

Role of LSTR in Root Canal Therapy Particularly against *Enterococcus faecalis* and *Enterococcus Faecium*

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ABSTRACT

Endodontic treatment success generally depends on the microbial suppression in the root canal and periapical region. Endodontic instrumentation alone cannot achieve a sterile condition during RCT. Sterilization of bacteria in root canal system is one of the prominent problems. Occasionally Some bacteria could remain in root canal even after using conventional medicaments. Evidence suggested that *Enterococcus faecalis* (*E. faecalis*) and *Enterococcus faecium* caused substantial root canal infections. It is important to evaluate root canal obturation because inadequate obturation may not only produce areas where bacteria may remain and survive, but also provides routes through which bacteria may migrate to the periradicular Tissues. Radiographs have often been used to examine root canal systems and evaluate obturations of the root canal, partly because of its non-destructive nature. Consequently, elimination of such organism is important to achieve Endodontic treatment success. In addition, *E. faecalis* has been frequently found in re-infected root canal-treated teeth in prevalence values ranging from 30% to 90% of the cases and re-infected root canal-treated teeth are about nine times more likely to harbor *E. faecalis* than cases of primary infections.

Keywords

Endodontic treatment, Re-RCT, *Enterococcus faecalis* and *Enterococcus faecium*.

Materials and Methods

Many studies have been directed towards finding an effective way to eradicate and/or prevent *Enterococcus faecalis* and *Enterococcus faecium* from gaining access to the root canal space. *Enterococcus faecalis* and *Enterococcus faecium* can gain entry into the root canal system during treatment, between appointments, or even after the treatment has been completed. Antibiotic medium 3 was used for testing drug resistance.

Antibiotic concentrations (micrograms per milliliter) used in selective plates were as follows: ciprofloxacin, minocycline and metronidazole (0.5 mg of each) MP with Bile Esculin Agar (BEA). In-vitro *Enterococcus faecalis* and *Enterococcus faecium* both were culturing under LSTR(3MIX-MP) to determine the Inhibitions zones of bacterial colony.

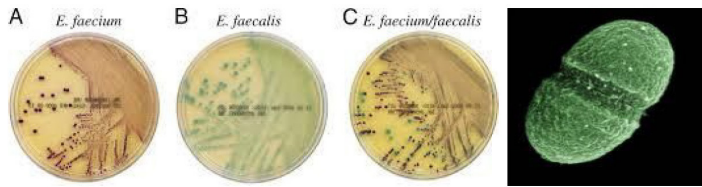
Discussion

Enterococcus faecalis and *Enterococcus faecium* Characteristics and Strains

Enterococci are gram positive cocci that can occur singly, in pairs, or as short chains. They are facultative anaerobes, possessing the ability to grow in the presence or absence of oxygen, Enterococcus species live in vast quantities colony-forming units (cfu per gram of feces] in the human intestinal lumen and under most circumstances cause no harm to their hosts. They are also present in human female genital tracts and the oral cavity in lesser numbers. They catabolize a variety of energy sources including carbohydrates, glycerol, lactate, malate, citrate, arginine, agmatine, and many keto acids Enterococci survive very harsh environments including extreme alkaline pH (9.6) and salt concentrations. They resist bile salts, detergents, heavy metals, ethanol, azide, and desiccation. They can grow in the range of 10 to 45°C and survive at a temperature of 60°C for 30 min.

There are currently 23 Enterococci Species and these are divided into five groups based on their interaction with mannitol, sorbose,

and arginine.



Categorization of *Enterococcus* species and two physiologically related gram-positive cocci based on phenotypic characteristics*

Group	Species
Group I (+) acid formation in mannitol broth (+) acid formation in sorbose broth (-) arginine hydrolysis	<i>E. avium</i> <i>E. gilvus</i> <i>E. malodoratus</i> <i>E. pallens</i> <i>E. pseudoavium</i> <i>E. raffinosus</i> <i>E. saccharolyticus</i>
Group II (+) acid formation in mannitol broth (-) acid formation in sorbose broth (+) arginine hydrolysis	<i>E. faecalis</i> <i>E. faecium</i> <i>E. casseliflavus</i> <i>E. gallinarum</i> <i>E. mundtii</i> <i>Lactococcus sp.</i>
Group III (-) acid formation in mannitol broth (-) acid formation in sorbose broth (+) arginine hydrolysis	<i>E. dispar</i> <i>E. durans</i> <i>E. hirae</i> <i>E. porcinus</i> (<i>E. villorum</i>) <i>E. ratti</i>
Group IV (-) acid formation in mannitol broth (-) acid formation in sorbose broth (-) arginine hydrolysis	<i>E. asini</i> <i>E. cecorum</i> <i>E. sulfureus</i>
Group V (+) acid formation in mannitol broth (-) acid formation in sorbose broth (-) arginine hydrolysis	<i>E. columbae</i> <i>Vagococcus sp.</i>

Collected from Teixeira and Facklam

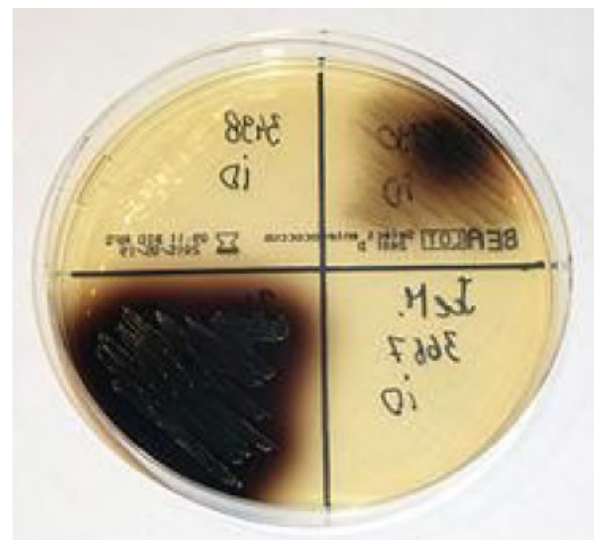
Survival and virulence factors

- Endures prolonged periods of nutritional deprivation
- Binds to dentin and proficiently invades dentinal tubules
- Alters host responses
- Suppresses the action of lymphocytes
- Possesses lytic enzymes, cytolysin, aggregation substance, pheromones, and lipoteichoic acid
- Utilizes serum as a nutritional source
- Resists intracanal medicaments (e.g. calcium hydroxide)
 - o Maintains pH homeostasis
 - o Properties of dentin lessen the effect of calcium hydroxide
- Competes with other cells
- Forms a biofilm

Study Technique or procedure

The strains were separately inoculated into tubes containing 2ml of sterile 0.9% normal saline Solution. Microorganisms suspension were inoculated into Bile Esculin Agar (BEA) media.

Enterococcus faecalis and *Enterococcus faecium* ATCC29212 and antibiotics disks were made from LSTR ciprofloxacin, minocycline and metronidazole (0.5mg of each) and were placed in BEA Media.



Enterococcus faecalis and *Enterococcus faecium* colonies (black) growing on BEA.

Growth Conditions Temperature:
37°C
Atmosphere:
Aerobic

Enterococcus faecalis
(ATCC® 29212™)



Results

Studies indicate that the prevalence of *Enterococcus faecalis* and *Enterococcus faecium* low in primary endodontic infections and high in persistent infections which inhibits by using LSTR. (3MIX-MP).

In vitro antibacterial efficiency of a mixture of ciprofloxacin, metronidazole, and minocycline (3Mix antibiotics), against oral bacteria of children was assessed by Sato et al. The antibiotic combinations were observed to be effective against both carious and endodontic lesions *in vitro*. Hoshino et al. determined that 25 µg each/ml of ciprofloxacin, minocycline, and metronidazole antibiotic mixture to be effective in sterilizing the infected root dentin *in vitro*. Sato et al. studied the ability of a mixture of ciprofloxacin, minocycline and metronidazole (0.5mg of each) in an *in vitro* study to eliminate experimental infection in deep layers of root dentin by *E. coli*. In other, *in vitro* study, the minimal inhibitory combination for ciprofloxacin and minocycline against *E. faecalis* and *E. faecium* were found to be 5 and 20 µg respectively and metronidazole was reported to have no inhibitory effect. However, as the combination (100µg each /ml) they inhibited the growth of every strain completely. Thus this suggests 3mix may be effective in persistent endodontic infection. Resistant bacteria *E. faecalis* may result in Root Canal Treatment (RCT) failure. It may impregnate in tiny dentinal tubules. It usually not killed by most of the medicament or any single antibiotic. The present study shows that LSTR-3mix MP therapy has a significant antibacterial effect on *E. faecalis* when compared to a single antibiotic. Two antibiotics of LSTR- 3mix showed susceptibility with *E. faecalis* except for metronidazole. It was also useful in pulpotomy and pulpectomy for primary or premature permanent teeth.

Conclusion

Endodontic failures would then routinely undergo re-treatment based on the attending clinician's best judgment. A possible alternative treatment for such cases is Lesion Sterilization and Tissue Repair (LSTR) therapy. The basic concept of this therapy is the elimination of the cause of infection, in this case, bacteria, by introducing the appropriate antibacterials into the tooth thereby resulting in disinfection of the lesion and resolution of inflammation. The main component of LSTR therapy is a macrogol-propylene glycol-antibiotic mixture (3Mix-MP) enterococcus faecalis & faecium are the species most frequently isolated from failed endodontic treatments because it can survive under stress conditions imposed by root canal treatment.

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