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Cleaning Efficacy of Mtwo and BioRaCe Rotary Systems: A Microbiological Evaluation

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Abstract

Background and Aim: Effective cleaning and shaping of the root canal system is an essential part of a successful root canal treatment and is considered as one of the basic features of rotary instruments. The present study aimed to compare the efficacy of two types of nickel-titanium rotary files namely BioRaCe and Mtwo, in reduction of *Enterococcus faecalis (E. faecalis)* count in the root canal system.

Materials and Methods: In this in vitro study, 50 extracted human single-rooted teeth were assigned to two groups plus a positive and a negative control group (20 samples in each group). First, the root canals were sterilized and were then inoculated with *E. faecalis* (ATCC: 11700). Subsequently, all the root canals were instrumented with BioRaCe and Mtwo nickel-titanium rotary instruments. Then, samples were collected from inside the root canals and cultured in a special medium to evaluate the remaining colonies. The Chi-square goodness of fit test was used to assess the difference in bacterial colony count after root canal preparation with the two rotary systems (alpha=0.05).

Results: Complete bacterial growth was observed in both the BioRaCe and Mtwo groups. There was no statistically significant difference between the two groups regarding bacterial elimination from the root canal system (P>0.05).

Conclusion: Under the limitations of the current study, the two rotary systems exhibited no superiority over each other in terms of their ability to eliminate microorganisms from the root canal system.

Key Words: *Enterococcus Faecalis*; Root Canal Therapy; Root Canal Preparation

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Introduction

Success of endodontic treatment depends on prevention and elimination of root canal system infection. This goal can be achieved by proper cleaning, shaping, and sealing of the root canal system [1]. Mechanical preparation diminishes bacteria in the dentinal tubules, enabling deeper penetration of irrigants [2]. Although many materials have been suggested for washing and disinfection of the root canal of permanent teeth, developmental, anatomical, and physiological differences between primary and permanent teeth cause some differences in the criteria for

dental materials and equipment used for pulpectomy [3].

Studies on teeth with failed endodontic treatment have often reported the presence of *Enterococcus faecalis* (*E. faecalis*), a facultative Gram-positive microorganism, in persistent or secondary infections [4]. High resistance of *E. faecalis* may be due to its invasion to dentinal tubules, adaptation to unfavorable conditions inside the root canal, and resistance to detergents and intracanal medicaments. Several strategies have been proposed to remove *E. faecalis* from the root canal system [5, 6].

Mtwo rotary instruments (VDW, Munich, Germany) with an S-shaped cross-section have two nearly perpendicular cutting edges that increase flexibility. Studies have revealed that this system provides centrality even in curved root canals [7-10].

However, by increasing the apical preparation size, in addition to eliminating more bacteria from the dentinal tubules, the irrigating syringe can penetrate deeper into the root canal system, improving the flow of the irrigant and its efficacy [8]. For this purpose, BioRaCe rotary system (FKG Dentaire, La-Chaux-de Fonds, Switzerland) was introduced to the market. In addition to features similar to those of RaCe, which include an electro-polished surface, sharp cutting edges, and a noncutting safe tip and alternating cutting edge, this system provides the benefits of both the crown-down technique and apical enlargement with fewer files [9, 11, 12].

Although previous studies have reported comparable shaping ability and safe dentin removal by both Mtwo and BioRaCe systems [9, 13], no similar study is available comparing the efficacy of these two rotary systems in decreasing bacterial count in the root canal system. Therefore, the current study compared the efficacy of Mtwo and BioRaCe nickel-titanium rotary instruments in reducing *E. faecalis* colonies in the root canal system.

Materials and Methods

Sample preparation:

This in vitro study was conducted on 50 mature single-canal mandibular premolar teeth. The sample size was calculated according to a previous study [14]. The study protocol was approved by the ethics committee of Tabriz University of Medical Sciences (ethical code: TBZMED.REC.1394.466). The teeth had been freshly extracted for orthodontic or periodontal reasons, and had no caries, cracks or endodontic treatment.

The root canals were checked with a #15 K-file (Mani Inc. Tachigi-ken, Japan) to ensure apical patency and no severe curvature. Then, the teeth were debrided and stored in saline. The roots were cut from the cementoenamel junction with a diamond disc under water coolant to have a 13-mm root length.

Initial coronal enlargement was performed with #2 and #3 Gates-Glidden drills. Then the root canals were prepared to the working length with a #20 K-file to remove the smear layer. The root canals were irrigated with 17% EDTA for 10 minutes and 5.25% NaOCl solution for 10 minutes, followed by irrigation with 5 mL of saline after using each solution to eliminate the remaining chemical agents. All the specimens were then separately placed in small test tubes containing brain-heart infusion broth and autoclaved for 20 minutes at 21°C and 15 psi pressure until the turbidity of the medium was faded away to ensure sterility [15].

Five samples were assigned to the positive control group with no instrumentation, and five were assigned to the negative control group without bacterial inoculation [15].

Bacterial inoculation:

E. faecalis (ATCC: 11700), obtained from Tabriz University of Medical Sciences, was cultured at 37°C in an aqueous agar medium for 48 hours. Then, 0.5 mL of *E. faecalis* bacterial suspension with a concentration of 1.5×10^8 CFUs/mL was injected into the root canals

except for the negative control group. Finally, the samples were incubated at 37°C under aerobic conditions for 72 hours [16, 17].

Cleaning and shaping:

The samples were divided into two experimental groups (n=20). To confirm *E. faecalis* contamination after initial root canal preparation, dentin debris was randomly collected from two root canals in each group under aseptic conditions and cultured in bile esculin agar for 48 hours at 37°C. This specific culture medium exhibited turbidity in presence of *E. faecalis* [18]. In the first group, root canal preparation was performed using BioRaCe rotary system by the crown-down technique, including BR0 (tip size no.25, taper 0.08) for coronal preparation followed by BR1 (tip size no. 15, taper 0.05), BR2 (tip size no. 25, taper 0.04), BR3 (tip size no.25, taper 0.06), BR4 (tip size no 35, taper 0.04), and BR5 (tip size no.40, taper 0.04).

The Mtwo rotary system was used for root canal preparation in the second group using the following sequence: (10, 4%), (15, 5%), (20, 6%), (25, 6%), (30, 5%), (35, 4%), and (40, 4%). All instruments were used to the working length as for the single length technique. After using each file, the root canals were irrigated with 2 mL of 2.5% sodium hypochlorite.

Bacterial culture:

After cleaning and shaping, and irrigation with 2 cc of 2.5% NaCl, dentin chips were collected from the root canals using a #1 peeso reamer and cultured in bile esculin agar medium. Bacterial colonies were counted after 24 hours of incubation. Presence of over 100,000 colonies was considered as complete bacterial growth, presence of <100,000 colonies was considered as relative growth, and presence of no colonies was considered as no growth [13, 19].

Statistical analysis:

The Kolmogorov-Smirnov test was used to assess the normality of data distribution. The Chi-square goodness of fit test was used to compare the colony count between the groups after preparation. The positive and negative control samples were not combined with the study samples, and no statistical analysis was carried out for them. SPSS version 22.0 (Chicago, IL, USA) was used for data analysis. P<0.05 was considered statistically significant.

Results

All samples in the positive control group showed bacterial growth and complete turbidity of the culture medium (Figure 1). However, no bacterial growth and no turbidity of the medium were seen in the negative control group, confirming the accuracy of the experiment (Figure 2). In the Mtwo rotary file group, the specimens showed similar results, and 19 out of 20 specimens exhibited complete bacterial growth, and one sample showed relative bacterial growth (less than 100,000 colonies were counted) (Figure 3). In the BioRaCe rotary group, complete bacterial growth was evident in all 20 specimens after 48 hours (Figure 4).

Based on the results, there was no significant difference in bacterial count of the root canal system after cleaning and shaping with Mtwo and BioRaCe rotary systems (P>0.05, Table 1).

Table 1. Frequency distribution of *E. faecalis* colony count in the two rotary groups

Group	Number (percentage) of samples without bacterial growth (<100.000 colonies)	Number (percentage) of samples with bacterial growth (>100.000 colonies)	P-value*
BioRaCe system	Zero (zero)	20 (100)	0.962
Mtwo system	Zero (zero)	20 (100)	

* Chi-square goodness of fit test

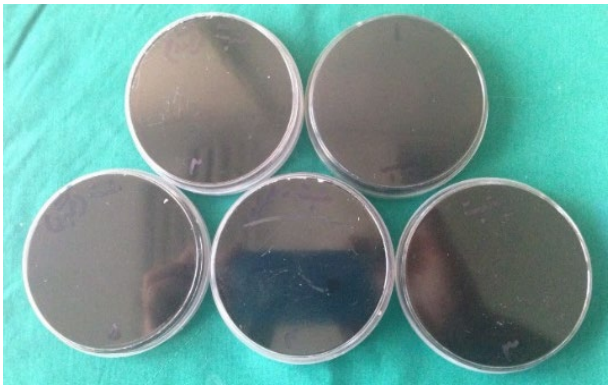

Figure 1. Positive control group. Bacterial growth caused complete turbidity of the culture medium.

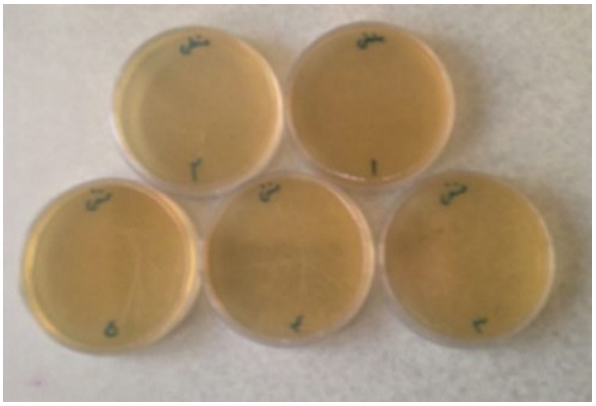

Figure 2. Negative control group. No change in ambient color indicates no bacterial growth.

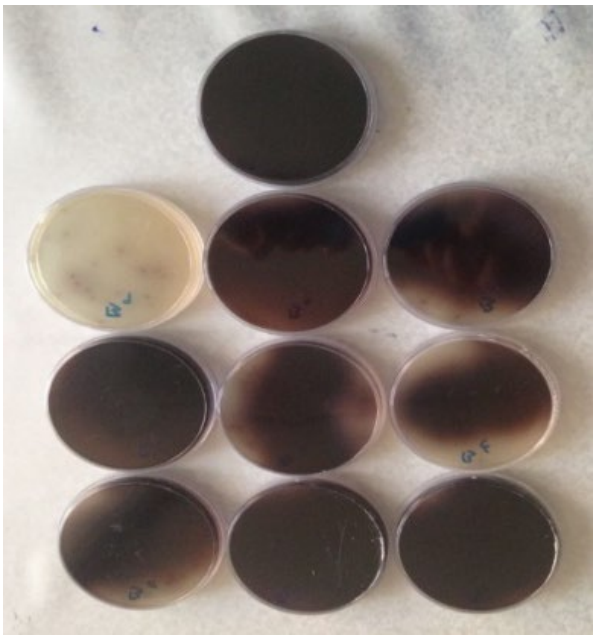

Figure 3. Plates related to the Mtwo rotary file group (complete growth of bacteria and change in color of the medium)

Figure 4. Plates related to the BioRaCe rotary file group (complete growth of bacteria and change in color of the medium)

Discussion

This study compared root canal debridement with Mtwo and BioRaCe nickel-titanium rotary files from a microbiological point of view. The results revealed that the two systems had no superiority over each other in terms of bacterial elimination.

Apical periodontitis is mainly a biofilm-induced condition, and removing the biofilm is the key to a successful endodontic treatment. *E. faecalis* was selected for this microbiological study because of its clinical importance, biofilm formation ability, and deep penetration into dentinal tubules. It is often involved in persistent endodontic infections and postoperative flare-ups [20].

Due to their flexibility and efficient cutting edges, Mtwo and BioRaCe systems have been shown to remove a safe amount of dentin and preserve the root canal centrality [2, 8]. The BioRaCe system is known for its focus on apical preparation. The apical files and alternating cutting edges of BioRaCe files result in proper apical enlargement. Despite a slight change in

the original root canal morphology that is not clinically important [21], it has shown good clinical antibacterial effectiveness and high success rate [22, 23]. On the other hand, the Mtwo system effectively prepares the root canals by better preserving the tooth structure because of its higher flexibility and S-shaped design [2, 24].

To minimize anatomical variations, we selected single-canal mandibular premolars with almost similar dimensions to standardize the samples. The incubation period for bacterial contamination was 72 hours under anaerobic conditions, and initial contamination was checked to be homogenous.

The colony counting method was used in the present study. However, this method has some specific shortcomings. Furthermore, effective sampling is highly important in root canal microbiological studies. Collecting debris with #1 peeso reamer, as used in the present study, from the area where the mechanical instrument's action is most effective leads to errors. Application of ultrasonic devices for sampling from less accessible areas would be more accurate.

The cleaning efficacy of BioRaCe was expected to be superior due to more effective preparation of the apical region, but the results proved otherwise. The results of the present study are consistent with those of Saluja et al. [25], and Coldero et al. [14], who compared different apical preparation sizes and reported no significant difference. Otero et al. [26] concluded that the WaveOne system with the aid of passive ultrasonic irrigation is superior to the multi-file systems like ProTaper Gold. This controversy can be attributed to the ultrasonic activation of irrigant. There is no similar study comparing BioRaCe and Mtwo systems in terms of microbiological cleaning ability. A previous study compared SAF and the ProTaper rotary systems and showed that SAF was more efficient in reducing *Enterococcus* colony count [27]. Matos Neto et al. [28] compared three systems of ProTaper, K3, and HeroShaper. They found no significant difference in reduction of *E. faecalis* colony count. The results of the above-mentioned studies were consistent with the present results. In a similar study, Schäfer et al. [10] demonstrated that the RaCe rotary system created lower amount of residual debris than Mtwo and K3. Gorduysus et al. [29] compared the elimination of *E. faecalis* from the root canal system by three types of rotary files namely ProTaper, K3, and HeroShaper, and reported that all these methods effectively decreased *E. faecalis* bacterial colonies, but the reduction in bacterial colonies in the ProTaper group was significantly less than that in the other two methods. However, it should be noted that in their study, the mesial root canals of mandibular molars were evaluated, which have a more complex anatomical diversity than single-canal teeth, causing more errors.

It has been noted that regardless of the type of rotary system, complete elimination of bacteria from the root canal system is not achievable, due to the bacteria taking refuge in inaccessible areas [18, 30]. Therefore, it is noteworthy that achieving sufficient taper in the root canal helps penetration of irrigants to the apical third while maintaining the integrity of the root canal structure with no need for excessive apical enlargement [18, 25, 31]. Furthermore, a lower number of bacteria within the dentinal tubules might not interfere with periapical healing in case of provision of a proper canal seal [32].

Based on the results of the present study and similar studies [33,34], complete eradication of bacteria from the root canal system is not possible; however, with proper tapering of the canal and with the aid of irrigation methods such as laser [35] and ozone [36], and sufficient sealing of the root canal, high success rate in root canal treatment may be achieved, without excessive apical enlargement.

Conclusion

Within the limitations of the present in vitro study, the results showed that the BioRaCe and Mtwo rotary systems were not capable of complete elimination of bacteria from the root canal system.

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