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Microbiology of endodontic diseases: A review article

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Abstract
Microorganisms play an unequivocal role in infecting root canal system. Endodontic infections are different from the other oral infections in the fact that they occur in an environment which is closed to begin with since the root canal system is an enclosed one, surrounded by hard tissues all around. The endodontic infections constitute almost 40—50% of the overall oral diseases. Pulpal and periapical pathology are the commonest debilitating form of oral disease with systemic implications. Bacteria detected from the oral cavity fall into 13 separate phyla, namely, Firmicutes, Bacteroidetes, Actinobacteria, Proteobacteria, Spirochaetes, Fusobacteria, Synergistes, SR1, TM7, Chloroflexi, Deinococcus, Acidobacteria, and Cyanobacteria. Microbes seeking to establish in the root canal must leave the nutritionally rich and diverse environment of the oral cavity, breach enamel, invade dentine, overwhelm the immune response of the pulp and settle in the remaining necrotic tissue within the root canal. During that time they have to compete in a limited space with other microbes for the available nutrition. As long as the enamel and cementum layers are intact, the pulp and root canal are protected from invasion, but loss of these structures by caries, cracks or trauma opens an avenue for penetration of bacteria through the dentinal tubules. From all the cases which report back with pain and infection after the endodntic therapy, it has been observed that E. faecalis is the most commonly found, with prevalence values reaching up to 90%.Other bacteria isolated in similar cases are streptococci P. alactolyticus, P. propionicum, F. alocis, D. pneumosintes, and D. invisus. This article gives an in-depth view of the microbiology involved in endodontic infections during its different stages.

Keywords: endodontic infections, enamel, microorganisms, pulp

Introduction
Microorganisms were observed in samples from teeth by Leeuwenhoek soon after he invented the microscope in 1684. Since Babylonian times, it was believed that a ‘tooth worm’ lived in the hollow portion of the tooth and caused decay. Leeuwenhoek challenged the ‘tooth worm’ theory of decay by identifying worm-infested cheese that he thought may be the source of disease [1]. Leeuwenhoek also described microorganisms that he scraped from teeth as ‘cavorting beasties’. However, it took over 200 years before his observation was confirmed and a cause and effect relationship was suggested by Miller [2]. Since 25 1890, when Miller first observed microorganisms associated with pulp tissue, microorganisms have been implicated in infections of endodontic origin.

Microorganisms play an unequivocal role in infecting root canal system. Endodontic infections are different from the other oral infections in the fact that they occur in an environment which is closed to begin with since the root canal system is an enclosed one, surrounded by hard tissues all around [3, 4]. Most of the diseases of dental pulp and periradicular tissues are associated with microorganisms [5]. Endodontic infections occur and progress when the root canal system gets exposed to the oral environment by one reason or the other and simultaneously when there is fall in the body’s immune response [6]. To begin with, the microbes are confined to the intra-radicular region when the ingress is from a carious lesion or a traumatic injury to the coronal tooth structure. However, the issue if not taken care of, ultimately leads to the egress of pathogens and their by-products from the apical foramen to the periradicular tissues. In total, bacteria detected from the oral cavity fall into 13 separate phyla, namely, Firmicutes, Bacteroidetes, Actinobacteria, Proteobacteria, Spirochaetes, Fusobacteria, Synergistes, SR1, TM7, Chloroflexi, Deinococcus, Acidobacteria, and Cyanobacteria.

Life is not easy for an endodontic pathogen. Microbes seeking to establish in the root canal

must leave the nutritionally rich and diverse environment of the oral cavity, breach enamel, invade dentine, overwhelm the immune response of the pulp and settle in the remaining necrotic tissue within the root canal. During that time they have to compete in a limited space with other microbes for the available nutrition. It is no accident that microbes berth in a particular environment there are ecological advantages for them to establish and flourish if conditions are favorable. Through genetic exchange and mutation, microbes have developed specialized systems that facilitate their ability to find, compete and survive in these very specific environments [7].

The micro-flora comprising the root canal is typically unique and specific. The endodontic infections constitute almost 40—50% of the overall oral diseases. Pulpal and periapical pathology are the commonest debilitating form of oral disease with systemic implications. Though success rate of endodontic therapy ranges from 30-90% of treated cases, failure rate is equally high accounting to millions. The blossoming advanced technological aids have probably detracted our attention from primary problem of endodontic disease.

The comprehension of the microbial location, characteristics and behavior in the root canal assists in decoding the disease process. However, for impromptu treatment outcomes, there is a need for this data to be translated into clinical practice. The dominance of certain species in some locations and geography-related pattern occurrence has been confirmed by Community-profiling. Most of the current knowledge of endodontic taxonomy is based on international reports. The geographical variation, ethnicity, food habits, oral hygiene and many other local factors can influence the type of flora and their behavior. Hence it is significant to establish the microbial profile of the local population to deliver specific targeted therapy. The culture technique has been the standard method of studying microflora in infectious diseases. If pathogens are uncultivable, detailed characterization of few species and also population, molecular (genomic) analysis can be an excellent tool. In endodontics, many are proteomic analyses but limited genomic studies. But molecular analyses have few inadequacies in envisaging physiology, function and pathogenicity of the disease and host factors. In-spite of limitations of culture technique, they are still excellent choice for their broad range approach and phenotypic characterization. Hence the combination of both the techniques can contribute immensely in microbial analysis.

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Microbial invasion (Routes of micro-organisms ingress)

One of the primary functions of tooth enamel is to exclude these microorganisms from the underlying dentine–pulp complex. As long as the enamel and cementum layers are intact, the pulp and root canal are protected from invasion, but loss of these structures by caries, cracks or trauma opens an avenue for penetration of bacteria through the dentinal tubules [7]. Dental caries is the most common cause of pulp injury. Most authors believe that acid-producing bacteria invade the dentinal tubules and demineralize the tubule walls. Thereafter proteolytic bacteria follow, acting on the organic matrix, which is denuded in the enlarged dentinal tubules. The bacteria in the front of the carious process are the first to reach the pulp. Gram-positive bacteria predominate among the advancing bacteria in a carious process. Some investigators have found exclusively lactobacilli whereas others also report *streptococci* (*S. mitis, S. milleri; now termed S. oralis and S. anginosus*), *propionobacteria*, *Actinomyces*, and some obligatory anaerobic gram-negative and gram positive non sporulating rods. Most of the bacteria in the carious process are non-motile. Therefore bacterial penetration in the dentinal tubules is slow; the acids and other metabolites and toxic products produced by the bacteria diffuse faster. A reaction of the pulp occurs only a few hours after experimental application of bacterial products into dentin cavities [8].

In addition to caries, pathways for the entry of microorganisms into the pulp space include direct pulp exposure (e.g. trauma, and dental procedures), dentinal tubules, lateral/accessory/furcation canals, and anachoresis.

The heterogeneous microflora comprises of 65% bacteria, 30% fungi and 5% of other organism [9, 10]. Lack of microbial specificity affecting the treatment outcome may be predisposed by the dearth of consistent evidence. The ability of the microorganisms to survive the most challenging environment of the root canal is remarkable. The complexity of root canal anatomy, nutrition supply, oxygen tension and microbial interaction makes the root canal environment very unique [11]. The advanced Molecular biology techniques are the forefront of microbial analysis. It provides rapid, sensitive and specific tools for the analysis of DNA, RNA and proteins.

Behavior of Microorganisms in Root canal

Most of the oral microorganisms have the inherent capacity to invade the pulpal space and deep into the dentinal tubules, isthmus, accessory and lateral canals. Studies have contradicted the development of apical periodontitis from necrotic pulp tissue and stagnant fluid in the absence of microorganisms. However, root canal flora comprises of more restricted species compared to oral flora, implying selective pressures playing role in the survival of few microorganism.

Pathogenicity of endodontic microflora

The microflora of infected root canal has the ability to initiate periapical inflammation through combination of species. Prevotella and Porphyromonas resist abscess resolution by increasing the accumulation of leukocytes. In polymicrobial infections, phagocytosis, intracellular and growth factors might protect the host to certain extent. But in mixed cultures, obligate anaerobes can interfere with this defense mechanism of facultative anaerobic partners. Also synergistic lowering of local oxygen concentration by facultative anaerobic bacteria, facilitate the invasion and replication of anaerobic bacteria in polymicrobial infections. Pathogens produce resorption and tissue destruction enzymes and also inactivate human plasma proteins. The host defense mechanisms of immunoglobulins and complement factors, as well as plasma proteinase inhibitors and plasma proteins of the clotting, fibrinolytic and kinin systems play important roles in various phases of the microbial invasion. Data on microbial morphology provides clues for the identification of most microorganisms and

physiological traits are often ambiguous [12, 13].

Bacteriology before Obturation

It is essential to analyze the quality and quantity of bacteria before obturation of the root canal system to evaluate the efficacy of the cleaning and shaping procedure. It not only helps to selfevaluate the productiveness of mechanical instrumentation done but also the antimicrobial efficiency of irrigating solutions used during the procedure. Though this protocol is not clinically feasible during each case of endodontic therapy but it has a definitive academic role while testing various instruments and irrigating solution which are launched new in the dental profession from time to time. In severe situations with persistent infections, this protocol is performed in clinical cases too. It has been observed that an average of 1 to 5 bacterial species have been found in the root canals after completion of cleaning and shaping procedure and the counts were found to be reaching up to 102 to 105 cells *per* canal [14].

It has been observed that the microbes which persist after the chemo-mechanical preparation are most commonly anaerobic rods such as F. nucleatum, Prevotella species, and C. rectus or Gram-positive bacteria such as Streptococci (S. mitis, S. gordonii, S. anginosus, S. sanguinis, and S. oralis), P. micra, Actinomyces species (A. israelii and A. odontolyticus), Propionibacterium species (P. acnes and P. propionicum), P. alactolyticus, Lactobacilli (L. paracasei and L. acidophilus) and E. faecalis [14].

Microbes in Endodontically treated teeth

It is a well-established fact that despite flowing the standard protocol of endodontic treatment, some cases still fail. These failures are due to multiple reasons but the microbiological factors have a significant role to play. From all the cases which report back with pain and infection after the endodntic therapy, it has been observed that E. faecalis is the most commonly found, with prevalence values reaching up to 90%.Other bacteria isolated in similar cases are streptococci P. alactolyticus, P. propionicum, F. alocis, D. pneumosintes, and D. invisus.[3] As far as the fungi are concerned, it is the Candida species that have been most commonly seen to be involved in as many as 18% of the cases. To be more specific, it has been observed that C. albicans is the most commonly detected fungal species in retreatment cases [15].

Beyond the border: Extraradicular infections

Extraradicular infection refers to the infection of the periradicular region. The infection can be either dependent or may be independent of intraradicular infections. While most of these infections are a sequel to the intraradicular ones, apical actinomycosis, caused by Actinomyces species is an example of extraradicular infection independent of the intraradicular infections. Species which have been reported by many studies to be involved in the extrardicular infections include: Actinomyces species (A. israelii, A. naeslundii, A. odontolyticus, A. viscosus), P. acnes, P. propionicum, P. gingivalis, P. intermedia, Prevotella oralis, P. micra, and F. nucleatum [6].

Microbes in endodontic flare-ups

The mid treatment flare ups during endodontic therapy are a night mare for the treating dentist because at times these flare ups exhibit in the form of an acute emergency, expressing itself in the form of pain or swelling or both.[16] The flare ups can be immediate post obturation also and the etiology in both the cases can be mechanical, chemical or microbial injury to the pulp and periradicular tissues. Of all the mentioned factors, bacteriological ones have definitely a major role to play. Chavez de Paz examined root canal microbiota and revealed F. nucleatum to be associated with flare-up pain and swelling. Other microbes isolated with flare-ups were Gram negative obligate anaerobic rods belonging to the genera Prevotella and Prophyromonas (Black pigmented bacteria) [17]. Chavez de Paz suggested that the combination of F. nucleatum, Prevotella spp. and Prophyromonas species may provide a risk factor for endodontic flare-ups by acting in synergy to increase the intensity of periapical inflammatory reaction. In a study done by Sundqvist, *et al*. in necrotic dental pulp, a relationship was established between certain microorganisms and painful teeth. In all cases of flare-up, an anaerobic gram negative rod, Bacteroides melaninogenicus was found. A new bacterial species has also been identified with failed endodontic treatment in two patients with failed endodontic treatment and persistent signs and symptoms. The bacteria were similar to each other and were classified as Actinomyces radicidentis.

Molecular biology in endodontics

The current substantial knowledge of the endodontic ecosystem is by conventional culture technique. It may not provide an accurate data of the microbial load as many organisms fail to endure for identification under routine laboratory conditions.

The phenotypic identification have certain drawbacks namely impossible to culture large number of extant species, non recovery of all viable microbes, need immediate processing, expensive, time consuming (several days to weeks), low specificity, low sensitivity, microbiologist' expertise and need for transport media. They also not favor the culture of all species either due to their specific nutritional requirements and growth factors. The toxicity of the culture, injunctive substances released by other microbes or metabolic dependency can hamper the culture technique. All these factors "underestimated" the endodontic pathogens responsible for pulp and periapical infections. Hence the role of culture as a gold standard in microbial taxonomy is not completely justified.

This has made comprehensive data collection still unreality. This has paved way for culture independent techniques – Molecular biology techniques. The study of DNA, RNA and proteins has opened new avenues for clinical research that revolutionized the identification of new species, understanding micro flora and led to rapid diagnostic tool [18, 19].

Conclusion

The endodontic infections can be symptomatic and can have serious bearing on the integrity of the tooth in the arch. The presence of apical periodontitis is linked to systemic implications like-infective endocarditis, bacteremia etc. The role of microorganism in causing the root canal infections is well established. Contextually, community microbial profiling unveils the pathogens and serves as a rationale for setting the targeted clinical protocols. The dominance of certain species in some locations and geography-related pattern can been confirmed by the analyses. Also the correlation of various parameters from diagnosis to treatment outcome can provide valuable information. Although the incidence of virulence factors are more pronounced in hospital infections, endodontic isolates are exhibiting emergence of bacterial

resistance to conventional regimens used in dental procedures. The different expression profiles of virulence factors can be explained by geographic differences, diet, infection stage and systemic condition.

The culture technique has been standard method of diagnostics in infectious diseases since many centuries. Due to time consumption, low sensitivity and specificity led to the era of molecular analysis. Genomic analysis is rapid, highly specific and sensitive and can directly analyze the sample. The major setback of the technique includes cost, overestimation of flora and inability to assess functional relationship. Hence the combination of both the techniques can expand and further refine the current knowledge regarding the species and communities associated with different clinical conditions. It can witness a huge change in the trend of treatment approaches.

References

1. Cruse WP, Bellizzi R. A historic review of endodontics, 1689–1963, part 1. Jornal of Endodontics. 1980; 6:495-499.
2. Henderson B, Wilson M. Commensal communism and the oral cavity. Journal of Dental Research 1998; 77:1674-1683.
3. Siqueira JF, Rocas IN. Distinctive features of the microbiota associated with different forms of apical periodontitis. J Oral Microbiol, 2009.
4. Figdor D, Sundquist G. A big role for the very small-understanding the endodontic microbial flora. Aust Dent J. 2007; 52(1):38-51.
5. Dudeja PG, Dudeja KK, Srivastva D, Grover S. Microorganisms in periradicular tissues: Do they exist? A perennial controversy. J Oral Maxillofac Pathol. 2015; 19(3):356-363.
6. Siqueira JF, Rocas IN. Diversity of Endodontic Microbiota Revisited. J Dent Res. 2009; 88(11):969-981**.**
7. Slots J, Sabeti M, Simon JH. Herpesviruses in periapical pathosis: An etiopathogenic relationship? Oral Surgery Oral Medicine Oral Pathology Oral Radiology Endododontics. 2003; 96:327-31.
8. Siqueira JF, Janeiro R. Endodontic infections: Concepts, paradigms, and perspectives. Oral Surgery Oral Medicine Oral Pathology Oral Radiology Endodontics 2002; 94:281-93.
9. Pinheiro ET, Gomes BP, Ferraz CC, *et al*. Microorganisms from canals of root-filled teeth with periapical lesions. Int Endod J. 2003; 36(1):1-11.
10. Sjogren U, Figdor D, Persson S, *et al*. Influence of infection at the time of root filling on the outcome of endodontic treatment of teeth with apical periodontitis. Int Endod J. 1997; 30(5):297-306.
11. Peciuliene V, Maneliene R, Balcikonyte E, *et al*. Microorganisms in root canal infections: a review. Stomatologija. 2008; 10(1):4-9.
12. Relman DA. The search for unregognized pathogens. Science. 2009; 284(5418):1308-1310.
13. Chan EC, McLaughlin R. Taxonomy and virulence of oral spirochetes. Oral Microbiol Immunol. 2000; 15(1):1-9.
14. Vianna ME, Horz HP, Gomes BP, *et al*. Microarrays complement culture methods for identification of bacteria in endodontic infections. Oral Microbiol Immunol. 2005; 20(4):253-258.
15. Peciuliene V, Reynaud AH, Balciuniene I, Haapasalo M. Isolation of yeast and enteric bacteria in root filled teeth endodontic with chronic apical periodontitis. Int Endod J. 2001; 34(6):429-434.
16. Ingle JI, Backland LK, Baumgartner JC. Endodontics 6th ed. Hamilton: BC Decker Inc, 2008.
17. Sipaviciute E, Maneliene R. Pain and flare-up after endodontic treatment procedures. Stomatologija. 2014; 16(1):25-30.
18. Munson MA, Pitt-Ford T, Chong B, *et al*. Molecular and cultural analysis of the microflora associated with endodontic infections. J Dent Res. 2002; 81(11):761-766.
19. Mandlik J. Microbial identification in endodontic infections with an emphasis on molecular diagnostic methods: a review. IIOABJ. 2016; 7(6):60-70.