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Applications of the oral microbiome in personalized dentistry

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Running title: oral microbiome in personalized dentistry

Abstract

Objective: In the era of personalized medicine, it is imperative that oral health is integrated into this concept. The oral cavity fosters a highly individualized microbiome that has evolved to promote oral health, and which exists in a dynamic balance with the host. Microecological changes to the biology of the mouth [e.g. in the host diet and lifestyle, or status of the immune system] may drive deleterious shifts in the composition or metabolic activity of the oral microbiome ['dysbiosis']. This review aims to explore how knowledge of the oral microbiome may be utilized for personalized dentistry at the point-of-care. **Design:** This is a comprehensive narrative review of the literature, summarizing the perspectives of the authors. **Results:** The huge increase in recent knowledge on the ecology and microbiology of the oral cavity generated by 'OMIC' technologies may indeed be clinically translated to support patient care, in terms of prevention, monitoring, risk classification or early diagnosis. The identified clinical applications may not only include dental caries and periodontal disease, but also dental implants and orthodontics. Population-based applications may include systemic health, pregnancy and elderly populations. **Conclusions:** Applications of selected oral microbiome and host-related biochemical parameters [e.g. the saliva proteome] for personalized dentistry can be customized for different clinical applications or individual populations, at point-of-care hubs.

Keywords: Personalized dentistry, oral microbiome, oral ecology, oral disease, chair-side diagnostics, point-of-care

Background

The human microbiome plays an essential and active role in the health and well-being of their host. There is a symbiotic relationship between the microbiome and the host; the host provides a secure, moist and nutritious environment while the microbiota deliver important benefits. These benefits range from the exclusion of potential pathogens from colonising available surfaces, to regulating the cardiovascular system and contributing to the development of the host defences early in life or the immunomodulation throughout life (Blum, 2017; Devine, Marsh, & Meade, 2015; Kilian et al., 2016) (Table 1).

The mouth supports the second largest and most diverse microbial community found in the body. The oral microbiota grow as multi-species biofilms on exposed surfaces, and between 100-300 species can be isolated from a single individual (Kilian et al., 2016). Desquamation ensures that microbial load on mucosal surfaces is relatively light but the mouth is unique in having non-shedding surfaces [teeth, implants, dentures], which permit the accumulation of substantial biofilm levels, unless controlled by effective oral hygiene.

The symbiotic relationship between the microbiota and host exists in a dynamic balance (microbial homeostasis). Any substantial change to the local environment can perturb microbe-microbe interactions, which may then alter the host-microbe equilibrium, thereby increasing the risk of disease. The breakdown of this microbial homeostasis is termed dysbiosis. Dysbiosis in the mouth is associated with caries, periodontal diseases, yeast infections, and increases the risk or severity of several systemic diseases including diabetes, cardiovascular disease and rheumatoid arthritis. Initially, in the twentieth century, the oral microbiota was characterised using culture-based methods, and there was an aspiration that an association between specific pathogens and disease might be

found, as had been the case with many classical infections. Over time, however, and with the advent of culture independent approaches, it has become apparent that disease is associated more with a shift in the oral microbiota away from species that predominate in health to a greater abundance of communities and consortia comprising taxa that were previously minor components of the microbiome. In this way, oral diseases differ from many classical infections, which are due to colonisation by species (and often by single species) not normally found at that site, often involving the production of specific virulence factors, and where the 'pathogen' can be diagnostic of that disease. Prevention or treatment can involve the use of antibiotics or vaccines.

The mouth is the most accessible habitat in which to study the relationship between the host and the microbiome. Individual surfaces and sites can be sampled both for the microorganisms but also for the activity of the host defences, using saliva or gingival crevicular fluid. Following the application of various 'OMIC' approaches, there is now a mass of information on the composition of the oral microbiome [approx. 770 taxa (Takahashi, 2015)] and the saliva proteome [1000+ proteins (Bostanci & Bao, 2017)], and functional profile of the innate and adaptive immune response. Despite this extensive knowledge and the development of relevant databases of microbial and biochemical information, however, it has not yet proved possible to translate this mass of genomic, transcriptomic, proteomic and metabolomic information of oral communities in health and disease into new diagnostic or treatment strategies. The aim of this review was to address how our current knowledge of the oral microbiome and general oral ecology could be applied to the concept of personalized dentistry, providing suggestions for its practical applications in the clinic.

Principles in pursuit of personalized dentistry

The vast oral microbiome and proteome discoveries achieved via high-throughput datasets now need to be clinically translated into dental public health benefit, for prevention, early diagnosis and predictive treatment (Belibasakis & Mylonakis, 2015). This will require further longitudinal studies of changes in the oral microbiome and proteome during the transition from oral health to disease, in order to be able to reliably link biomarkers to the early stages of dysbiosis. In pursuit of this, it is important to perceive oral disease as an ecological change of a human ecosystem that has led to the dysbiosis of highly individualized beneficial resident microbial communities. The new paradigm for understanding and evaluating oral disease requires shifting away from concepts applied in medical microbiology, such as a quest for specific pathogens and their elimination, or using broad-spectrum agents and standard laboratory criteria to define the efficacy of antimicrobials. This necessitates monitoring of microbiomic and metabolomic changes, rather than seeking individual units as diagnostic or prognostic factors for oral disease. Species with different taxonomic classifications may perform the same role or function within a microbial consortium (Takahashi, 2015). Indeed, according to the human microbiome project, microbial taxa may well vary but their metabolic pathways in a given niche remain relatively similar (Human Microbiome Project, 2012). The oral cavity is the ideal site to study ecological relationships between the microbiome and the host, due to its vast taxonomic diversity and the easily accessible and non-invasive sampling (e.g. dental plaque or saliva) of precise sites in the mouth.

Application of the knowledge of the human microbiome should aim at preserving the highly intra- and inter-individual diversity of the microbiota at a site, while protecting against its loss. Monitoring large arrays of these parameters can help develop

bioinformatic algorithms for defining whether the observed trends are symbiotic or dysbiotic. Measured parameters of the heterogeneity and functional status of a sampled ecosystem should include a) the diversity of its taxonomic components (i.e. microbes), b) their products (i.e. microbial metabolites, i.e. the metabolome), c) their biological responders (e.g. host-related biochemical parameters), and d) acquired environmental parameters (e.g. diet, status of the host defences). In turn, process will eventually help stratify individuals into groups with similar properties of their oral ecosystem, so called 'ecotypes' (Zaura et al., 2017) which could help explain the carriage and persistence of putative 'pathogens' at specific sites, the degree of 'resilience' to change of the microbiome of an individual, and the risk for developing certain oral diseases, while also supporting the diagnosis or predicting the outcome of treatment. As 'one size' does not fit all, there is a need for a tailored oral care according to their ecological type, so the future challenge will be the identification of the right preventive strategy for each individual. To achieve that, longitudinal studies will be needed, to identify biomarkers that predict disease before tissue destruction occurs, per individual. For meaningful and routine screening of changes in the microbiome, proteome or metabolome, the analytical process should be performed by chair-side technological utilities at a point-of-care (POC) hub. POC hubs could be considered as the general practice, dental practice, nursing home, pre-natal clinics, or private home.

Potential applications for oral diseases

Screening for symbiotic or dysbiotic trends can be performed on an overall or a site-specific manner in an individual. Overall implies the use of saliva, whereas site-specific manner implies the sampling of dental biofilms on the tooth surface/periodontal pocket, and/or the analysis of gingival crevicular fluid (GCF) from the targeted gingival

sulcus/periodontal pocket. Confirmation of 'health' can be actively managed by motivating and encouraging the patient to preserve on-going behavioural and dietary habits commensurate with oral health. On the other hand, dysbiotic trends can be identified early, and if not adequately corrected with enforced oral hygiene and behavioural changes, the patient can be referred to secondary healthcare for remedial action. A major challenge in this quest to identify biomarkers will be the ability to differentiate normal and transitory shifts in the oral microbiome/proteome from the more genuine and persistent changes that could reliably herald dysbiosis.

Saliva in particular can act as a diagnostic fluid for overall microbiome, proteome, or genome sequencing for personalized monitoring. Downstream bioinformatics analyses may lead to evidence-based diagnostic or treatment options that include dietary changes and education, enrichment of 'risk-assessment' tools for deciding on the frequency of visits to dental practices (Featherstone & Chaffee, 2018), use of probiotic bacteria or prebiotics, or targeted antibiotics or other small molecules. An example of such an approach is considering patient 'ecotypes' and whether belonging to a particular ecotype increases the susceptibility to dental caries or periodontal disease (Zaura et al., 2017). For instance, high ecological complexity of the microbial community, with predominantly proteolytic anaerobic taxa, high resting pH and low lysozyme activity of saliva was proposed to indicate early dysbiosis towards periodontal disease, while reduced microbial diversity with predominantly saccharolytic taxa and low resting pH together with high proportion of lipid degradation products among salivary metabolites was indicative of a person with dysbiosis towards caries (Figure 1). To identify these subgroups or ecological types of individuals, one needs to consider and monitor easily collectable and analyzable biochemical parameters in saliva, or at the site of interest (dental surface, gingival crevice/pocket). In the case of caries, these may include, but

not restricted to, pH raising factors in the oral cavity, salivary buffering capacity, production of high pKa acids (such as acetic acid), generation of alkali by bacterial metabolism of urea or arginine that may counteract the low pH, etc. In contrast, in the case of periodontal disease, these may include increased pH in the gingival crevice/pocket as a result of inflammation, high alkali production, high proteolytic activity conferred by either bacterial metabolic or host inflammatory products, reduction in redox potential or oxygen partial pressure, indicative of anaerobiosis, etc. Screening of saliva and GCF of periodontitis patients and healthy controls using proteolytic activity tests showed the potential of specific substrates to determine the presence of *Porphyromonas gingivalis* proteases, underlying the potential of specialized chemical substrates for the detection of bacterial proteases, envisaging improvements to the specificity of the current enzyme-based diagnosis of periodontitis (Kaman et al., 2012). Screening for the development of red plaque fluorescence could also have important diagnostic implications in identifying patients susceptible to gingival inflammation. This property of dental plaque correlated with gingival inflammation in experimental gingivitis, and measuring it allowed stratification of the study subjects into those who responded with gingival inflammation to plaque accumulation and those who did not before the signs of inflammation were clinically detectable (van der Veen, Volgenant, Keijser, Ten Cate, & Crielaard, 2016). Monitoring ecological changes in the oral cavity can be useful not only in defining diagnostically the deterioration of health status or the risk for disease progression, but can also aid in evaluating the outcomes to different interventions in dental practice. As mentioned earlier, this will require further longitudinal studies to trace the dysbiotic changes in the oral microbiome and proteome, when transiting from health to disease, and *vice versa*. For instance, monitoring the effects of probiotics is also a matter of

consideration in personalized dentistry, due to lingering debates on whether they can be applied for the prevention or treatment of oral disease. The majority of gut probiotics can drop the pH in favor of aciduric and acidophilic bacteria, which could in-turn pose a risk for dental caries. Yet, the environmental changes accompanied by a reduced pH may suppress the alkalophilic and proteolytic taxa (mainly periodontal pathogens), causing a shift to health-associated taxa, at least in terms of periodontal health. This has been the case when studying the periodontal pathogen *Porphyromonas gingivalis* and cariogenic pathogen *Streptococcus mutans* in laboratory biofilm models (Jasberg, Soderling, Endo, Beighton, & Haukioja, 2016). The use of probiotics has also yielded clinically promising results in improving gingival health and periodontal status (Teughels et al., 2013), although less is certain in terms enamel health and dental caries. It should also be taken under consideration that probiotics applied for gut applications may not necessarily be probiotic for oral health applications (Pham et al., 2009). Probiotic strains derived from the dairy industry and from gastro-intestinal applications may not be suitable for oral care applications for a variety of reasons, including their inability to colonize the mouth and compete with the resident oral microbiome for nutrients. However, recently, a number of oral streptococcal strains, such as *Streptococcus dentisani* (Lopez-Lopez, Camelo-Castillo, Ferrer, Simon-Soro, & Mira, 2017) and *Streptococcus* A12 (Huang et al., 2016) have been isolated from individuals with low caries experience; these strains can generate alkali from arginine and produce inhibitors against the cariogenic mutans streptococci, while being able to naturally colonize the mouth. *Streptococcus oralis* can also outcompete *S. mutans* when cultured together in multi-species biofilms (Thurnheer & Belibasakis, 2018). Hence, changes in the diversity and composition of the oral microbiome exposed to probiotics may help identify individuals, or populations, responsive to this application, or ones for which it

may have negative or no effects in terms of oral health.

Potential applications for patient populations

Personalized dentistry implies the screening not only of individuals, but also applications on highly targeted populations. This section discusses some of the potential applications where monitoring of changes in the oral cavity could well be applicable for prevention or diagnosis.

Applications in relation to systemic health

There have been many studies over the past decade on the association between oral disease and conditions that compromise systemic health. It is still debatable whether the involved mechanistic links account for microbiological or immunological triggers, and certainly no definitive causal inference has been established. Yet, monitoring of the respective microbiological or immunological changes in the oral cavity may be an indirect way to follow changes in systemic health parameters for evaluating the risk of development or the deterioration of existing conditions.

Apart from a focused impairment on the functionality of the oral cavity, periodontal disease may generate a chronic inflammatory burden to the human body, with potential systemic effects (Nibali et al., 2007; Tonetti et al., 2007). In patients with periodontitis, the periodontal inflamed surface area is estimated to reach approximately 44 cm^2 (Hujoel, White, Garcia, & Listgarten, 2001; Nesse et al., 2008). As the gingival wound size in patients with periodontitis corresponds to the palm surface area of an adult hand, this creates “a wide window” for oral bacteria/their products or locally produced inflammatory mediators to escape into systemic circulation. Indeed, significant links have been elucidated between periodontitis and diabetes mellitus (Lalla & Papapanou,

2011) and cardiovascular diseases (Beck & Offenbacher, 2005). Conversely, successful treatment of periodontitis by reducing bacterial load and local inflammation can have a beneficial effect on systemic inflammatory status (Lalla & Papapanou, 2011). As a reciprocal association between the oral cavity and other systems of the human body exists, there is merit to use "oral health risk indicators/factors" as predictors of general health. Most of the available association studies are based on clinical indicators of periodontitis (e.g. probing pocket depth, clinical attachment level, bleeding on probing scores), while taking less consideration of molecular parameters of the disease, such as microbial counts in subgingival plaque, or host response proteins in saliva or GCF. It is highly unlikely that there is a single individual marker decisively responsible for the cross-talk between oral disease and systemic health, but combinations of several factors may rather predict the associated risk. There is a massive volume of data available derived from the analysis of the subgingival microbiota, GCF and saliva that is obtained through conventional or high-throughput platforms. Oral bacteria associated with periodontal disease are among the primary candidate risk markers, due to the high levels of bacteremia that are observed after routine oral prophylaxis procedures, such as tooth brushing and flossing. Although not fully conclusive, some differences in the composition of the subgingival plaque microbiome have been reported in subjects with systemic diseases (Demmer et al., 2015; Fak, Tremaroli, Bergstrom, & Backhed, 2015). For example, higher colonization levels of "red complex" species (*P. gingivalis, Treponema denticola, Tannerella forsythia*) have been shown to be associated with a higher prevalence of pre-diabetes among diabetes-free adults (Demmer et al., 2015). The periodontal bacterial burden has also been linked with an increased risk of cardiovascular disease (Rosenfeld & Campbell, 2011) and *P. gingivalis* is among the top infectious agents that can be recovered within carotid atherosclerotic plaques

(Desvarieux et al., 2005). Although there is sparse knowledge about the salivary proteome in patients suffering from systemic diseases who also suffer from periodontitis, certain salivary proteins may be associated with higher risk of systemic inflammation. Hence, identifying markers with higher specificity and sensitivity is of importance especially where comorbidities exist. For example, a newly emerged biomarker, a soluble form of triggering receptor expressed on myeloid cells-1 (TREM-1), is associated with increased systemic inflammatory burden in patients with periodontal disease (Bostanci, Ozturk, Emingil, & Belibasakis, 2013; Nylund et al., 2017) and deregulated local immune responses in the ageing population (Ozturk, Belibasakis, Emingil, & Bostanci, 2016). Another candidate biomarker that may confer high specificity and sensitivity for the connection between oral disease and systemic conditions is matrix metalloproteinase (MMP)-8 (Rathnayake et al., 2013; Rathnayake, Buhlin, et al., 2017; Rathnayake, Gieselmann, Heikkinen, Tervahartiala, & Sorsa, 2017). There is a clear need for more accurate molecular measures that identify the systemic burden of oral infections and their impact on general health. Improved patient management should result from the incorporation of these measures into routine dental and medical practices. Nevertheless, the main contemporary challenge is how to combine and make best use of the generated data sets for the benefit of the patient.

Applications in pregnancy

Oral health is compromised during pregnancy as a result of the establishment of hormonally-primed gingival inflammation. Poor oral health has been associated with adverse pregnancy outcomes, such as preterm birth, pre-eclampsia, and foetal growth restriction (Riche et al., 2002). While periodontal therapy leads to improved periodontal conditions in pregnant women, it does not necessarily reduce overall rates of pre-term

birth and low birth weight (Iheozor-Ejiofor, Middleton, Esposito, & Glenny, 2017). The biological mechanisms behind this association are not clear, but may involve oral microorganisms, or their components, reaching the foetal-placental unit, or circulating inflammatory mediators impacting this unit. Apparent microbiological changes in the oral cavity during pregnancy include increased levels of *Prevotella intermedia* and *Prevotella melaninogenica* attributed to progesterone or oestradiol (Kornman & Loesche, 1980). *Fusobacterium nucleatum* is also associated with preterm birth and has been isolated from the amniotic fluid of women delivering prematurely (Han et al., 2004). Pregnancy-specific inflammatory changes include an increase in salivary Annexin-1 or host-derived proteases (Gursoy et al., 2010; Hassan et al., 2018). Measurement of the levels of suspected pathogens, the associated hormones, Annexin-1 or matrixmetalloproteinases can be easily and routinely measured in saliva during a visit at point-of-care (POC), which could be the dental office or the pre-natal clinics. Collective changes in these 'ecological' parameters associated with a deteriorating oral health during pregnancy, could help cluster sub-populations at risk. Guidelines for pre-pregnancy prophylaxis and screening during pregnancy should then be laid out for reducing potential associated risks. These may necessitate further oral hygiene instruction, more regular visits to the dental hygienist for maintenance care, or visit to the obstetrician for evaluation of the pregnancy and foetal status.

Applications in the elderly populations

Ageing can lead to increased carriage and prevalence of opportunistic microbial species in the oropharyngeal region, such as enterobacteria, staphylococci and *Candida albicans* (Belibasakis, 2018). Dental biofilms of elderly individuals harbour 'superinfecting' microorganisms, including enteric rods, yeasts and pseudomonads,

which could predispose to postulated 'geriatric' forms of periodontal disease and increase the risk of inhalation pneumonia (Slots, Feik, & Rams, 1990). More striking age-related differences in the gut and oral microbiota may be found at the extremity of life, such as in the centenarian populations (Biagi et al., 2010; Rampelli et al., 2013). These atypical oral colonizers may favor the development of opportunistic infection, such as oral candidiasis, respiratory infections, or bacteremia in individuals with poor systemic health, or indicate a deteriorating immune response. Therefore, measuring the oral carriage and levels of such microorganisms in frail elderly individuals may help identify subjects at higher risk, so that individual prevention and handling protocols could be designed. This application could be particularly useful at nursing home POC hubs, where these measurements can be auxiliary to the already established handling operational guidelines, but can also help prevent the epidemic spread of communicable infections within the facility.

Applications in dental implant patients

Dental implants are a well-established treatment option in the toolbox of reconstructive dentistry. Yet, dental implant treatment comes with a risk for the development of peri-implantitis, a disease that progressively destroys the implant-surrounding tissues, with eventual implant loss. Active periodontitis is a risk factor for peri-implantitis, and incomplete treatment of the former may increase the occurrence of the latter. A number of microbiological similarities exist between the two diseases, with some distinctive differences. These include a higher frequency of detection of aerobic Gram-negative bacilli (such as enterobacteria), staphylococci and *Candida* spp. (such as *C. albicans*) in peri-implantitis, though it is likely that differences are larger based on studies using metagenomics (Belibasakis, Charalampakis, Bostanci, & Stadlinger, 2015; Robitaille,

Reed, Walters, & Kumar, 2016). Screening for these microorganisms that unilaterally seem to associate with implant, rather than tooth, at the dental POC could be a valuable asset during the treatment planning phase, or during the monitoring phase, following implant installation. Their detection or increased levels would necessitate pre-treatment or maintenance protocols aiming at their elimination, hence amending an oral ecology conductive for peri-implantitis.

Applications in orthodontics

Orthodontic treatment is a common modality in adolescent populations that serves the restoration of functional and aesthetic needs of the patients. Microbial biofilms are able to form on orthodontic appliances in the oral cavity, similarly to natural teeth. This may predispose the patients to two oral health related issues, namely dental caries, and opportunistic fungal infections. During the course of orthodontic treatment, increased colonization by acidophilic and acidogenic mutans streptococci and lactobacilli is observed, both on the orthodontic appliances and the oral cavity in general (Jung, Kim, Park, Cho, & Ahn, 2014). Though the percentage of *S. mutans* and *Lactobacillus* spp. may not be predictive of 'white spot' lesions (Beerens, Ten Cate, & van der Veen, 2017), it is still ecologically plausible that their colonization and biofilm activity may increase the demineralization rate of the enamel, increasing the risk for dental caries. On the other hand, *Candida albicans* also naturally colonizes orthodontic appliances. Orthodontic patients free of oral *Candida* may transit to *Candida* carriers after orthodontic appliance insertion (Hibino, Wong, Hagg, & Samaranayake, 2009), yet systemically healthy individuals do not develop *Candida* infection due to this. However, caution should be placed in orthodontic treatment of immuno-compromised children, as they may run a higher risk of opportunistic fungal infection. Therefore,

screening for cariogenic mutans streptococci, lactobacilli, potentially in combination with measuring acidogenicity in dental plaque or saliva, or *Candida albicans* before or during the course of orthodontic treatment may confer benefits to the susceptible orthodontic patient populations. Their presence, or the detection of increasing levels, can serve as an indicator for antimicrobial or antifungal interventions or poor compliance level with dietary recommendations, aiming to prevent the development of white spot lesions, or opportunistic *Candida* infection, respectively.

Conceptualizing the practice of personalized dentistry at point-of care

The key concept to introducing biological/biochemical parameters in personalized dentistry would be the ability to measure and monitor over time those at the POC, in a rapid manner during the patient visit, complementing the clinical measurements. POC hubs could be considered as the general practice, dental practice, nursing home, pre-natal clinics, or private home. Dispatching the collected material to a centralized laboratory is time-consuming and not cost-effective. The possibility to screen the microbiological or immunological oral status of the patient in a cost-effective and routine fashion will require non-invasive sample collection, such as saliva, and the use of customized devices coupled to adapted micro-laboratory assays. Open-ended nucleic acid-sequencing or proteomic platforms would yield maximal results for customized bioinformatic analysis. However, this is still a distant possibility to be applied in POC hubs in terms of current technology, including lack of bioinformatic tools that can provide a straightforward and clinically meaningful interpretation. What is achievable, however, is the selection of a finite set of microbiological or immunological parameters that are well established and accepted in the literature, some of which were mentioned in the sections above. Suitable platforms for such analyses at POC include scaled-down

molecular laboratory assays, integrated onto a cartridge rather than on a purpose-built device. These microfluidic technologies have enabled the construction of miniature laboratories on a disc conformation, which can rapidly measure in saliva the levels of selected oral bacterial species by a real-time polymerase-chain reaction based assay, and selected immune-markers on an enzyme-linked immune-sorbent assay (Mitsakakis et al., 2016).

Last but not least, routine incorporation of microbiological and immunological parameters in daily clinical practice will enrich the awareness of the clinicians on the underlying biological mechanisms and ecological considerations of the disease. This would lead to a paradigm shift in oral care by guiding clinicians to treat the cause rather than just the consequence of disease. In the course of such developments, they will validate the usability of the gathered biological information for the benefit of the patient, and will propose modifications or further applications for development that are practically relevant for their practice and the benefit of the patient. This will enable the true achievement of personalized dentistry.

Conflict of interest

The author Georgios Belibasakis is an Associate Editor for the journal.

Author contributions

All authors contributed both conceptually and in terms of writing individual sections, to the manuscript. In addition, GB co-ordinated submission, and edited the final version.

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Figure legend

Figure 1. Schematic distribution of clinically healthy individuals based on the state of their oral ecosystem, as defined by salivary analysis. The individuals could be stratified as being in a state of symbiosis – indicated by purple, red and blue portions, corresponding to three adaptive 'ecotypes' described by Zaura et al (Zaura et al., 2017), and in a state of early dysbiosis – indicated by green and cyan-blue coloured portions, corresponding to the outliers of the distribution plot. These outliers have highly specialized microbial communities within an ecosystem with specific physiological, metabolic and microbial properties. Examples of a few of these properties are indicated in the horizontal triangles. Each property is highest at the base of the triangle, and lowest at the tip of it.

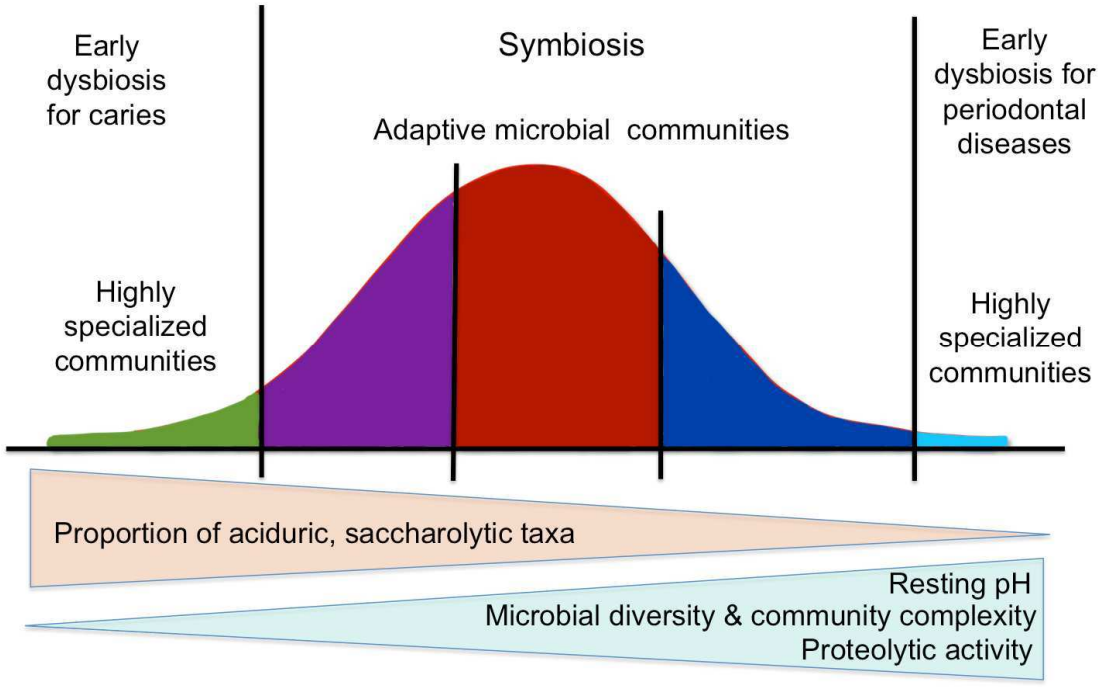

Table 1. Benefits and dis-benefits of the human microbiome

Benefits of the human microbiome [symbiosis]	**Dis-benefits of the human microbiome [dysbiosis]**
Colonization resistance [pathogen exclusion]	Auto-immune & inflammation-mediated diseases
Development of the host defences [priming a healthy immune system early in life]	Obesity
Regulation of the cardio-vascular system	Malnutrition
Gut morphology and motility	Allergies
Vitamin production	Neurological disorders [e.g. anxiety, autism, etc]
Catabolism of the diet & energy production	Cancer
Maintaining a balance with the host throughout life [preventing a detrimental immune response throughout life]	Pregnancy complications [e.g. premature birth, low birth weight babies]

Further details can be found in references: Devine et al., 2015; Kilian et al., 2016; Blum et al., 2017.