provided they can incite and/or withstand inflammation. Mixed microbial communities foster competitive and cooperative interactions, influencing the overall microbiome composition and function. For instance, Passariello et al. noted a positive correlation between CMV, EBV, HSV-1, and several periodontal pathogens like P. gingivalis, T. forsythia, and F. periodonticum. Our recent research also linked increased viral diversity, including Gammaretrovirus and Porcine type-C oncovirus, to alveolar bone loss and periodontal ligament widening. Similarly, Zhao et al. found that HBV correlated positively with Neisseriaceae family abundance on the tongue dorsum. However, further investigation is needed to ascertain all critical triggers for periodontitis [27].

Commonly identified dysbiotic microbiome signatures in periodontitis include the red complex triad (T. denticola, T. Forsythia, P. gingivalis) and the orange complex triad (F. nucleatum, P. intermedia, P. micra), along with other pathogens like A. actinomycetemcomitans, C. rectus, E. corrodens, and Filifactoralocis. Bacteria associated with periodontitis can enter the bloodstream, potentially causing bacteremia, transient or continuous presence of bacteria in the bloodstream. Lockhart et al. observed positive bacteremia rates post-tooth extraction and toothbrushing, with Streptococcus viridans, A. actinomycetemcomitans, P. gingivalis, Micromonas micros, and Actinomyces species commonly identified. Circulating bacteria could colonize other tissues, linking the oral microbiome to systemic diseases[28,29].

Roles of the Oral Microbiome:

The human body's microbial communities contribute significantly to critical physiological, metabolic, and immunological functions, encompassing food digestion, energy generation, and the development of mucosal and immune systems. They also regulate fat storage, process environmental toxins, maintain skin and mucosal barriers, modulate the immune system, resist pathogen colonization, and prevent disease. The relationship between the microbiota and the host profoundly influences health outcomes, with the oral microbiome, often in the form of a biofilm, playing a central role in maintaining oral health and preventing disease. Understanding the composition of the microbiome and its interactions with neighboring organisms is essential for unraveling its mechanistic contributions.

METHODS OF STUDYING THE ORAL MICROBIOME:

Traditional methods for microbe identification have evolved from culture-dependent approaches focusing on single species to complex in vitro studies of multispecies communities. Recent technological advancements have enabled culture-independent characterization of the entire microbiota in vivo, ranging from analyzing individual gene expression to meta-omic analysis.

The most significant recent advancement lies in the development of culture-independent "omics" techniques, which encompass the study of DNA, RNA, proteins, or metabolites of the entire microbial community. Two emerging fields of research, microbiomics and metagenomics, have emerged to detect and identify microbes in the body and understand microbiome activity in health and disease. Metagenomics involves techniques that detect non-culturable bacteria and assess the genomic diversity of microbes through comprehensive genomic analysis of the entire microbial community. Metagenomics provides insights not only into the types of organisms present but also their functional potential by analyzing metabolic pathway genes and utilizing protein-coding sequence databases. It involves sequencing the entire DNA from a given sample.
The oral microbiome is arguably the most extensively studied human microbiome to date, facilitated by the ease of sample collection.Traditional methods for identifying bacterial taxa relied on culture-dependent techniques, such as microscopy and biochemical tests, which have limitations in culturing many bacterial species present in biological samples. Despite efforts, only about half of the estimated 700 oral bacterial species have been successfully isolated and classified through culture-based methods.

In response to these limitations, culture-independent techniques, including gel-based methods like denaturing gradient gel electrophoresis (DGGE) and PCR-based methods such as real-time quantitative PCR and PCR-DGGE, have emerged for high-throughput analysis of microbial communities. DNA sequencing approaches like 16S rRNA sequencing and metagenomics have also been employed to study uncultivated oral microbial communities. While 16S rRNA sequencing provides insights into taxonomic composition, metagenomics offers a broader view by sequencing the entire genome.

Next-generation sequencing (NGS) technologies, such as 454 pyrosequencing, Illumina, and Pacific Biosciences, have revolutionized microbial diversity studies by enabling rapid large-scale sequencing projects. However, meaningful interpretation of NGS data requires advanced bioinformatics capabilities for data quality control and analysis. These advancements allow researchers to profile microbiomes and metagenomes at unprecedented depths, offering insights into microbial diversity and function. However, careful consideration of study design and analysis steps is crucial for robust and accurate results in microbiome studies [30].

CONCLUSION

Microbiome research is in its early stages, with ongoing studies and continual data accumulation. However, there is inconsistency in the results obtained across various studies, which could be attributed to differences in techniques, standardization methods, and sample sizes. To establish more consistent patterns and generate reliable data, larger studies encompassing diverse sites in both healthy and diseased states are necessary. Such research efforts can help identify distinct biomarkers and facilitate the development of targeted therapies and personalized medicine, ultimately enhancing patient care in clinical practice.

REFERENCES:

1. Deo PN, Deshmukh R. Oral microbiome: unveiling the fundamentals. J Oral MaxillofacPathol2019;23:122–8.
2. Sender R, Fuchs S, Milo R. Revised estimates for the number of human and bacteria cells in the body. PLoSBiol 2016;14:e1002533.
3. Dovrolis N, Kolios G, Spyrou GM, Maroulakou I. Computational profiling of the gut-brain axis: microflora dysbiosis insights to neurological disorders. Brief Bioinf2019;20:825–41.
4. NIH Human Microbiome Project Data Portal.
5. Kreth J, Merritt J, Qi F. Bacterial and host interactions of oral streptococci. DNA Cell Biol 2009; 28: 397–403.
6. Radaic A, Ganther S, Kamarajan P, Yom SS, Grandis JR, Kapila YL (2021) Paradigm Shift in the Pathogenesis and Treatment of Oral Cancer Focused on the Oralome and Antimicrobial-Based Therapeutics . Periodontol 2000.
7. Bandara HMHN, Panduwawala CP, Samaranayake LP. Biodiversity of the human oral mycobiome in health and disease. Oral Dis 2019;25:363–71.
8. Ghannoum MA, Jurevic RJ, Mukherjee PK, Cui F, Sikaroodi M, Naqvi A, et al. Characterization of the oral fungal microbiome (mycobiome) in healthy individuals. PLoSPathog 2010;6:e1000713.

9. Sowmya Y. A review on the human oral microflora. Res Rev. 2016;4:1–5.
10. Marsh PD. Role of the oral microflora in health. Microbial Ecol Health Dis. 2009;12:130–7.
11. Patil S, Rao RS, Amrutha N, Sanketh DS. Oral microbial flora in health. World J Dent. 2013;4:262–6.
12. Palmer RJ., Jr Composition and development of oral bacterial communities. Periodontol 2000. 2014;64:20–39.
13. Petersen C, Round JL. Defining dysbiosis and its influence on host immunity and disease. Cell Microbiol2014;16:1024–33.
14. Sultan AS, Kong EF, Rizk AM, Jabra-Rizk MA. The oral microbiome: a Lesson in coexistence. PLoSPathog 2018;14:e1006719.
15. He X, McLean JS, Guo L, Lux R, Shi W. The social structure of microbial community involved in colonization resistance. ISME J 2014;8:564–74.
16. Baker JL, Edlund A. Exploiting the oral microbiome to prevent tooth decay: has evolution already provided the best tools? Front Microbiol2018;9:3323.
17. Marsh PD. In sickness and in health-what does the oral microbiome mean to us? An ecological perspective. Adv Dent Res 2018;29:60–5.
18. Zaura E, Brandt BW, Prodan A, Teixeira de Mattos MJ, Imangaliyev S, Kool J, et al. On the ecosystemic network of saliva in healthy young adults. ISME J 2017;11:1218–31.
19. Takeshita T, Kageyama S, Furuta M, Tsuboi H, Takeuchi K, Shibata Y, et al. Bacterial diversity in saliva and oral health-related conditions: theHisayama Study. Sci Rep 2016;6:22164.
20. He X-S, Shi W-Y. Oral microbiology: past, present and future. Int J Oral Sci 2009;1:47–58.
21. McLean RJC, Lam JS, Graham LL. Training the biofilm generation–a tribute to J. W. Costerton. J Bacteriol2012;194:6706–11.
22. Marsh PD. Are dental diseases examples of ecological catastrophes?. Microbiology (Reading, Engl) 2003;149:279–94.
23. Takahashi N, Nyvad B. The role of bacteria in the caries process: ecological perspectives. J Dent Res 2011;90:294–303.
24. Zargar N, Ashraf H, Marashi SMA, Sabeti M, Aziz A. Identification of microorganisms in irreversible pulpitis and primary endodontic infections with respect to clinical and radiographic findings. Clin Oral Investig2020;24:2099–108
25. Nomura R, Matayoshi S, Otsugu M, Kitamura T, Teramoto N, Nakano K.Contribution of severe dental caries induced by streptococcus mutans to the pathogenicity of infective endocarditis. Infect Immun 2020;88.
26. Hajishengallis G, Lamont RJ. Beyond the red complex and into more complexity: the polymicrobial synergy and dysbiosis (PSD) model of periodontal disease etiology. Mol Oral Microbiol2012;27:409–19.
27. Nobile CJ, Johnson AD. Candida albicans biofilms and human disease. Annu Rev Microbiol2015;69:71–92.
28. Horliana ACRT, Chambrone L, Foz AM, Artese HPC, de Rabelo M de S, Pannuti CM, Romito GA. Dissemination of periodontal pathogens in the bloodstream after periodontal procedures: a systematic review. PLoS One 2014;9:e98271.
29. Lockhart PB, Brennan MT, Sasser HC, Fox PC, Paster BJ, Bahrani-Mougeot FK. Bacteremia associated with toothbrushing and dental extraction. Circulation 2008;117:3118–25.
30. Deo PN, Deshmukh R. Oral microbiome: Unveiling the fundamentals. J Oral MaxillofacPathol. 2019 Jan-Apr;23(1):122-128. doi: 10.4103/jomfp.JOMFP_304_18. PMID: 31110428; PMCID: PMC6503789.