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Efficacy of Various Decontamination Methods and Sterilization on Contaminated and Inoculated Diamond-Coated Burs

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Abstract

The objective of this study was to evaluate the effectiveness of various decontamination methods and subsequent sterilization on contaminated and inoculated diamond-coated burs. Diamond-coated burs and extracted human molars were sterilized with a steam sterilizer. Enamel and dentin from the extracted teeth were abraded utilizing diamond-coated burs using a high-speed handpiece. The burs were subsequently inoculated with one of the following microorganisms: Enterococcus faecalis ATCC 19433, Staphylococcus aureus ATCC 6538, Pseudomonas aeruginosa ATCC 15442 or Geobacillus stearothermophilus ATCC 7953. Twenty-four hours after inoculation, the burs were subjected to various cleaning treatments, sterilized, and then cultured for bacterial contamination. The number of CFU/mL was determined per group. Except for the positive control group, no CFU/mL or growth was found for all treatment groups and for all bacterial types**.** In conclusion, the contaminated and inoculated diamond-coated burs tested in this study were successfully sterilized to eliminate the tested bacteria. The use of a cleaning stone with manual cleaning or an ultrasonic cleaner resulted in the least amount of remaining tooth debris on the diamond-coated bur heads.

Keywords: Diamond-coated burs, cleaning, sterilization

INTRODUCTION

Dental burs are one of the most commonly used dental instruments within a dental practice. Of those dental burs, diamond burs with their unique cutting structures are essential dental rotary instruments used for both operative and fixed restorative dentistry [1]. A conventional “diamond bur”, more accurately called a diamond-coated bur, is a metal rod that is coated by galvanic deposition with diamond powder during manufacturing. The shape of the diamond granules imbedded on the bur, resulting in its complex surface roughness, is often a source that invites the accumulation of dental debris, microorganisms and other materials, which in turn make diamond-coated burs more difficult to clean and sterilize [1].

Diamond-coated burs were first introduced in the late 19th century.

Depending on the manufacturer, brand, or cost, the perception of single-use versus multi-use diamond burs varies between different clinical practices and remains controversial. In recent years, however, to eliminate any chance of cross contamination, there has been a push to classify diamond-coated burs as single-use devices [1]. In October of 2002, the Medical Device User Fee and Modernization Act of 2002 (MDUFMA) amended the previous Federal Food, Drug, and Cosmetic Act by providing new regulatory requirements for reprocessed single-use devices (SUDs) [2]. This new amendment removed the previous pre-marketed exemption for diamond-coated burs and now requires manufacturers to provide validation data which includes cleaning, sterilization, and functional performance [3].

Several studies have evaluated the cleaning and sterilization of endodontic files and carbide burs, but limited research has been published investigating diamond-coated burs. For the debridement of endodontic files, a study by Perakaki et al. found that the use of an ultrasonic cleaner for 10 minutes was more successful in cleaning debris from the structurally complex endodontic file than a washer disinfector [4]. Similarly, another study found that an ultrasonic cleaner had a significant effect on the cleanliness of the endodontic files; pre-soaking did not benefit sterilization; and, the optimum time for ultrasonic cleaning was between 5 and 10 minutes [5]. For the debridement of carbide burs, a 2016 case-control study found that the use of a high-pressure autoclaving session followed by a low-pressure steam autoclave session resulted in no bacterial growth on used carbide-fissure burs [6]. A study by Kumar et al. stated that autoclaving or glutaraldehyde was an effective method to sterilize carbide-steel burs [7]. This was further supported in a study by Mathivanan et al., who found that the use of autoclave and hot air ovens were relatively the best method of bur sterilization in comparison to a glass bead sterilizer [8]. However, these studies did not evaluate diamond-coated burs. One study examined the effectiveness of pre-cleaning diamond-coated burs covered with a dye and found that none of the pre-cleaning methods were effective in removing the dye and that the diamond-coated bur head was the most frequently contaminated site on the bur [9]. An additional study on diamond-

coated burs found that none of the cleaning or sterilization methods were absolutely efficacious, but the study did not examine a combination of cleaning and sterilization. [10].

Limited research has been published examining the efficacy of various decontamination methods and sterilization of diamond-coated burs. In addition, no research has been published examining the Clean-A-Diamond Mini Square (Premier, Plymouth Meeting, PA), which is an autoclavable, reusable dressing stone that can reportedly be used to unclog coarse- and medium-grit diamond-coated burs. Therefore, the aim of this study was to evaluate the effectiveness of various decontamination methods and subsequent sterilization on contaminated and inoculated diamond-coated burs. The null hypotheses were there would be no difference between various decontamination methods in: (1) microorganism elimination or (2) debridement of contaminated and inoculated course diamond-coated burs.

METHODS

The Institutional Review Board at Wilford Hall Ambulatory Surgical Center, Joint-Base San Antonio, Lackland, Texas approved this protocol (#FWH20190066N). A total of 7 groups with 20 diamond-coated burs (5847.31.016 FG Super Coarse Flat-End Cylinder Diamond, Brasseler, Savannah, GA) per each group were evaluated. The diamond-coated burs along with extracted human third molars were sterilized with a steam sterilizer (Amsco 400, Steris, Mentor, OH). Each bur was heavily contaminated with enamel and dentin debris via abrasion for 30 seconds with a high-speed handpiece (Forza F5, Brasseler, Savannah, GA) and water coolant. One group of burs was tested after removal from their original packaging and did not receive any contamination with tooth debris.

Three microorganisms: Enterococcus faecalis ATCC 19433, Staphylococcus aureus ATCC 6538, and Pseudomonas aeruginosa ATCC 15442, were grown on trypticase soy agar with 5% sheep blood (TSA II) and incubated (Thermo Forma Steri Cycle 370 CO_2 Incubator, Thermo Fischer, Waltham, MA) at 35 +/- 2°C ambient air for 24 hours. Geobacillus stearothermophilus ATCC 7953

was incubated at 50 +/- 2°C ambient air for 24 hours and grown on TSA II. Staphylococcus aureus and Pseudomonas aeruginosa were chosen due to their extensive use in evaluating sterilization and disinfection procedures by the Environmental Protection Agency [11]. Additionally, Staphylococcus aureus, Pseudomonas aeruginosa and Enterococcus faecalis have been proven to be prevalent in hospital acquired infections [12-14]. Geobacillus stearothermophilus has been utilized to monitor steam sterilization, hydrogen peroxide gas plasma, and liquid peracetic acid sterilizers [15]. Inoculation suspensions of the microorganisms were prepared by cultivating the organisms in Trypticase Soy Broth to a concentration of approximately 1.5 x 10^8 CFU/mL, then a 1:10 dilution of the suspension was made with sterile saline resulting in an inoculum suspension of approximately 1.5 x 10^7 CFU/mL. The diamond-coated burs were inoculated (except negative control and new, unused, pre-packaged bur groups) by immersing them in 1 mL of the inoculum suspensions. They remained in the inoculum for 10 minutes (represents the approximate amount of time the burs would be in the patient's mouth). The burs were then placed in a sterile container for 24 hours. After being contaminated with enamel and dentinal debris, the burs in the negative control group were divided into four groups with 5 burs in each group. Each bur underwent one of four different decontamination methods with subsequent sterilization. This process was to determine if there was outside microorganism contamination at any step during the experiment. Table 1 outlines the various treatment methods completed per group.

The diamond-coated burs from each group were immersed in 1 mL of sterile saline and vortex mixed (Fisher Heavy Duty Vortex Mixer, Fisher Scientific, Waltham, MA) for 2 minutes to remove microorganisms from the bur. The saline from the positive controls were serially diluted (1:10) and plated on TSA II. Saline from each of the cleaning protocol groups (4-7), the negative control, and the new, unused, pre-packaged burs were plated on the TSA II as before. E. faecalis, S. aureus, and P. aeruginosa plates were incubated at 35 +/- 2°C in ambient air for 24 hours. G. stearothermophilus plates were incubated at 50 +/- 2°C in ambient air for 24 hours.

After incubation, the number of colony forming units (CFUs) on the plates were counted and CFU/mL recovered were calculated. The mean CFU/mL and standard deviation was determined per group. The bur heads from Groups 4 through 7 were examined under a light microscope (Nikon SMZ-1B, Melville, NY) at 10x magnification and rated as none (0), minimal (1), moderate (2), or heavy (3) for level of remaining enamel and dentin debris. Representative images of the burs were taken with a stereomicroscope (SZX16, Olympus, Shinjuku, Japan). See Figure 1. The tooth debris data were analyzed with statistical software (SPSS, version 25, IBM, Armonk, NY). All the groups were analyzed with the Kruskall Wallis test (alpha = 0.05). The Mann Whitney U test was used to make comparisons between groups. The alpha value was adjusted to 0.008 with a Bonferroni correction because multiple comparisons were completed simultaneously.

RESULTS

Except for the positive control (Group 1), no CFU/mL or growth was found for all treatment groups and for all bacterial types. None of the diamond-coated burs from the negative control group (Group 2) demonstrated any bacterial growth. Also, none of the new, unused, prepackaged burs demonstrated any bacterial growth (Group 3). See Table 1. For remaining tooth debris, the results of the Kruskall-Wallis test found a significant difference between groups ($p=0.0001$). Using the Mann-Whitney U test, there were significant differences between all the groups ($p<0.008$) except between groups 5 and 7 ($p=0.46$). Group 4 (Median=2, IQR=1) had significantly more debris than all other groups. Group 6 (Median=1, IQR=2) had significantly less debris than Group 4, but significantly greater debris than Groups 5 and 7. Group 7 (Median=0.5, IQR=1) had the lowest level of debris, but it was not significantly less than Group 5 (Median=1, IQR=1).

DISCUSSION

The complex surface structure of diamond-coated burs retains tooth debris and may make them more difficult to sterilize than carbide burs. However, in this

study, there was no difference in microorganism elimination based on decontamination method along with sterilization due to no observable growth of microorganism. Therefore, the first null hypothesis was not rejected. A study by Sajjanshetty et al. found that none of the individual techniques (i.e., manual scrubbing, hot air oven, glass bead sterilizer, ultrasonic cleaner, autoclave) were absolutely efficacious on diamond-coated burs. Of the methods tested, the autoclave was the most effective in decreasing the colony-forming units of Streptococcus mutans [10]. In contrast, this study found that the use of a steam sterilizer in combination with various pre-cleaning procedures resulted in no growth of any of the four tested bacteria on coarse diamond-coated burs. Additionally, this study reinforced the manufacturer's claim that diamond-coated burs are sterile in their individual packages and require no further action prior to first use.

Previous laboratory research found that an ultrasonic cleaner was successful in reducing debris from endodontic files [4,5]. However, Gul et al. found that various pre-cleaning methods (i.e., manual, ultrasonic, manual with enzyme, manual with ultrasonic) were not effective in removing a dye from diamond-bur heads [9]. Comparisons to this study are difficult because the removal of dye would be different from removing debris. This study demonstrated significant differences based on debridement method, so the second null hypothesis was rejected. No pre-cleaning methods were completely efficacious at removing all the dentinal debris. The manual cleaning procedure (Group 4) resulted in the removal of significantly less tooth debris than all the other groups with over 70% of the burs retaining a moderate to heavy level of debris. The use of an ultrasonic cleaner (Group 6) resulted in significantly more debris removal compared to the manual cleaning procedure (Group 4), but significantly less debris removal compared to the use of the manual cleaning and Clean-A-Diamond stone (Group 5) or ultrasonic cleaning and Clean-A-Diamond stone (Group 7). The use of an ultrasonic cleaner along with a Clean-A-Diamond stone (Group 7) resulted in the least amount of remaining tooth debris with 96% of the burs demonstrating minimal or no remaining debris. However, it was not significantly different from the use of manual cleaning and a Clean-A-Diamond stone (Group 5) with 94% of the burs demonstrating

minimal or no remaining debris. The additional use of the Clean-A-Diamond stone appeared to make a dramatic improvement in the decontamination of the diamond-coated burs compared to the use of either the manual cleaning procedure or ultrasonic cleaner alone. See Figure 2.

The disposal of a multi-use diamond-coated bur after one use may not be cost effective. According to the manufacturer, (technical representative, Brasseler), their conventional multi-use diamond-coated bur evaluated in this study should be used a maximum of 12 times before discarding, but only on one patient. A pack of 5 burs is approximately $60, and equates to approximately $12 per bur if not sterilized and re-used. With the new single-use diamond burs, the manufacturer recommends that each bur be discarded after only one preparation. The price of a 25 bur pack is approximately $50, which equates to about $2 per preparation. Differences in costs will vary depending on the manufacturer and type of bur. Cutting efficiency and lifespan is dependent on numerous factors such as coarseness of bur, substrate material (e.g., enamel, dentin, composite, ceramic), preparation time, and the speed and torque of the handpiece and the amount of water spray. Limitations to this study include the use of only one type and coarseness of a diamond-coated bur and only one cycle of debridement and sterilization.

Based on the results of this study, the increased cost and waste created through the single use of diamond-coated burs may be unwarranted since the contaminated and inoculated diamond-coated burs evaluated were satisfactorily debrided and sterilized after one use. If using a diamond-coated bur multiple times, it may be the most efficacious for practitioners to consider either utilizing the protocol outlined in Group 5 - a two-second debridement with a Clean-A-Diamond stone followed by the manual cleaning cycle and one cycle of steam sterilization, or in Group 7 - a two-second debridement with a Clean-A-Diamond stone, a 15 minute ultrasonic cleaning, and one cycle of steam sterilization. Either of these two protocols should ensure a sterile diamond-coated bur that is reasonably free of tooth debris.

CONCLUSION

The contaminated and inoculated diamond-coated burs tested in this study were successfully sterilized to eliminate the tested bacteria. The use of a cleaning stone with manual cleaning or with an ultrasonic cleaner provided the least amount of remaining tooth debris on the diamond-coated bur heads.

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Group		Contamination	Decontamination Method	Sterilization Method
1	Positive Control	Tooth debris only	None	None
2	Negative Control	Tooth debris only	Burs were divided into four groups of five, each undergoing one of the decontamination and sterilization methods noted in Groups 4-7.	Steam-one cycle of steam sterilization✯
3	New, unused, prepackaged	None	None	None
4	Manual cleaning (Brasseler IFU)	Tooth debris and bacteria	One-minute rinse under cool running water 10-minute immersion in a neutral-pH cleaning solution+ One-minute brush in the solution+ One-minute rinse under warm water until visibly clean	Steam-one cycle of steam sterilization✯
5	Clean-A-Diamond stone & manual cleaning (Brasseler IFU)	Tooth debris and bacteria	2 seconds of debridement with the Clean-A-Diamond stone^ One-minute rinse under cool running water 10-minute immersion in a neutral-pH cleaning solution+ One-minute brush in the solution+ One-minute rinse under warm water until visibly clean	Steam-one cycle of steam sterilization✯
6	Ultrasonic Cleaning (Brasseler IFU)	Tooth debris and bacteria	15-minute sonication in an ultrasonic unit*	Steam-one cycle of steam sterilization✯
7	Clean-A-Diamond stone & Ultrasonic cleaning (Brasseler IFU)	Tooth debris and bacteria	2 seconds of debridement with the Clean-A-Diamond stone^ 15-minute sonication in an ultrasonic unit*	Steam-one cycle of steam sterilization✯
+Dawn Ultra, Proctor & Gamble, Cincinnati, OH; *1000 Pro-Sonic, Sultan Healthcare, York, PA; ✯Amsco 400; ^Mini Square, Premier, Plymouth Meeting, PA				

Table 1: Cleaning, decontamination, and sterilization methods by Group.

Treatment Groups	CFU/mL (range)			
	Enterococcus faecalis	Staphylococcus aureus	Pseudomonas aeruginosa	Geobacillus stearothermophilus
Group 1	1.2-5.3 x 10^5	1.1-7.9 x 10^5	1.2-6.7 x 10^6	1.0-1.6 x 10^5
Group 2	No growth	No growth	No growth	No growth
Group 3	No growth	No growth	No growth	No growth
Group 4	No growth	No growth	No growth	No growth
Group 5	No growth	No growth	No growth	No growth
Group 6	No growth	No growth	No growth	No growth
Group 7	No growth	No growth	No growth	No growth

Table 2: Bacterial growth of each of the Groups.

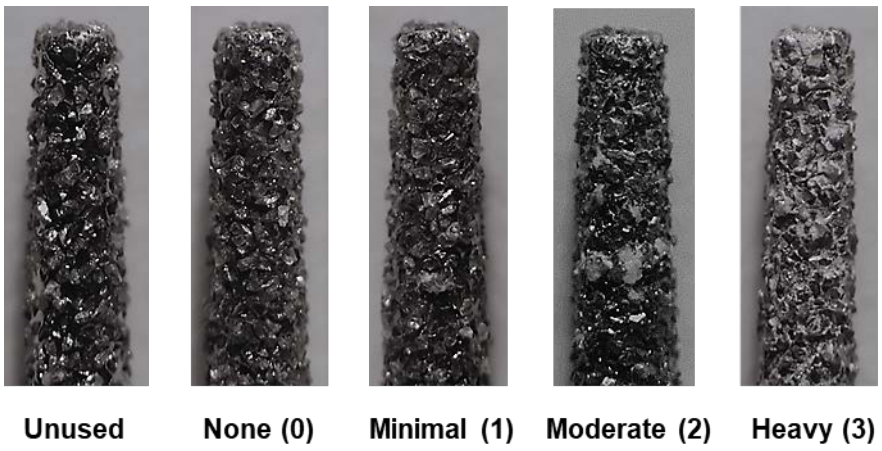
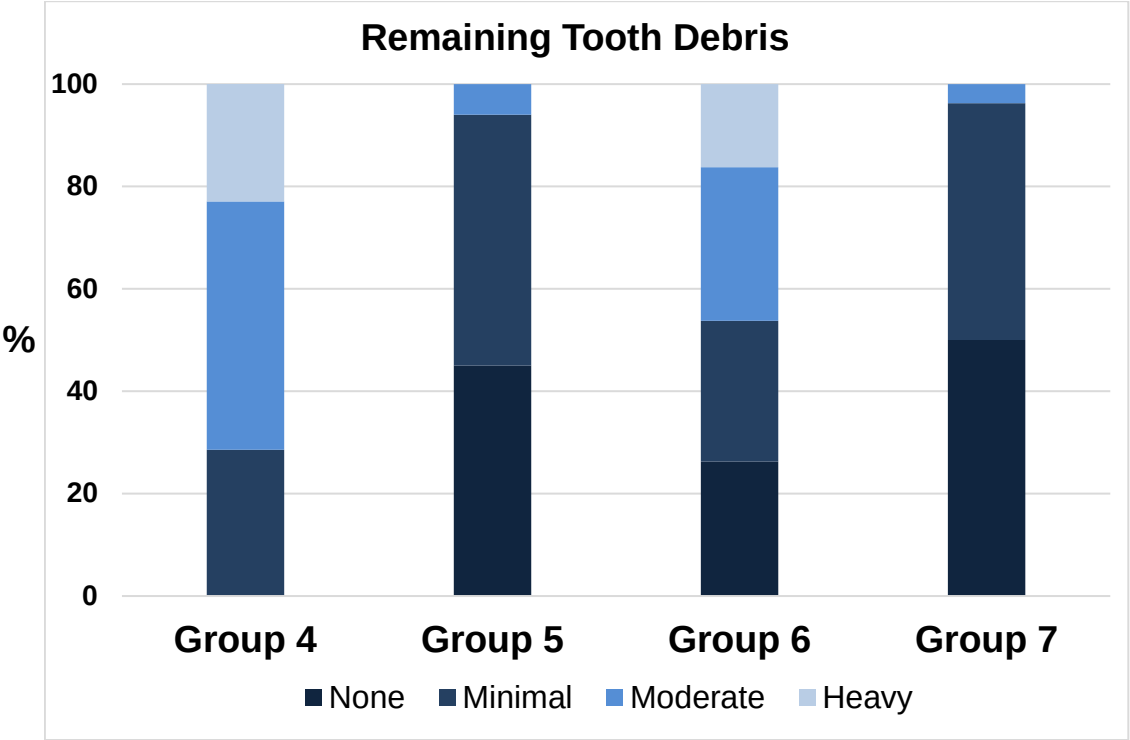


Figure 1: Representative image of unused bur and burs with various of level of tooth debris rated as None (0), Minimal (1), Moderate (2), or Heavy (3). After decontamination and sterilization, each bur from Groups 4 – 7 were rated for level of debris.



Remaining Tooth Debris
100
80
60
40
20
0
Group 4
Group 5
Group 6
Group 7
None
Minimal
Moderate
Heavy

Figure 2: Level of remaining tooth debris for Groups 4 through 7.