

Molecular Characterization of Microorganisms Involved in Endodontic Infection

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Abstract

Periodontitis and poor dental hygiene may be related, according to recent studies. Microorganisms are the primary cause of the majority of endodontic infections. Apical periodontitis is fundamentally an inflammatory, infectious sickness with a microbial origin, and a root canal infection is its biggest factor. The lack of microbiological specificity influencing the treatment's success could be caused by the demise of reliable data. Most oral bacteria have the innate ability to enter the pulpal space and deeply into the dentinal tubules.

Both historically and currently, herbal treatments are used all over the world. In recent decades, there have been more studies conducted on this alternative therapy technique. Cleaning and sanitizing of root canals and intracanal medications used in between sessions are among the endodontic uses of medicinal plants. This study's purpose was to compare the effectiveness of ginger extract, a new natural irrigant, to the conventional irrigant NaOCl in treating two groups of infected root canals.

Keywords: Ginger; Culture; Molecular; Endodontic Infection; Sodium hypochlorite; Irrigation.

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Introduction:

The oral microbiota, which significantly affects dental health, also has a significant impact on general health. The mouth contains a variety of bacterial species, some of which are beneficial to health and others of which may be harmful (Martino et al., 2022).

Operative dental therapy is addressed in endodontic treatment. It is performed when a previous root canal treatment fails, the dental pulp develops an irreversible inflammatory condition, or it becomes necrotic. Numerous reasons, like dental trauma, ongoing

irritation, and deep cavities, can lead to pulp inflammation (Raducka et al., 2023)

It is widely accepted that microorganisms are the primary cause of most endodontic infections. Looking beyond the symptoms and researching the etiopathogenesis of disease processes is a major focus of modern medicine (Siqueira Jr & Rôças, 2019). Apical periodontitis, which is essentially an infectious and inflammatory disease with a microbial etiology, is primarily brought on by a root canal infection. There is currently no method that can ensure the complete

cleaning and sealing of the whole canal system of a dead tooth because the healthy tooth's root is formed of a highly porous substance that constantly exudes fluids when the tooth is alive (figure 1). Furthermore, root canal therapy can only minimize the amount of infection in the dead tooth since it is impossible to completely eradicate an infection once it has taken hold in a tooth. The primary emphasis of endodontic practice is the extraction of infected tooth pulp and its replacement with filling material, a process known as root canal therapy. Numerous pathways exist for bacteria to enter endodontic tissues, entering the bloodstream through damaged tissues, periodontal membrane, exposed cavities, dental tubules, and contamination from infected tissues. When bacteria or their byproducts are able to activate the periapical tissues, the infection propagates apically and causes apical periodontitis. This occurs when the root canal becomes infected coronally.

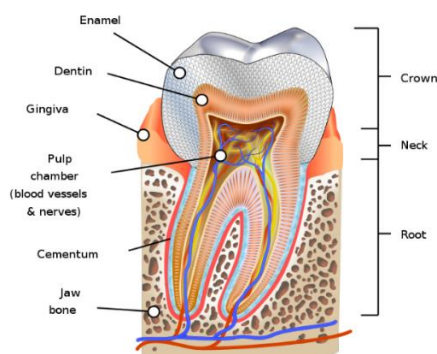


Figure 1: Diagram showing healthy tooth

Depending on a number of bacterial and host-related factors, endodontic infections may result in either chronic (asymptomatic) or acute (symptomatic) apical periodontitis (Figure 2) (Madhu & Mathew, 2018; MÖLLER *et al.*, 1981) Additionally, it can be categorized as an extraradicular infection or an intraradicular infection depending on the anatomic site.

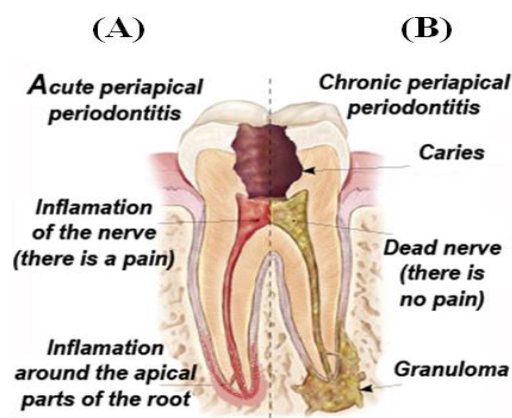


Figure 2: Acute (symptomatic; A) and chronic (asymptomatic; B) endodontic infection

cultivable bacterial species have been linked to apical periodontitis by molecular approaches, which have also

Behavior of Microorganisms in Root Canal. The periradicular tissues can be seriously harmed by bacteria that have invaded the apical root canal. The periradicular tissues are eventually affected by an inflammatory response brought on by chemicals generated from the bacterial biofilm in the canal, which finally destroys the periodontal ligament and bone. The majority of oral bacteria have the innate ability to penetrate the dentinal tubules, isthmus, and lateral canals as well as the pulpal area.

Molecular Biology in Endodontics

Standard culture techniques account for a large portion of existing knowledge regarding the endodontic ecology, but this method may not be able to accurately measure the microbial burden because many organisms cannot withstand routine laboratory conditions for identification. There are some drawbacks to phenotypic identification, including the inability to culture many extant species, the non-recovery of all viable microbes, the need for immediate processing, the cost, the length of time required (a few days to weeks), and the requirement for microbiologist expertise. The culture process may be hampered by the culture's toxicity, dangerous chemicals secreted by other bacteria, or metabolic dependence (Jhajharia, 2019). The endodontic microorganisms that cause pulp and periapical infections were "underestimated" by all of these parameters. Thus, the use of culture as the sole criterion for microbial taxonomy is not entirely warranted.

Because of this, full data collecting is still impossible. This has made way for molecular biology approaches that are independent of culture. Conrads *et al.*, (1997) introduced the first Molecular Biology strategy in endodontics, among of the initial strategies that Mullis *et al.*, (1992) came up with was Polymerase chain reaction (PCR). By using PCR, it has been feasible to isolate previously unidentified species from endodontic infection (Siqueira Jr & Rôças, 2017a). The 16S rRNA gene (or 16S rDNA) has been the most frequently used of the many genes chosen as targets for bacterial identification because it is universally distributed among bacteria, long enough to be highly informative and short enough to be easily sequenced, has conserved and variable regions, and provides reliability for inferring phylogenetic relationships (Yarza *et al.*, 2014). A group of species believed to be crucial in the etiology of apical periodontitis have been identified through culture research (first generation) (Siqueira Jr & Rôças, 2013). Furthermore, results from culture-based approaches have not only been validated, but they have also been greatly expanded upon by results from culture-independent molecular biology methods. Many

can destroy soft tissues and clothing because it is a bleaching agent. If NaOCl is accidentally introduced

uncovered new putative endodontic pathogens (Siqueira Jr & Rôças, 2017b). In various forms of endodontic infections, more than 400 distinct bacterial species have previously been identified. Comparatively, just 32% of these were discovered by culture studies alone, whereas roughly 45% of them were reported only by molecular biology investigations. By utilising both culture and molecular investigations, 23% of the overall bacterial species richness has been identified (Siqueira JR & Rôças, 2014). As a result, it is clear that molecular techniques have modified the endodontic microbiota (Jhajharia *et al.*, 2015).

Irrigating solutions for endodontic infections

In order to clean and lubricate the root canal, remove debris from the canal system, and dissolve both organic and inorganic tissues, endodontic treatment has demonstrated the importance of employing root canal irrigant solutions (Haapasalo *et al.*, 2010). A root canal irrigant's ideal properties include: (a) having a broad antimicrobial spectrum; (b) the ability to dissolve proteins and necrotic tissue; (c) prevent the formation of a smear layer during instrumentation; (d) the presence of low surface tension to reach areas that are inaccessible to the tools (dentin tubules). Also, it should be reasonably priced and simple to use (Karpagam & Raj, 2018).

Sodium hypochlorite (NaOCl)

Root canal irrigation with sodium hypochlorite solutions (in concentrations ranging from 1 to 5.25%) is now a method that is routinely used (Verma *et al.*, 2019). It functions as an efficient antibacterial and tissue-dissolving endodontic irrigant. Its low viscosity makes it simple to introduce into the canal architecture and is reacting with amino acids and fatty acids in tooth pulp to cause the liquefaction of organic tissue (Spencer *et al.*, 2007). It has drawbacks, the main ones being the toxicity of its action to vital tissues and metal corrosion in dental applications. It also affects all living tissues with the exception of keratinized epithelia. It has a very terrible flavor and is highly alkaline and hypertonic. The predicted shelf life can only be attained by following the right storage techniques. By restricting the usage of hypochlorite to the pulp chamber and root canal, endodontists can avoid the majority of these drawbacks. Rubber dams are a critical part of irrigation techniques. As an endodontic irrigant, sodium hypochlorite is not used in any commonly agreed-upon concentration. Depending on the dilution and storage procedures followed by particular practitioners, it is utilized down to 5.25% (Clarkson & Moule, 1998) hypochlorite's antibacterial and tissue-dissolving effects increase, but so does its toxicity. Accidental leakage of this substance

outside of the root canal system, significant soft tissue or nerve damage, and even airway compromise, may occur. So that many studies have investigated the efficacy and potential uses of novel and all-natural materials for root canal disinfection (Murray *et al.*, 2008). As phytotherapy has developed as a science, there is increased interest in analyzing plant extracts that might have therapeutic uses in dentistry.

Ginger as a model for the phytotherapeutic irrigant

An efficient complement to modern medicine is herbal medicine. Numerous studies indicate that pure phytochemicals, essential oils, and plant extracts have the potential to be developed into medications that are used to treat or prevent periodontitis. More studies on the safety and effectiveness of these products are required to determine whether they have medicinal value, either on their own or in conjunction with traditional treatment options, which can help lower the global burden of oral diseases, despite the positive results of the many clinical trials for these products.

(Mosaddad *et al.*, 2023)

Ginger (*Zingiber officinale*) is commonly used to cure a number of illnesses and is known as "The Great Medicament". This tropical rhizome is known to be a traditional gastric tonic and is advised for gastrointestinal discomfort, nausea, vomiting, diarrhea, coughs, and colds. Moreover, it acts as an anti-inflammatory and cures rheumatologic diseases. So, it should come as no surprise that Chinese fishermen used ginger to both prevent and cure seasickness. It contains more than 20 active ingredients, such as derivatives of gingerol and curcumin (Tan & Vanitha, 2004). Rhizome's antimicrobial action has been extensively demonstrated *in vitro* and that is related to its chemical components, - Oleo-resin: (Gingerols, shogaols, paradols, zingerone, gingerenone-A, and 6-dehydrogingerdione) - Essential oil: (β -Bisabolene and zingiberene, zingiberol, zingiberenol, ar-curcumene, β -sesquiphellandrene, β -sesquiphellandrol (cis and trans), phellandrene, camphene, geraniol, neral, linalool, d-nerol) - Others: (Starch, lipids, proteins and amino acids) (Sharifi-Rad *et al.*, 2017). A number of bacteria and fungi were discovered to grow less quickly when exposed to the components contained in the rhizomes of *Z. officinale*. Ginger has antibacterial properties that are effective against *S. sanguinis* and *Streptococcus mutans* (Pushpalatha *et al.*, 2022a). Besides that, compared to commercially available antibiotics, it displays stronger antibacterial action against *Staphylococcus aureus* and *Streptococcus pyogenes* (Pushpalatha *et al.*, 2022b). Although ginger has a wide range of pharmacotherapeutic benefits, it has been prescribed to

handle dental conditions. Ginger has been demonstrated to have an inhibitory effect on the growth of microorganisms that cause tooth decay in terms of oral health. In addition to serving as a dental anesthetic, it can help with dentine remineralization. Teeth can be kept fresh, free of tartar, and cavities with the use of ginger toothpaste and tooth powder.

Conclusion

In conclusion, it can be said that ginger extract functions as a substitute for other naturally occurring extracts that are readily available for irrigation in regular dental clinical practice. The 16S rRNA gene sequence comparison is one of the best techniques for identifying prokaryotes. In this investigation, the traditional identification of the isolated isolates was largely corroborated by the 16S rRNA sequence analysis.

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