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Review

Rationale for treatment of oral infectious diseases based on a micro-ecological concept

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ABSTRACT

The most common oral infectious diseases are caries, endodontic infections, periodontitis and peri-implantitis. Early diagnosis and total control of these conditions remain a major challenge for clinicians. In this review, we describe infectious oral diseases and evaluate critically the current treatment strategies using a holistic aetiological approach. We draw attention to the local environment, the micro-ecology, where opportunistic pathogens may survive and thrive. We explain why elimination of bacteria from the disease site is probably not feasible and may not be critical; acceptance of this idea would represent a paradigm shift in understanding these conditions. We demonstrate that a crucial step for long-term success of treatment interventions and shift from disease to health involves a change in the local environment to create conditions in which pathogenic bacteria cannot survive and grow. We argue that measures that do not entail local micro-ecological change at affected sites will fail to prevent the recurrence of infectious oral diseases. Our further hope is that the idea of micro-ecology in dentistry will provide a model and pedagogical tool that will help clinicians, in their quest to counter oral diseases of infectious origin, to evaluate treatment approaches in dental care.

KEYWORDS: antibiotics, antiseptics, bacteria, caries, dental implant, ecology, endodontic treatment, infection, periodontitis, probiotics

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INTRODUCTION

Oral infectious diseases are bacterially induced pathological conditions that occur in the oral cavity. Examples of oral infectious diseases are dental caries, endodontic infection, necrotic ulcerative gingivitis, periodontitis, peri-implantitis and pericoronitis. They are all plaque-mediated diseases, underlining the many different pathways bacteria can follow when growing in different ecological environments, such as on teeth, at or below the gingival or mucosal margin. Obviously, caries, endodontic infection, periodontitis and peri-implantitis are the most devastating ones because they may lead to tooth or implant loss. They progress by way of sudden bursts of activity, somewhat resembling earthquakes. We can ascertain where they hit but not when; thus oral diseases are largely unpredictable. Clinical signs and symptoms of the disease follow the 'burst' and guide us to the pathological site. Their intensity, nevertheless, depends on the nature of the infectious process (acute or chronic).

Dealing with infectious oral diseases remains a challenge, both from a diagnostic and a treatment perspective. Ideally, we would like to have identifiable valid prognostic markers, to mark the exact timing of the pathological event. In this respect, researchers should pay attention to the bacterial activity that takes place in the specific environment and tips the balance from health to disease. Early diagnosis is important for successful treatment. Control is a much more realistic treatment endpoint compared with cure because the risk of recurrence of these diseases cannot be

eliminated. Patients should also take this message home; it implies regular check-up appointments and maintenance visits to ensure the long-term success of the treatment.

Several aspects of the pathogenesis of these chronic diseases remain obscure, and we are unable to propose treatment options and to choose the most appropriate and reliable treatment regime. Given the wide and promising options available at the hands of the clinician, it is crucial to be able to differentiate between the treatment philosophies that would work or would fail in the complex oral environment, so as to combat oral infectious diseases in the most effective way. The aim of this paper is to propose a holistic clinical and microbiological concept for all oral infectious diseases and apply it to current and future prophylactic and therapeutic treatment strategies. This concept provides a persuasive model and an effective pedagogical tool that underpins our understanding of the disease process and biological responses to therapy.

A holistic clinical and microbiological concept: the ecological rationale

The reductionist approach, which views plaque-mediated oral diseases as mono-infections, has been refuted and slowly abandoned over the last three decades. The study of individual bacterial species in the oral cavity has moved to the study of biofilms, i.e. surface bound complex inter-dependant microbial communities. Substantial effort has been invested in identifying and naming all the specific units of biofilms both in oral health and disease. The newest sophisticated molecular techniques have contributed significantly to their taxonomy compared with the traditional culture techniques, which were unable to reveal the most fastidious and difficult-to-culture bacteria. The clinical significance and the contribution of these modern techniques in understanding the pathogenesis of the disease need not be all that promising. Identifying the panorama of the oral microbiota may reveal the complete picture but will not improve disease diagnostics and consequently therapeutics. Biofilms have received great attention in research, but they are still not the key to understanding the pathogenesis of the disease; rather they describe how the microbes are organised, but do not shed further light on the disease process. Microbiologists should continue to study microorganisms in biofilms, not as independent entities but always in the context of their specific environments and with a view to gaining insights into their function in these environments. This is in line with the so-called ‘ecological plaque hypothesis’ and should play a role in future research.

The ‘ecological plaque hypothesis’, described by a British microbiologist [1] but based on a Swedish researcher [2], seeks to explain the pathogenesis of oral diseases. It states that oral bacteria require a specific environment to cause disease and clearly explains why bacteria are essential but not sufficient to cause caries, periodontitis or peri-implantitis. Even in the case of endodontic infection, where bacteria are present in sufficient numbers to cause apical lesions, the degree of pathogenicity of the bacteria involved in the lesion and thus the severity and extent of the lesion are determined by local environmental factors. This theory lays the foundation for investigation of the functions of microorganisms in their natural environments both in health and disease. This constitutes a paradigm shift; we move from static descriptive diagnostics to dynamic functional diagnostic efforts. This change of focus should guide future microbiological research directly into the complex clinical environment, that is beyond just the *in vitro* settings.

Establishing the pathways of pathogenesis of a disease in a correct setting is very critical for treatment strategies, as this formulates the rationale behind what and how to treat. Thus, by ‘mapping out’ the ecology of specific clinical condition, we are able not only to explain the pathological mechanisms behind plaque-mediated oral diseases but also to identify true clinical endpoints of our therapy. We can design and apply effective treatments to achieve these ends. Until now, microbiological aspects of the disease have been mostly descriptive and not fully linked to the clinical reality. Microbiology combined with ecological principles becomes a new vista in the eyes of the clinician, as it reflects not only the names and counts of bacteria, but a point of reference by which one could judge present and future treatment options, leading to successful treatment planning.

Effective treatment of caries from an ecological perspective

Dental caries is an infectious process, wherein bacteria trigger damage to the hard tooth structure (enamel, dentine and cementum). The progressive breakdown of the dental tissues leads ultimately to the creation of deep cavities and pulp exposure. The classical animal studies [3-5] firmly established the principle that dental caries is a bacterial infection. Since the later part of the nineteenth century, it has been widely acknowledged that caries results from the production of metabolic acids by oral bacteria through fermentation of sugars leading to the demineralisation of dental hard tissues. Caries development is a highly dynamic series of processes with alternating periods of progression and regression or arrest [6-7]. Bacteria are essential but not sufficient to develop caries.

The microbial aetiology of caries is of low specificity. Mutans streptococci and *Lactobacillus* spp. have been mainly discussed to be associated with the aetiology of caries. Among the species of mutans streptococci, *Streptococcus mutans* has been claimed to be the most important. *S. mutans* has three main virulence factors making it the most common cariogenic bacteria: acidogenicity, acidurity and adherence [8-9]. Traditionally, *S. mutans* adheres to oral surfaces and produces extracellular insoluble polyglucans from sucrose. Moreover, it has the ability to adapt (by stress response), to tolerate and to survive under acidic conditions, which inhibit many other plaque bacteria. The increased number of *S. mutans* that is seen concomitantly with increased caries activity and caries lesion development may thus be a result of an ecological change, rather than due to *S. mutans* itself. Caries is not a mono-infection and the presence of *S. mutans* alone neither implies a carious process [10] nor predicts future caries. *Lactobacillus* spp. have long been regarded as potential aetiological agents in dental caries due to their acidogenicity and acidurity (acid tolerance). They may be of some significance in a limited number of lesions, but they are probably more important in the progression of established lesions [9, 11]. Other bacteria, all belonging to the normal flora of the oral cavity, have also been associated with caries, implying that caries is an endogenous, polymicrobial, opportunistic infection [12]. Other species of the genus *Streptococcus* such as *S. sobrinus*, *S. intermedius*, *S. gordonii*, *S. oralis*, *S. salivarius* as well as *Veillonella* spp. (*V. denticariosi*) and *Actinomyces* spp. (*A. gerencseriae*, *A. israelii*, *A. naeslundii*, *A. oris* and *A. odontolyticus*) may also have cariogenic potential and may be capable of initiating the caries process at susceptible tooth sites [9, 13-15]. Even some strict anaerobes such as *Prevotella* spp. and *Fusobacterium* spp., not only tolerate acidic conditions [16] but also produce acid, to an extent that may not greatly differ from streptococci [17].

The ecological plaque hypothesis is very attractive as members of the resident flora under specific ecological circumstances obtain a selective advantage over other species and perturb the homeostatic balance of the biofilm [1]. A number of local or systemic factors could give rise to such ecological shifts in the plaque microbiota, including dietary influences (particularly sugar intake) and alterations to the flow of saliva. An obvious example relevant to the initiation and development of caries would be the increased availability of fermentable carbohydrate, leading to acid production and lowering of the local plaque pH. Such a pH-based change in environmental conditions would in turn favour an ecological shift towards more acid-tolerant species, such as mutans streptococci and *Lactobacillus* spp. [18]. An extended ecological plaque hypothesis has been suggested with regard to caries [19]. According to it, the presence of *Actinomyces* and ‘low pH’ non-mutans streptococci may, under conditions of frequent sugar supply or reduced salivary secretion, adapt to produce acid (stress-response), which destabilises the homeostatic biofilm, causing a change to a more acidogenic plaque biofilm. The presence of mutans streptococci and *Lactobacillus* spp. is, at this stage, facilitated and may lead to further acceleration of the carious process. However, it would be a narrow-minded approach to ‘frame’ groups of bacteria for specific roles in the carious process, as they may vary on an individual level, and pathogenicity of microorganisms is highly dependent on the specific ecological determinants of the site; thus, such statements cannot be generalised.

Implicit in the ecological plaque hypothesis is the concept that, in order to arrest caries, we should interfere with ecological factors that drive the deleterious shifts towards disease (e.g. lower the acidic challenge). Targeting directly the 'putative pathogens' (e.g. mutans streptococci and *Lactobacillus* spp.) [20, 21] may not necessarily bring the desired ecological changes. On the other hand, ensuring an ecology shifted towards health will lead to reduction of 'pathogens' to a level equivalent to the host balance (homeostasis) and this is the consequential effect. The focus of interest should be shifted from elimination of specific cariogenic microorganisms to nurturing, selecting and favouring a non-acidogenic biofilm. Alkalogenic flora, bacteria that produce alkali, could serve as acid blockers which lower the acidic challenge and help to re-establish health [22]. A process of using genetically engineered alkali-producing streptococci has been described in [23]. This alkaline environment resulting from ammonia produced from arginine and urea would also be supported by the ecological plaque hypothesis [24]. Randomised controlled clinical trials need to be launched to support the application of this potential therapy.

To modulate the microbial ecology of caries by using relatively simple approaches such as diet control, oral hygiene and usage of fluoride leads to predictable and successful results. Diet control and selection of non-cariogenic foodstuffs modify the substrate and deprive the acidogenic and aciduric bacteria of harmful exogenous nutrients, which are critical for their survival and growth. Limiting the frequency of sucrose intake also reduces the time that the substrate is in the mouth. Mechanical removal of plaque from the tooth surface, by efficient oral hygiene measures, disrupts the biofilm, lowers the number of micro-organisms in contact with the tooth and maintains the ecological homeostasis despite environmental stresses. However, the chemical effect of fluoride in most dentifrices seems to be even more critical against caries development and may alone explain the dramatic reduction in caries in recent decades in the western world. Dentifrices with even higher fluoride concentration (5,000 mg/L) proved to be an important vehicle for caries prevention in a two-year clinical trial [25]. Fluoride in such high concentrations may have an enzyme inhibitory effect and thus slow down the metabolic activity of cariogenic bacteria, if delivered close to sugar exposure. Other anti-plaque agents such as quaternary ammonium compounds, bisbiguinides (chlorhexidine), enzymes, metal salts, essential oils and plant extracts have been used, mostly as mouthwashes. The critical issue in combating caries effectively is that these substances retain their antimicrobial effect in an open growth system. The relatively short contact time between the inhibitor in the rinse and the mouth makes substantivity very important. Dentifrice is another vehicle for the unsuspended delivery of antimicrobial agents but many proven antimicrobial and anti-plaque agents, such as chlorhexidine, are incompatible with the components of toothpastes and lose their bioactivity.

Other approaches to modulate the flora and control caries, which seem more sophisticated and attractive in concept and delivery, are vaccine development [26] and replacement therapy [27]. Nevertheless, no experiment in which animals were immunised [28, 29] demonstrated an absolute protection against caries. Vaccination is no longer a topic under discussion, mainly because the long-term effect of altering the indigenous oral microbiota is highly unpredictable. Moreover, the epidemic character of caries has declined and no longer justifies the use of a vaccine as a caries prevention vehicle, given the substantially reduced prevalence of caries in western countries over the past several decades. Replacement therapy - currently named probiotic therapy - was another idea based on the possibility that indigenous antagonistic organisms could be exploited to block the major caries pathogens. In this approach, a harmless effector strain is permanently implanted in the host's microbiota. Once established, the presence of the effector strain prevents the colonisation or outgrowth of a particular strain, such as *S. mutans* in the case of caries [30]. Neither immunisation nor replacement therapy, though they may sound promising, can guarantee elimination of the disease. They are directed specifically at mutans streptococci and, despite potential reduction of these species in caries, no clear ecological changes are triggered and caries may not necessarily be arrested. Caries control

presupposes changing the pH balance from acidic to alkaline, which in turn slows down the growth and metabolism of cariogenic bacteria.

Effective treatment of endodontic infection from an ecological perspective

Endodontic infection is initiated as soon as microorganisms enter into the root canal system. Apical periodontitis serves an important protective function and seeks to prevent the spread of bacteria and bacterial elements from the root canal system to other body compartments. An early observation, relevant to the pathogenesis of apical periodontitis, was proposed by Miller [31] and proven approximately 70 years later by a classic study in germ-free rats [32], subsequently confirmed by numerous studies [33-35]. Like other 'plaque-mediated' diseases and unlike classic medical diseases, apical periodontitis does not have a 'single species aetiology'. Indeed, studies using anaerobic culture and sophisticated molecular biology methods [36-38] have demonstrated that apical periodontitis is a mixed, polymicrobial opportunistic infection. A set of endogenous oral bacteria usually organised in biofilm communities is involved in the disease process.

The main ecological difference in the case of the pulp compared with the periodontal tissues is that we are not dealing with an open growth environment. An intact pulp is bacteria-free and thus, never in contact with the commensal bacteria in the oral cavity. Moreover, when the previously intact pulp finally succumbs to bacterial attack, the infection is confined to a contained space, within hard tissue walls. Limited communication with the oral cavity results in limited access to nutrients through blood and serum permitting only a restricted number of microorganisms in the root canal compared with the much higher number in the periodontal pocket [39]. This explains plausibly why endodontic bacteria, in contrast to subgingival bacteria, are less likely to be translocated into the circulatory system and trigger systemic complications. They are relatively few in numbers and virtually contained within the root canal.

Elimination of bacteria from the root canal space would mimic the healthy situation prior to infection. Moreover, it is an achievable goal from an ecological perspective because the actual space in which bacteria survive and thrive is limited and delimited. It remains a challenge to fully eliminate the bacteria due to the anatomical complexity of the root canal system and specifically the presence of dentinal tubules, isthmuses, ramifications, accessory canals and the periapical delta, where bacteria can reside and become inaccessible.

The goal for the treatment of apical periodontitis should be an ecological change in such a way that bacteria find it virtually impossible to grow within the root canal. Due to the exposure of the root canal to oral bacteria, it is also critical to prevent the iatrogenic introduction of oral bacteria into the infected root canal system. Thus, a prime aim is to work in an aseptic environment using the rubber dam routinely, to maintain a clean and dry operative field and to prevent access of extraneous microorganisms from the saliva and the gingival exudate to the wound site. Without the use of rubber dam, proper asepsis in endodontics cannot be attained and we run the risk of contamination, leading often to long-lasting infections that are difficult to control. Since with current technology we have not reached the optimal goal of regenerating lost pulpal tissue, we must try to eliminate bacteria from the root canal space, so as to mimic the bacteria-free condition of the pulp prior to infection. This is achieved by biomechanical preparation of the root canal system with mechanical (filing) and chemical (irrigation with agents) means in one or two visits [40]. A permanent root filling is performed by obturating the instrumented root canal space in order to prevent recurrence of the disease. Mechanical instrumentation of the root canal disrupts the biofilm, just as it does in the case of periodontitis. Chemical means are also combined with the instrumentation process. They clean out debris and dentine shreds by anti-septical irrigation [41]. The main types of antibacterials used are sodium hypochlorite, chlorhexidine digluconate, alcohol, iodine and complex commercially available medicaments. They have been used both as irrigants at the time of instrumentation as well as intracanal medicaments between appointments.

The different ecological conditions that exist in the root canal compared with the periodontal pocket explain why the chemical approach may have some effect in the first case but not in the second. The root canal occupies a defined space;

the pulpal tissue is encased within rigid hard tissue walls. This feature coupled with the fact that the vital pulp is devoid of microorganisms, makes chemical infection control of the root canal predictably effective [42]. In addition, irrigation takes place in a dry, controllable and not an open growth environment where antimicrobials can perform at an optimal level retaining their concentration and antibacterial properties.

Despite the adjunctive effective role of antiseptics during mechanical instrumentation, we frequently face failure in endodontic treatment. One of the main reasons for endodontic treatment failure is the inability to clean the entire root canal system due to its complex anatomy [43]. Moreover, bacteria in this complex system are organised in biofilms, which are considered to be difficult therapeutic targets [44] and this adds to the difficulties of creating a bacteria-free environment [45]. The irrigation acts quickly, via flushing of the root canal and often does not give sufficient time for the active chemical agent to interfere with the bacteria. Increased concentration of the active agent is one option but the ideal concentration of sodium hypochlorite and the potential side effects still remain a controversial topic. The best antiseptic concentration of 5% sodium hypochlorite should be used [46] if we are determined to utilize the maximum potential of the available antiseptics towards eradication of the endodontic infection. At the same time, part of the 'price' is accepting the side effects. Every battle has its losses but as long as the endpoint of the therapy is achieved, some compromise is acceptable. Various intracanal medicaments have been used as a dressing to augment the time the active chemical agent remains in contact with the infected root canal system [47]. The long-term use of such medicaments may be important for long-term successful outcomes, especially when we are dealing with persistent root canal infections.

The ecological principles also explain a paradigm shift in the treatment of apical periodontitis. It was common practice in the past, in order to combat severe apical periodontitis lesions, to leave the tooth open for drainage without a temporary filling for a limited period of time [48]. This practice was based on the argument that, by direct communication between the root canal and the oral cavity, we induce aerobic conditions, which are unfavourable for the growth of the anaerobic bacteria of the primary pulp infection. Today, understanding the ecological principles, we now realise that by leaving the pulp chamber open, we create an open growth system, similar to a periodontal pocket and thus facilitate bacterial invasion [39]. This creates a secondary persistent infection, often harder to eradicate.

Following chemo-mechanical debridement, root filling is performed, aiming at preventing recurrence of the disease. An impermeable root seal is critical for obturating the disinfected root canal space and sealing any remaining bacteria in the dentinal tubules, their branches and apical delta. Thus, it may be utopian to seek to eliminate bacteria totally from the root canal system, due to the inability of chemo-mechanical agents to penetrate such tortuous anatomical configurations. Bacteria colonising such niches may survive but as long as they do not grow and their metabolic activities are close to zero, they do not pose any threat to the longevity of the root-filled tooth. We may have to accept incompletely healed apical lesions around root-treated teeth and thus some local inflammation at the apex, as long as the size of the inflammation and the lesion do not increase over time.

Effective treatment of periodontitis from an ecological perspective

A clear paradigm shift in the pathogenesis of periodontal disease has occurred in recent decades, summed up by the statement that 'periodontitis is an endogenic, polymicrobial opportunistic infection' [49]. Clustering different groups of bacteria, as attempted and demonstrated by the use of red and orange complexes [50, 51], is still not the key in understanding the pathogenesis of periodontitis as there are divergences from this clustered model in the clinical reality [52].

In periodontitis we are dealing with an open growth system and not a closed, bacteria-free environment. Bacteria are present and have the chance to become opportunistic pathogens more than once, meaning that treatment does not necessarily imply life-long periodontal health. Thus, a rational goal is to seek to control the disease, not to cure it. Clinical endpoints of successful treatment would be i) no repeated bleeding

following pocket probing and ii) pocket elimination, a probing pocket depth (PPD) of ≤ 4 mm. Another sign suggesting successful control of 'subgingival infection' is increased radiographic density of the marginal bone crest or at angular defects. Following the ecological principle, we should choose treatment strategies that establish clear ecological change, as well as facilitating optimal oral hygiene.

Supragingival plaque control is the cornerstone of periodontal treatment because it establishes long-term periodontal stability and guarantees the long-term successful result of our professional intervention. The role of the dental clinician is very crucial in helping the patients to establish and maintain a high status of oral cleanliness. The constant application of meticulous supragingival plaque control measures is more critical in the case of maintenance of patients because it will directly affect the result of our non-surgical and/or surgical periodontal treatment. If supragingival plaque is present after meticulous professional pocket debridement, a subgingival microbiota similar to that of the untreated periodontitis site is re-established within 4-6 weeks, posing the risk for recurrence of the disease [53, 54].

The first intervention in periodontal therapy is subgingival mechanical debridement, in order to disrupt the subgingival biofilm. Whereas in the past, periodontal debridement was primarily performed by hand instruments, today power-driven instruments are used more often. Root debridement either by hand or ultrasonic instruments achieves the clinical therapeutic goal of pocket depth reduction. It makes a clear ecological change by the soft tissue management it triggers. Gingival recession and gain in clinical attachment are the result of the reduction of the microbial load accompanied histologically by fewer inflammatory cells, richer in collagen connective tissue and closely adapted to the junctional epithelium of the tooth or root surface, leading clinically to increased resistance to probing and thus, decreased probing pocket depth measurements.

The traditional purpose of initial periodontal treatment is to perform scaling and root planing (SRP) by jaw quadrant-wise SRP (Q-SRP) at a series of consecutive appointments [55]. A challenge to this traditional approach was proposed by a group of Belgian researchers, who argued in 1995 for the 'full-mouth disinfection' approach. It entailed undertaking all scaling and root planing in one stage within 24 hours [56]. However, attempting to eliminate bacteria from all intraoral niches in an open growth environment might be unrealistic and also this may not be essential. Re-colonisation of the treated sites by microbes from untreated sites may occur, but since the ecological changes have been achieved by mechanical debridement, microbial homeostasis is not disturbed in the long-run only by translocation of microbes. Such phenomena constitute part of the moderate environmental stresses. Recent reviews also conclude that there is no clinical benefit from the full-mouth approach compared with the staged approach in initial debridement [57, 58].

The adjunctive use of chemical agents (antimicrobials) in mechanical subgingival infection control has been discussed extensively in the literature. Different sorts of agents have been tested in different forms: as mouthrinses, as irrigants to flush out the periodontal pocket, and as gels or chips with the intention of a more sustained effect in the pocket. From an ecological perspective, to create a 'bacteria-free' environment temporarily is not necessary for periodontal stability. Strong *in vitro* promises with regard to their killing effect do not reflect the real *in vivo* clinical situation, where hopes of true additive effect are shattered. Substantivity is a very critical property, which antiseptics are unable to maintain in an open growth environment such as the oral cavity, where a variety of clearance mechanisms exist. We cannot have extreme expectations of an antiseptic, which has to be harmless to teeth and tissues and thus cannot be used in extremely high concentrations, and in addition is expected to be useful in an unfriendly, moist environment. As soon as it comes into contact with saliva or gingival crevicular fluid, it is inactivated and loses most of its antibacterial properties. Chemical agents applied locally in the periodontal pocket have only slight and short-term effects on the subgingival microbiota, as shown by early reports [59, 60] and highlighted in review studies [61, 62].

In cases where the response to a non-surgical approach is sub-optimal, such as the persistence of residual bleeding pockets, additional surgical therapy should be considered. One of the major

objectives of surgery is, by pocket elimination, to create a situation where it is possible to re-establish and maintain a healthy condition in the subgingival area by proper oral hygiene [63]. Reconstructive or resective procedures or combinations of these fulfil this aim. Bone re-contouring and apical repositioning of the flap, though they may seem invasive, should be preferred to open debridement. The strongest argument for open flap debridement is visibility and thus effectiveness against biofilm situated in inaccessible areas (concavities, hollows, furcations). Access flap surgery is primarily directed towards the elimination of the subgingival microbiota and may not induce the desirable changes in the environmental conditions and consequently the site may remain at risk for future re-colonisation. A pioneering study with an ecological perspective was that of Mombelli *et al.* [64], which demonstrated that health can be achieved by alteration of the local environment (from anaerobic to aerobic) alone, via apically repositioning flap surgery. This was sufficient due to the reduction in microbial load but not necessarily a change in the local environment. From a microbiological point of view, the only way to change the ecology completely is by pocket elimination.

Complete and predictable regeneration in periodontal defects is still a difficult and challenging goal. With regard to reconstructive approaches, one can understand the difference in philosophy between the use of Emdogain® (EMD), Guided Tissue Regeneration (GTR) and Bone Replacement Grafts (BRGs), having in mind an ecological perspective. A periodontal pocket, unlike a root canal space that needs to be 'filled', is not a bone disease but a soft tissue disease. The bony destruction is a secondary effect. The deposition of 'cosmetic' bone by bone grafting, which is unpredictable, is not a biological concept for true regeneration nor does it entail a true change of the ecological conditions at the site of the defect. GTR, though based on a biological principle [65], is still a mechanical strategy. It seeks to make space by insertion of a barrier. Space making is not a factor of prime importance for true periodontal regeneration, in contrast to other factors such as wound stability and primary intention healing. These may be disrupted by insertion of a membrane (barrier). A barrier *per se* does not prevent bacteria from colonising and invading the space. It is a 'foreign body' and, if exposed in the oral cavity, there is a potential risk of becoming infected due to lack of tissue coverage [66]. On the other hand, EMD seems to offer the most promising and predictable prospects for regeneration attempts. It mimics the events that take place during the nascent development of the root and the periodontal tissues [67, 68]. Thus, EMD promises true regenerative outcomes by mimicking nature and by not forcing nature via a barrier. It must be borne in mind that any regenerative attempt is meaningful only in a healthy environment, implying that debridement and cleaning of the site are prerequisites before any attempt at regeneration is made. Even EMD does not work in the wrong ecology, as it can be degraded and inactivated by bacterial enzymes [69].

Effective treatment of peri-implantitis: an ecological perspective

Peri-implantitis shares similarities with periodontitis with regard to terminology, risk factors, diagnosis and treatment. It is also a polymicrobial endogenous opportunistic infection that is attractive for the 'ecological plaque hypothesis'. One apparent difference is the much more complicated surface structure of an implant with which we are dealing. The tooth has predictable anatomy that is genetically determined and a smooth surface, favourable for instrumentation. The implant has a position and shape that varies in the dentition and is determined by the operator. The geometry of the implant with threads of different design, and often a rough surface to enhance osseointegration, may also impede the ability of the clinician to detect and remove calculus and plaque located below the mucosal margin. In certain cases, where the position of the implant is not ideal, the prosthetic superstructure can also be overextended for aesthetic purposes or constructed in a way that restricts access for oral hygiene. A thick prosthesis not only hampers the patient's efforts to brush effectively at the mucosal margin, but also renders difficult the correct probing by the clinician and the assessment of the true severity of the disease, even if thorough mechanical debridement can be achieved.

A recent retrospective study of peri-implantitis cases clearly showed that peri-implant health is difficult to establish [70]. According to the Cochrane review, there is very little reliable evidence suggesting which could be the most effective interventions for peri-implantitis [71]. The various designs of the implants make direct comparisons of different treatment modalities very difficult and extrapolating definite conclusions is almost impossible, resulting in a great heterogeneity of studies focusing on implant therapy compared with homologous therapy on teeth.

Peri-implant mucositis and mild incipient peri-implantitis lesions may be resolved using the cause-related measures and a non-surgical approach [72]. Non-surgical debridement can be hard to perform fully at implant surfaces, as we have no tactile sensitivity for the position of calculus and plaque and there are risks of damaging the implant surface and interfering with the established osseointegration. It is thus recommended that non-surgical debridement of implant surfaces to remove calculus and plaque should be restricted to the portion of the implant located coronally to or at the level of the mucosal margin [73]. While calculus may be chipped off using carbon fibre or plastic curettes, plaque is removed by polishing the implant surface with rubber cups and a polishing paste. Carbon fibre curettes should be preferred for implant instrumentation compared with conventional steel curettes or ultrasonic instruments with metal tips because they do not damage the implant [74].

Non-surgical therapy alone does not seem to be effective in moderate or severe peri-implantitis lesions [75]. The threaded design of the dental implant, coupled with the surface roughness, often promoted in the clinical practice for faster osseointegration, does not facilitate access for optimal cleaning by mechanical and chemical means. A recent study on the remaining biofilm forming intra-orally on titanium discs after cleansing revealed that on moderately rough surfaces, the biofilm was complex and firmly attached, whereas on turned surfaces it had a pattern of spread bacteria, forming fewer clusters [76].

In accordance with the ecological paradigm, among the surgical techniques proposed to arrest progression of peri-implantitis, access flap surgery alone may not trigger and sustain the ecological changes needed to favour health. In contrast, apically repositioned flap combined with bone remodelling, though radical it may be in certain cases, creates a positive architecture for hard and soft tissues and allows the patient to practice optimal oral hygiene. Favourable outcomes have been shown by use of this treatment [70, 77].

Regeneration of the lost peri-implant tissues, as in the case of periodontitis, is the most challenging and most desirable intervention. It does not involve resection of intact surrounding bone, which would imply apical displacement of the soft tissue margin and aesthetic concerns for the patient. The wide bony crest opening, as often encountered in typical saucer-like peri-implant defects, may enable the use of regenerative materials. Two systematic reviews [78, 79] comment on the high variability in the amount of bone filling, which may be due in part to different defect morphologies, different measurement methods and different investigation procedures. A recent clinical trial [80] reported favourable regenerative outcomes over a period of three years by the use of a bone substitute with or without a membrane in the treatment of peri-implantitis. Indeed, there are some severe cases where the only option might be the removal of the implant. This is critical before any re-osseointegration is attempted, so as to eradicate the problematic biofilm, associated with the heavily diseased implant.

Role of antiseptics

Antiseptics are general antimicrobial substances applied to living tissues that eventually kill or inhibit the growth of microorganisms. They are toxic to both infectious agents and host cells, are widely used in skin and mucous membranous infections [81] and have in general a little propensity to develop resistance. The most commonly used antiseptics are alcohol, essential oils, iodine, sodium hypochlorite, chlorhexidine and hydrogen peroxide. Antiseptics have been incorporated into dentifrices, mouthrinses, gels, varnishes or are used separately as irrigants directly or as slow-release medicaments with an overall goal to act as adjuncts to the mechanical approach in order to combat oral diseases more effectively.

In vitro studies of antiseptics with regard to their antimicrobial effect have often been very promising but the *in vivo* clinical reality does not always correspond. They can provide supportive data to clinical investigations but cannot stand alone as proof of efficacy *in vivo*. The concentrations of antiseptics in toothpastes and mouthwashes drop significantly by the time they reach the 'open growth system' of the oral cavity. A variety of clearance mechanisms take effect: i) actions such as swallowing, mastication, blowing the nose; ii) active motion via the ciliae (nose and sinus); and iii) most importantly, the wash-out effect of the salivary, nasal and crevicular fluid flow. Another critical issue is substantivity, the ability of antiseptics to retain their properties. Povidone iodine at 1% has a substantivity of only 60 minutes [82] whereas chlorhexidine, though more of a preventive than a therapeutic agent, shows persistent bacteriostatic action lasting in excess of 12 hours [83]. This implies that antimicrobial properties of antiseptics vary not only in magnitude but also in persistence. Prolonged persistence of antimicrobial action is also ecologically dependent; it is modified by the quantity and quality of biofilm and the surrounding organic material. In the case of chlorhexidine, its substantivity in root canals is much longer compared with periodontal tissues. In two *in vitro* studies, the substantivity of chlorhexidine solution 2% as irrigant of the root canal was found to be 72 hours [84] and 48 hours [85], respectively, due to the presence of organic material. The different ecological conditions explain the increased substantivity of the antiseptic in the root canal compared with the oral cavity, the periodontal pocket or the peri-implant pocket. The root canal system is a 'closed' system compared with the 'open growth system' of the oral cavity, because the root canal is a contained space that can easily be dried and thus is controllable.

The above ecological principles explain why different antiseptics, used as irrigants in the root canal system, have greater chances to be fully effective but are unable to be so in the case of periodontal tissues. Incorporation of antiseptics into dentifrices and mouthwashes has only a rinsing effect and the antimicrobial properties of such products are of too short a duration to be critical in the establishment and maintenance of oral health. Mouthwashes containing chlorhexidine can prevent plaque formation and are proposed for periodontal patients for a short period when they find it difficult to brush or they could disturb the wound healing process, such as after oral surgery. Antiseptics have also been used as subgingival and submucosal irrigants in chronic periodontal and peri-implant lesions, producing some slight additive effects [72, 86].

Role of air-polishing, photodynamic therapy and lasers

Interest in air-polishing for subgingival debridement was renewed when sodium bicarbonate was replaced by a patent, finely grained, low-abrasive glycine powder [87]. The principle is that this powder, inside a powder chamber, stirred up by pressurised air and a flow of air and water is transported to the tip of an air-polishing nozzle. Glycine powder air-polishing (GPAP) resulted in a significantly greater reduction in subgingival bacterial counts than hand-instrumentation and, it was suggested, this could be used effectively in debridement of shallow periodontal pockets [88]. A recent clinical trial using probing pocket depth reduction as the primary outcome variable failed to show enhanced efficacy of GPAP compared with ultrasonic debridement of moderately deep pockets in maintenance patients [89]. Another randomised controlled trial assessed the efficacy of GPAP in moderate to deep periodontal pockets and clearly showed that the improved microbiological outcomes were not accompanied by enhanced clinical outcomes, as probing pocket depth reduction and bleeding on probing remained largely unchanged [90]. This is in line with the ecological paradigm because transient reduction in total bacterial counts might not alone trigger and sustain an ecological change conducive to health. GPAP was also used in a six-month clinical trial in the non-surgical treatment of peri-implantitis with limited clinical efficacy and failure to reduce bacterial counts [91, 92].

Photodynamic therapy has also emerged as an alternative to antimicrobial agents and mechanical means of eliminating dental plaque bacteria, targeting their cytoplasmic membrane and DNA. It aims at generating singlet oxygen and free radicals that are cytotoxic to bacteria, based on the

concept that a photosensitiser which absorbs light can be preferentially taken up by bacteria and subsequently activated by light of appropriate wavelength [93]. The effect of light in killing bacteria organised in biofilms proved to be much weaker compared with bacteria in a planktonic state [94]. Whether the promising biological effect in treatment of caries, endodontic infection, periodontitis and peri-implantitis, as shown *in vitro*, is of any clinical significance needs to be validated by clinical assessments.

In the absence of efficient antimicrobials in the oral cavity, lasers seem attractive, but lasers are merely at the start of a long process before they are available for everyday use; their clinical effectiveness remains controversial. The erbium-doped yttrium-aluminium-garnet (Er:YAG) laser is the most promising for hard tissue applications in the oral cavity. A recent systematic review with meta-analysis [95] based on five trials failed to identify increased clinical efficacy of Er:YAG laser therapy compared with scaling and root planing at 6 and 12 months.

The role of antibiotics

Local delivery of antibiotics has been attempted in the treatment of chronic periodontitis, to provide an effective concentration of the drug at the target site with minimal systemic load. Once the drug reaches the site of action at an effective concentration, it must remain long enough for the pharmacological effects to occur at the site. This is not so easy because the drug is lost by crevicular fluid clearance in an established open growth system, as in the case of periodontitis. As far as we know, four local antibiotics have become commercially available: tetracycline fibres, metronidazole gel, minocycline ointment and doxycycline hyclate in a resorbable polymer. These drug delivery systems have not been shown to provide superior results compared with scaling and root debridement [61, 62].

With regard to systemic use of antibiotics, a lot of valid arguments based on the ‘ecological plaque hypothesis’ can illustrate the dilemma, to use or not to use. Antibiotics have no application in the carious process, where the infection is limited to dental hard tissue (enamel, dentine). In the case of periodontitis and apical periodontitis, the infection is in a highly vascularised ecological environment and there is a risk of infection spreading through the blood system to other parts of the body, causing systemic complications. The rationale behind the use of systemic antibiotics in oral infections is that the antimicrobial agents will reach the infected tissue through the blood system and eliminate the infection. Antibiotics are administered in oral diseases mostly *per os* and not intravenously, as the infections are sub-epithelial but not deep-seated. It is also important to realise that antibiotics inhibit bacterial growth and do not kill the microorganisms in the real clinical environment. Classification of antibiotics as bactericidal and bacteriostatic are used to indicate *in vitro* antimicrobial potency without having real clinical significance. This distinction is insufficient to predict clinical outcomes.

Acute forms of oral diseases should be countered with systemic antibiotics without any restrictions when there are clinical signs of systemic involvement. Fever, general discomfort and diffuse intraoral swellings are absolute clinical indications for administration of systemic antibiotics. In such acute conditions, antibiotics have optimal efficacy for two reasons. They target the high metabolic activity of bacteria (cell wall synthesis, protein synthesis or DNA/RNA synthesis) and can slow down or abolish their growth and thus intervene with their enhanced virulence under this sudden outburst. This is critical to prevent bacterial spreading, dissemination of the infection and severe complications. Moreover, it has been demonstrated for some pathogens that genes coding for many virulence factors are much more highly expressed in planktonic cells than in sessile (biofilm) cells, suggesting that planktonic cells are more likely to participate in acute infections [96]. In this planktonic local environment, cells are more susceptible to antimicrobial agents and phagocytosis [97-99] and assimilation of antibiotics is a lot easier compared with the biofilm environment, explaining their increased efficacy. Effective early management of the acute infection with drainage and systemic antibiotics is of prime importance. Such a treatment strategy clears the infection and either prevents further development of disease, if acute infection emerges for the first time, or

prevents progression if it emerges as an exacerbation of a chronic infection. In acute forms of periodontal disease (e.g. necrotising forms, periodontal abscess etc.) scaling is of secondary importance and is better to be postponed until the acute stage subsides.

Antibiotic use should not be our first and foremost priority in the treatment strategy against chronic infection. Chronic infection is the continuation of infection beyond the time when the host defense system might reasonably be expected to clear the acute infection. Once established, it reflects a diseased equilibrium between microbes and host. Thus, in the case of chronic periodontitis, soft and hard tissue management should be the prime aim of all treatment modes, rather than the elimination of bacteria, for long-term periodontal stability. One can argue that there is a strong body of evidence with clinical trials showing improved microbiological and clinical parameters when antibiotics are used as adjunct to mechanical debridement in chronic periodontitis [100-108]. Microbiologically, this is to be expected because the associated biofilm is in a dynamic state, containing also planktonic bacterial cells, which antibiotics may eliminate. In chronic periodontitis, the bacteria remain in a low metabolic state, and the clinical significance of the use of antibiotics is questionable.

Another burning issue in periodontics is the administration of systemic antibiotics in aggressive periodontitis patients. The term ‘aggressive’ has never been used for caries or apical periodontitis but was introduced in the most recent classification of periodontal diseases [109]. It is more of a descriptive term and not a diagnostic term as it does not imply different treatment modalities compared with chronic periodontitis. The differentiation between aggressive and chronic periodontitis is mainly done with regard to the time frame (span) in which the disease has developed. In the case of ‘aggressive periodontitis,’ this cannot always be assessed with certainty, as the primary criteria might not be covered in full [110]. Aggressive periodontitis does not necessarily imply acute infection at the time of diagnosis; it may have been established and have already developed to chronic infection. On the other hand, a chronic periodontitis may also have a sudden exacerbation and undergo an aggressive phase. What is critical in our differential diagnostic procedures is to try to identify the acute phase of the disease, because this knowledge will have a direct impact on long-term successful treatment plans. The term ‘aggressive periodontitis’ has been misused extensively in the literature and correlated with the absolute use of antibiotics, although we still face a chronic and non-acute phase of the disease. There is supporting evidence to confirm successful treatment of aggressive periodontitis without the use of antibiotics [111-113]. Thus, the answer to the dilemma on whether to administer antibiotics or not should be based on the characterisation of the infection (acute, subacute or chronic). Probably, the only exception for which the term ‘aggressive’ is truly relevant from a microbiological perspective is the case of the previously called ‘localised juvenile periodontitis’ or ‘early-onset periodontitis’ [114]. *Aggregatibacter actinomycetemcomitans* periodontal infection and the particular JP2 clone have clearly been implicated in the aetiology of periodontitis in adolescents [115]. Even in this classical monospecific infection, the lesion burns out and heals over time and this may well be explained by antibody production against leukotoxin.

Role of probiotics and prebiotics

Probiotics are live microorganisms that confer a health benefit to the host when administered in adequate amounts. They were first introduced to combat gastro-intestinal diseases and appear to act through colonisation resistance or immune modulation. The probiotics with the greatest number of proven benefits for intestinal microbial balance are *Lactobacillus* and *Bifidobacterium* spp. The effects of any probiotic depend greatly on the particular strain used. Varying test results imply great heterogeneity and no reproducible results should be expected from studies that employ different strains or species, varying formulations and diverse dosing schedules [116].

Attempts have been made to use probiotics in other environments, such as the oral cavity, in the hope of a similarly beneficial effect as in the intestine [117]. In the mouth, the aim was to use probiotics as a therapeutic tool for the prevention and treatment of dental caries and periodontal disease. Plausible mechanisms of probiotic action

within the oral cavity have been postulated on the basis of gastrointestinal studies and include direct interaction with dental plaque through competition for nutrients or binding sites on host tissue and other bacteria as well as indirect action via modulation of aspects of both innate and specific immune function [118]. *Lactobacillus rhamnosus* CG [119], *Lactobacillus casei* [120], *Lactobacillus reuteri* and *Bifidobacterium* DN-173 010 [121] have not demonstrated any ability to alter the colonization patterns of cariogenic bacteria permanently and thereby prevent caries.

Oral administration of probiotics has also been explored in the control of periodontal disease. A recent study [122] showed that the prevalence of hormofermentative lactobacilli, particularly *L. gasseri* and *L. fermentum*, in the oral cavity was greater among healthy participants than among patients with chronic periodontitis. This finding rejuvenated interest in a theory that lactobacilli could inhibit the growth of periodontal pathogens and prevent periodontitis. Long-term randomised controlled trials with large patient cohorts are needed to establish clearly the potential of probiotics in preventing oral infections. It has also been suggested in the literature that probiotics could be used adjunctively to scaling and root planing as replacement therapy (replacing microflora) in an attempt to inhibit periodontal pathogen recolonisation of subgingival pockets after scaling and root planing. The concept of guiding periodontal pocket regeneration [123] and the overall concept of probiotics in oral diseases should be regarded with great caution, however fancy and attractive they may sound. Such external environmental stresses cannot exert a sustained long-term beneficial effect on the local site, as bacteria are able to adapt to such challenges and retain their full potential to survive and thrive in the tissues.

The mucosal lining of the gastro-intestinal tract is able to sense and distinguish between molecular patterns shared by pathogens and non-pathogenic commensal microbes. Extrapolating probiotic colonisation behaviour on mucosal surfaces such as the intestine to the tooth surface, which is a non-shedding biofilm prone surface, seems to be a rather unrealistic scenario. True probiotic action would imply not just colonisation and adaptation to the oral ecosystem - that would be the easy step - but establishment of permanent residence in the oral cavity. There is no evidence that probiotics, even if they are of oral origin, are present in the oral cavity as a result of frequent consumption of dairy products, nor is there evidence that the oral cavity represents their natural and permanent habitat. *In vitro* studies have shown promising results of putative probiotic candidates maintaining their viability and integrating into the biofilm structure [124]. On the other hand, the *in vitro* situation does not reflect the *in vivo* situation, rendering the probiotic concept rather weak in exerting clear ecological changes and thus controlling oral diseases in the long term. In summary, probiotic bacteria seem to trigger merely transient effects as they fail to dominate in an already established ecological niche.

With the dynamic development of ‘functional food’ markets worldwide, the prebiotic concept, which was firstly introduced in 1995 [125], has also been attractive in relation to the control of plaque-mediated diseases. Prebiotics are selectively fermented ingredients that allow specific changes both in the composition and the activity of the gastrointestinal microflora that confer benefits on host well-being and health [126]. Prebiotics cause their effects through the metabolism of the bacteria they promote. Currently, only oligosaccharides in the fructo-oligosaccharide and galacto-oligosaccharide groups can be termed as prebiotics. Human milk contains oligosaccharides that have prebiotic characteristics [127]. It is known that the oral microflora of children are influenced to a large extent by the diet and that diet is a factor associated with caries. Within this concept, prebiotics could have a preventive effect on caries initiation and progression on condition that they induce clear ecological changes, such as change of pH. Research on prebiotics and oral health is still in its infancy.

CONCLUSIONS

It has been demonstrated that health in the oral cavity is not achieved in an environment devoid of microorganisms. Oral microbiota is symbiotic with us; we live together with it. In the case of pathogenesis of chronic oral diseases, we face a

paradoxical situation. The same indigenous species that support health also cause disease. In order to explain this, the ecological perspective is very beneficial. Oral health or disease is an adventitious event resulting from microbial communities adapting to the prevalent ecological conditions at a given moment. Identifying the key ecological determinants, as described above, is the critical step in decoding the transition from commensal microbiota to opportunistic pathogens and thus the transition from health to disease. Our current approach is that bacterial growth is a result of interactions between the bacteria and the oral environment and it is critical for such interactions to be deciphered. Features of the surrounding environment that trigger bacterial growth and thus pathogenicity should be identified and investigated. These ecological features not only contribute to the pathogenesis of oral diseases, by acting as determinants in the behaviour of potential pathogens, but they are also decisive for the effectiveness of treatments. We propose that various treatment strategies in the armamentarium of a dentist should be evaluated through the lens of ecology in order to understand how effective they are in combating oral diseases.

It is important to realise that eliminating microorganisms cannot be our prime aim, firstly because this is not feasible in an open growth environment but, most importantly, this is not necessary because creating a bacteria-free environment in the oral cavity does not mimic health. Treatment modalities should aim at changing the local environment in order to combat oral diseases successfully and to achieve long-term predictable treatment outcomes. The only exception is the root canal system in the case of primary infections, in which eliminating microorganisms is both essential and much more feasible due to the limited communication with the oral cavity. However, it might remain a challenge, even in this case, due to the complex anatomy of the root canal system and the consequential difficulties to reach the bacteria.

CONFLICT OF INTEREST STATEMENT

The authors declare that there are no conflicts of interest.

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