

Evaluation of a Ciprofloxacin and Metronidazole Encapsulated Biomimetic Nanomatrix Gel on *Enterococcus faecalis* and *Treponema denticola*

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Statement of Purpose

Over 70 percent of Americans suffer from an endodontic infection at least once in their lifetime [1]. Endodontic infections are mainly caused by vegetation of bacterial biofilm in the root canal and can potentially extend to underlying periodontal structures, which require massive antibiotic treatment. The use of oral antibiotics has been concerning as improper dosage application, duration of therapy, and nonspecific prescription lead to antibacterial resistance [2]. For instance, United States dentists prescribe between 200 and 300 million antibiotics each year, and about 10% of prescriptions create a high potential for development of antibiotic resistance [3]. The triple antibiotic mixture, ciprofloxacin (CF), metronidazole (MN), and minocycline (MC), has been developed and widely used in root canal treatment and is expected to induce root end closure (conventional apexification). However, unfavorable outcomes of crown discoloration, weakened cervical root structure, and insufficient root dentin thickness have been raised as limitations of the conventional root canal treatment. In order to improve the limitations from the conventional root canal therapy, an efficient root canal disinfection using two antibiotics (CF and MN), excluding MC, encapsulated within an injectable self-assembled biomimetic nanomatrix gel was developed. In order to improve on the limitations of triple mixture therapy, first, minimal bactericidal concentration (MBC) of each of the CF and MN was tested to determine bactericidal activity against 14 endodontic species. Secondly, CF and MN encapsulated within the injectable biomimetic nanomatrix gel were separately tested against *E. faecalis* and *T. denticola*.

Research Design and Methods

To evaluate a nanomatrix gel of peptide amphiphiles (PAs) short and YK (9 PA-YIGSR: 1PA-KKKKK) was prepared in a one to one ratio and encapsulated with each CF and MN at varying concentrations. The bactericidal effects of each antibiotic encapsulated biomimetic nanomatrix gel were tested separately on both *Enterococcus faecalis* and *Treponema denticola*. *E. faecalis* is known as the one of the most common and recurrent bacterial strains in endodontic infections and *T. denticola* is found in root canal infections and often used to evaluate endodontic disinfectants. The bacterial densities from the broth suspensions were determined by the McFarland standard with optical density (OD₆₀₀) values between 0.1 and 0.5.

Results

Concentrations (0.1-0.2 µg/mL) of each CF and MN encapsulated in biomimetic nanomatrix gels well demonstrated antibacterial effect against the two endodontic strains. In *E. faecalis* and *T. denticola*, 0.0625 µg/mL of CF killed the bacteria, while 0.0625 µg/mL of

MN presented bactericidal effects. Thus, the two antibiotics delivered separately using biomimetic nanomatrix gels may provide an alternative root canal treatment to the current triple antibiotics treatment avoiding such limitations and inducing a natural healing environment owing to the biomimetic nanomatrix gel.

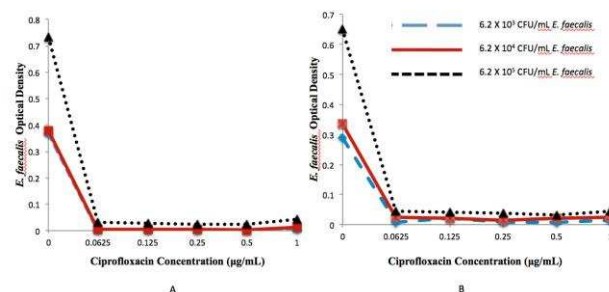


Figure 1. The Effect of ciprofloxacin (CF) against *E. faecalis*. Antibacterial effect of CF was measured in varying concentrations of 0 µg/mL, 0.0625 µg/mL, 0.125 µg/mL, 0.25 µg/mL, 0.5 µg/mL and 1 µg/mL on *E. faecalis* in concentrations of 6.2 X 10³ CFU/mL, 6.2 X 10⁴ CFU/mL and 6.2 X 10⁵ CFU/mL. (A) Without nanomatrix gel, (B) With antibiotic encapsulated in nanomatrix gel

Conclusion

Concentrations (0.0625-0.25 µg/mL) of each CF and MN encapsulated in the biomimetic nanomatrix gel demonstrated competitive antibacterial effects with controlled release on *E. faecalis* and *T. denticola*; both of these antibiotics combined in a 1:1 ratio may provide promising results in future studies. The results suggest this innovative antibiotics delivery system could be an effective alternative treatment over the conventional method for endodontic treatments.

References: [1] Center of Disease Control. Division of Oral Health. 2013.[2] Sweeney LC, Dave J, Chambers PA, Heritage J. Antibiotic resistance in general dental practice--a cause for concern? The Journal of antimicrobial chemotherapy. 2004;53:567-76.[3] Harrison L. Dentists Overuse Antibiotics in Children. 2012.

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