

Microbial life in the mouth

The variety of surfaces in the oral cavity offers a home to a whole range of microbial communities, as **Dave Spratt** describes.



◀ A healthy human mouth. *BananaStock / JupiterImages*

The oral cavity forms the top section of the gastrointestinal tract and provides a large number of diverse surfaces on which a wide variety of complex biofilms is able to form. These surfaces include soft shedding tissues of the buccal mucosa, papillae and crypts of the tongue and hard non-shedding surfaces of the teeth. Dental plaque is the term commonly used for the biofilm formed on teeth; however, the term plaque has now been extended to encompass biofilms on all the oral surfaces. These biofilms consist of a complex microbial community embedded in a matrix of polymers of bacterial and salivary origin.

More than 700 bacterial taxa have been recorded in the oral cavity (although only 100–200 occur in any particular mouth) and this rich and diverse flora contains bacterial species

principally found only in this habitat. The soft tissues in the oral cavity lack significant plaque accumulation due to the rapid rates of epithelial cell turnover, the exception being the dorsum of the tongue which is associated with a significant and characteristic microbiota. The hard non-shedding surfaces of the oral cavity, i.e. teeth; provide a far more stable substratum for the colonization of bacteria.

Acquiring the oral microbiota

Colonization of the oral cavity begins at birth. Neonates are usually sterile, despite encounters with the maternal resident microbiota during birth. The acquisition of the oral microbiota is via passive transmission from a variety of sources including food, milk, water and particularly saliva from the mother. However, the majority of these bacteria are present only transiently and only a limited range actually colonize. Streptococci predominate the primary colonization, especially *Streptococcus salivarius* and *S. mitis*. The richness and diversity of the microbiota increase rapidly with age and by the time the infant has teeth (6–18 months) numerous species are present. The microbiota becomes more complex during puberty (12–16 years) and notable increases are observed in Gram-negative anaerobes and spirochaetes. The

presence of sex hormones in the gingival crevicular fluid (serum-like exudate that bathes the tooth/gum interface) is thought to drive this. Few changes have been observed with further increases in age, except to note that loss of teeth and therefore habitat will influence the microbiota.

Dental plaque

In a healthy mouth the only non-shedding surface available for colonization is enamel (a hard, highly calcified tissue) of the tooth surface. This surface is covered with a conditioning film or pellicle, derived from the saliva, within seconds of cleaning and it is this surface which is rapidly colonized by the bacteria in saliva (up to 10^8 per ml). The colonization can be split into two broad processes, the initial attachment of bacteria to the pellicle and the secondary attachment of other cells to those already present. The process is far from random and cell-to-cell recognition of genetically distinct partner cell types plays an important role in development of micro-colonies and subsequent biofilm architecture. Co-aggregation interactions are thought to contribute or perhaps drive plaque development. Early plaque accumulation is facilitated by intrageneric co-aggregation among *Streptococcus* and *Actinomyces* species, as well as intergeneric

co-aggregation between these species. The partnerships between dental plaque bacteria are highly specific and lead to complex biofilms. These processes are likely to benefit the individuals involved and may have nutritional or protective roles. The plaque is therefore rich and diverse and contains numerous microenvironments with a variety of gradients present, including nutrients, oxygen, redox potential and pH. The plaque is not only heterogeneous in nature but differs depending on location on the tooth surface and indeed with time. Mature plaque is therefore an extremely complex and highly dynamic community.

Unknown quantities

It has been estimated that only about 50% of the oral microbiota can be cultivated. While this is a high percentage compared to some other microbial systems, it still means that we have very little knowledge about half of the oral microbiota. Culture independent techniques have clearly made inroads in determining the richness of this proportion (mainly via comparative 16S rRNA gene sequencing) of the microbiota. However, it has only been very recently that techniques have allowed the metagenome and functions of the whole community to be studied.

Oral infections

Small alterations in an environment can lead to ecological shifts and subsequent population changes, and in certain specific cases this may predispose to a more 'pathogenic' microbial community. This concept is termed the 'ecological plaque hypothesis' and can be used to understand the microbial aspects of a range of oral infections.

Dental caries and periodontal disease are some of the most prevalent infectious diseases of humans and are due to the accumulation of dental plaque on the tooth surface and at the tooth gum interface respectively.

Dental caries. This is the localized demineralization of the tooth tissue by various acids produced by bacterial fermentation of dietary carbohydrates and is arguably the most common, chronic infectious disease in humans. Approximately 90% of all dentate adults in the UK have at least one restored tooth as a result of caries with a mean frequency of seven per person. Caries can be simply and conveniently split into two categories: coronal (crown) caries and root-surface caries. Coronal caries can occur on all surfaces of the crown where the plaque biofilm is allowed to develop and mature. Demineralization occurs due to a shift in the microbiota brought about by an increase in the amount and frequency of dietary fermentable carbohydrates. The increase in acids, such as lactic acid, reduce the pH to a level which only favours the growth of acid-loving microbes such as *Streptococcus mutans*, which is additionally highly acidogenic. Root surface caries, as the name implies, occurs on root cementum or dentine and is secondary to

recession of the gums. This is clinically and microbiologically distinct from crown caries. The disease has been shown to have a definite progression, each with its own characteristic microbiology. In brief, initially the lesion is described as 'soft' and consists of a highly demineralized tissue replete with bacteria (increased numbers of lactobacilli and Gram-positive pleomorphic rods, *Actinomyces israelii* and *A. gerencseriae*, but fewer streptococci). The progression of the lesion leads to a change in appearance and is categorized as 'leathery', consisting of a re-mineralized surface overlaying a heterogeneous mix of bacteria, de-mineralized tissue and re-mineralized tissue. A further progression is to a 'hard' lesion which is fully re-mineralized and inactive with respect to caries (reduced numbers of *S. mutans*).

Root canal infections. Structures present in the mouth not normally exposed to the microbiota are usually sterile, for example, the endodontium – the pulp and root canal system within teeth. The root canals of teeth are complex systems of interconnecting channels containing the blood vessels and nerve tissue leading from the tooth apex to the pulp chamber. Endodontic infections are therefore defined as infections of the pulp and periapical tissues. Bacteria and bacterial products can gain access to the pulp chamber, often as a consequence of caries (demineralization of enamel and dentine). The resulting inflammation will lead to pulpal necrosis and progress to resorption of bone supporting the tooth, finally leading to tooth loss. The bacteria associated with individual lesions are surprisingly limited given the number of taxa potentially able to colonize and the large number of taxa associated with periodontal lesions. This reduced diversity implies special selective pressures operating within the root-canal system. Root-canal infections are invariably polymicrobial in nature and typically 4–12 bacterial isolates can be cultured. This microbiota is often diverse with respect to growth atmosphere, nutritional needs and virulence determinants and may be regarded as an 'infection team'. For example, primary colonization and adherence to dentine is carried out by streptococci which additionally utilize oxygen, thus making the environment more anaerobic and therefore suitable for the colonization and growth of strict anaerobes. Commonly, isolates from infected root canals include streptococci, *Actinomyces* spp., *Prevotella* spp., *Peptostreptococcus* spp. and *Fusobacterium nucleatum*.

Periodontal diseases. This broad group of diseases affects the periodontal tissues (gums and supporting bone). The most common of the periodontal diseases is gingivitis; this is usually brought about by poor oral hygiene. The microbiota and their extracellular products present at the gum margin cause a reversible (with good oral hygiene) non-specific inflammation of the gums. The plaque microbiota shifts from a streptococci-dominated community to one dominated by *Actinomyces* species.

Periodontitis refers to a group of more advanced and related diseases defined as 'an apical extension of gingival inflammation to involve the tissues supporting the tooth (periodontal ligament and bone)' and results in a periodontal pocket. By far the most common is chronic periodontitis which is the major cause of tooth loss in adults. The microbiota present in the periodontal pocket is extremely diverse, with up to 100 culturable species from a single pocket. Since such a rich microbiota is present, trying to identify the particular species responsible for disease initiation and progression is a very complex and difficult undertaking (especially as probably only half of the taxa are culturable). However, the World Workshop on Clinical Periodontology has designated three species as aetiological agents of periodontitis in a susceptible host: *Aggregatibacter* (formerly *Actinobacillus*) *actinomycetemcomitans*, *Porphyromonas gingivalis* and *Tannerella forsythia* (formerly *Bacteroides forsythus*). Numerous other types of periodontitis exist and the microbiology of each is thought to be different.

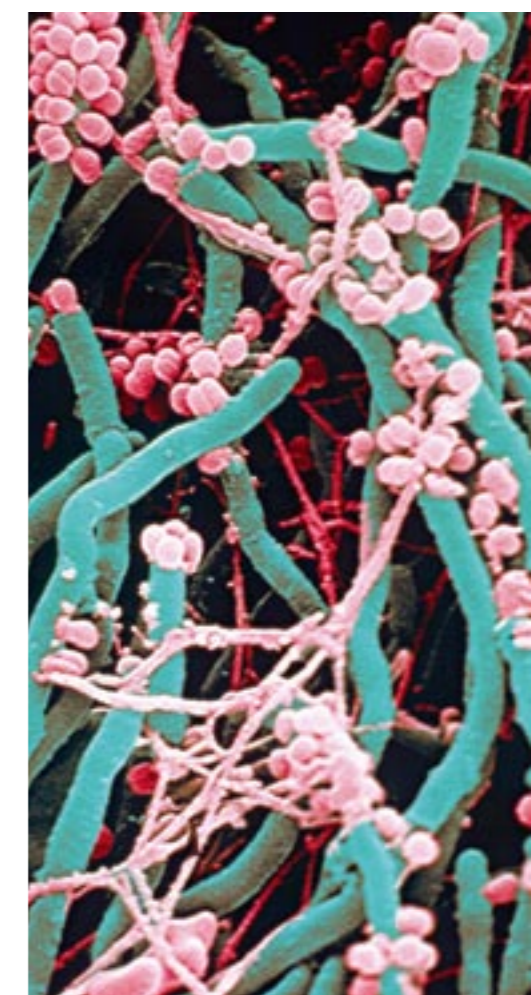
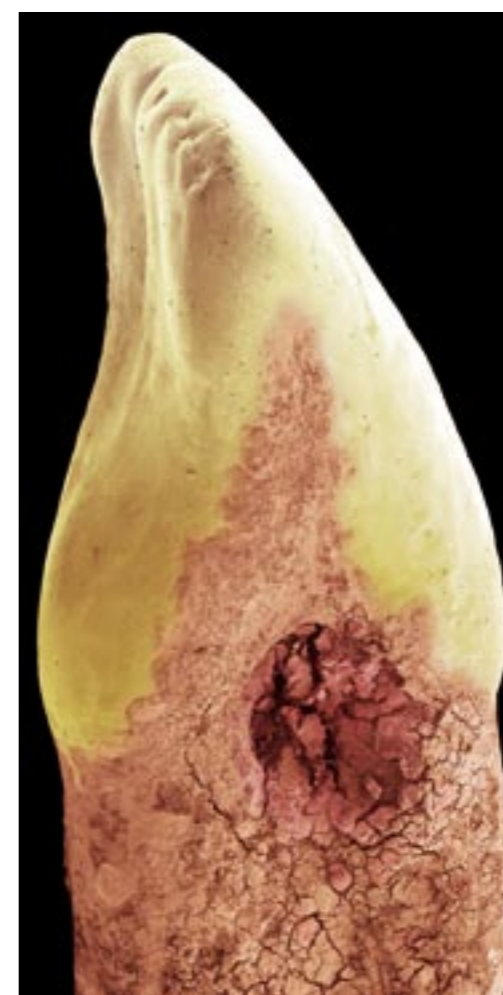
The oral microbiota is also responsible for a number of other oral problems including oral malodour, thrush, angular cheilitis, denture-associated erythematous candidosis, *Candida* leukoplakia and median rhomboid glossitis.

In summary

Life on us and especially our oral microbiota is complex, dynamic, rich and diverse. Changes in the community structure brought about by environmental alterations cause a range of diseases, the ecology and pathology of which remain, largely, unknown.

Dave Spratt

Senior Lecturer, Microbiology Unit, Division of Microbial Diseases, UCL Eastman Dental Institute, 256 Gray's Inn Road, London WC1X 8LD, UK (t 0207 915 1107; f 0207 915 1127; e d.spratt@eastman.ucl.ac.uk)



▲ Top. Mouth of a person reflecting a history of poor oral care: the gums are inflamed (gingivitis) and the lower incisor teeth are surrounded by deposits of tartar. Science Photo Library

▲ Lower left. False-coloured scanning electron micrograph of a cavity (lower centre) in a human incisor. Cavities are caused by dental plaque (brown), a film of bacteria embedded in a glycoprotein matrix. Steve Gschmeissner / Science Photo Library

▲ Lower right. False-coloured scanning electron micrograph of *Streptococcus mutans* bacteria (pink) in dental plaque. Manfred Kage, Peter Arnold Inc. / Science Photo Library