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Assessment of contamination on sterilised dental burs after being subjected to various pre-cleaning methods

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Abstract

Objective: To determine the effectiveness of different pre-cleaning methods by determining frequency and site of contamination on the sterilised dental burs using Phloxine B dye.

Method: The in-vitro experimental study was conducted from June to August 2017 at dental clinics of Aga Khan University Hospital Karachi. Diamond dental burs were selected and divided into two control and four test groups. The two control groups were classified as Negative (new burs) and Positive (used contaminated). The four test groups were classified as Manual (Group-1), Ultrasonic (Group-2), Manual + Enzyme (Group-3) and Manual + Ultrasonic (Group-4). Phloxine B dye was used to determine the contamination. The images of the burs were taken and enlarged at 15X before subjected to visual assessment. Association between contamination and pre-cleaning methods were determined. Data was analysed using SPSS version 22.

Results: A total of 210 burs were selected for the study which were divided in 6 groups of 35(16.66%) each. One (2.8%) bur in negative control group and all burs in positive control group showed contamination. In test groups, 27(77.1%), 29(82.8%), 27(77.1%) and 24(68.5%) burs showed contamination in groups 1, 2, 3 and 4, respectively. There was no association between type of pre-cleaning method with the frequency of contamination ($p=0.57$). The head of bur was the most frequently contaminated site ($p \leq 0.003$).

Conclusions: None of the pre-cleaning method was found to be effective. Head of bur was the most frequently contaminated site.

Keywords: Cross infection; Decontamination; Phloxine B; Sterilisation. (JPMA 68: 1188; 2018)

Introduction

In dentistry, infection control is a major concern due to risk of transmission of communicable diseases.¹ According to the universal guidelines, dental practitioners are required to sterilise instruments that come in contact with the saliva and blood during dental procedures.^{2,3} Proper sterilisation of the dental instruments by adhering to the universal infection control protocols can prevent the spread of infection.^{4,5} Various doubts have been raised over the effectiveness of instrument cleaning in the dental practices. An observational study in the United Kingdom (UK) showed that the cleaning of dental instruments is insufficiently managed and poorly controlled.⁵

Dental burs are used to accomplish a variety of objectives which include cavity preparation, access opening during root canal treatment, caries removal, crown preparation and a myriad of other dental procedures.¹ They can become contaminated heavily with saliva, blood, necrotic tissue and potential pathogens which can act as a source

of cross-infection.⁶ Due to complex architecture of dental burs, pre-cleaning and subsequent sterilisation is difficult to achieve.⁷ Proper attention to adequate cleaning, disinfection and sterilisation should be ensured in the routine dental practice to prevent cross-infection.⁶

Various methods, including bacterial culturing, microscopic evaluation and dye staining, are used to evaluate contamination on the dental burs.^{1,8,9} Phloxine B is an antibacterial dye that can be easily used to assess and visualise any contamination. The dye stains both gram-positive and gram-negative bacteria.¹⁰

A study reported that new dental burs, provided by the manufacturer, were not sterile, nearly 42% of those had contamination and 15% of the used burs had contamination after sterilisation.¹¹ Studies have reported ineffectiveness of various pre-cleaning methods to decontaminate used dental burs.^{12,13} To the best of our knowledge, there is no local data on the contamination assessment of dental burs. The current study was planned to investigate the effectiveness of different pre-cleaning methods by determining frequency and site of contamination remaining on sterilised dental burs using Phloxine B dye. The target was to identify areas that could improve pre-cleaning methods before sterilisation procedure and prevent the chances of cross-infection or

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limit the use of these dental burs to single use.

Materials and Method

The in-vitro experimental study was conducted from June to August 2017 at the dental clinics of Aga Khan University Hospital, Karachi. New high-speed diamond dental burs either sterilised or untreated were included while corroded dental burs or those with manufacturing defects were excluded.

Approval from the institutional ethics committee was obtained. Sample size was calculated using World Health Organisation (WHO) sample size calculator and using data from an earlier study,^{14,15} which reported the prevalence of bacterial contamination at 45% on dental burs. Absolute precision was kept at 20% and confidence interval (CI) at 95 %. The required sample size was 24 per group. We inflated it to 35. The dental burs included (MANI, INC, Utsunomiya, Tochigi, Japan) were divided into four test and two control groups.

During collection, the burs were handled using sterile technique to prevent any contamination. After collection, all the burs were randomly allocated to positive control, negative control and test groups.

All the burs of test groups and positive control group were then used for access opening in permanent teeth and after the procedure the burs that were contaminated from bacteria, blood, saliva, tooth and tissue debris were carried using sterile technique to the sterilisation department except positive control group burs where different cleaning methods were used prior to steam autoclave.

The negative control group had uncontaminated burs (new/unused) while the positive control group consisted of used contaminated burs. Among the test groups; Group-1 had contaminated burs which were subjected to manual cleaning using bur brush (Ash instruments, Dentsply Ltd, Weybridge, Surrey). Burs were subjected to 20brush strokes moving from shank to head portion and holding with a sterile tweezer under running water. Group-2 consisted of contaminated burs which were subjected to ultrasonic cleaning in ultrasonic bath (Coltene/Whaledent, Inc. Biosonic UC50D, Altstätten Switzerland) containing distilled water for 10 minutes as per manufacturer instructions. Group-3 had contaminated burs which were subjected to manual scrubbing and placement in enzymatic solution (InstruZym, Dr. Deppe, Kempen, Germany) for 10 minutes as per manufacturer instructions. Group 4 consisted of contaminated burs which were subjected to manual scrubbing, ultrasonic cleaning for 10 minutes, and

placement in enzymatic solution for 10 minutes.

After cleaning, all the dental burs of test groups were sterilised using steam autoclave (Melag, Vacuklave 23 B+, Berlin, Germany) for 3.5 minutes at 134°C. The identification numbers were labelled on the sterilisation pouches. For staining, all the burs of control and test groups were placed in an individually labelled sterile micro-centrifuged tube (Axygen Scientific, Sweden) containing 2ml of Phloxine B dye (100µg/ml) (Sigma-Aldrich, St. Louis, Missouri, United States). The tubes were then placed in an ultrasonic bath for 10 minutes and rinsed with de-ionised water and allowed to air-dry. These processed burs were assessed visually and then photographed using digital single-lens reflex (DSLR) camera (Canon EOS 70D body and 1000mm lens). All photographs were examined by two separate investigators who were not involved in the cleaning process twice at two different intervals on a computer screen magnified at 15X for evaluation of staining. Inter-examiner reliability was examined on a subset of 50 randomly selected burs and agreement between two investigators was determined using Kappa statistics that turned out to be excellent at 0.80. All the burs that were contaminated, as shown by staining, were then examined for the determination of most frequently contaminated site. Each bur was examined at head, neck and shank for the presence or absence of staining. Data was analysed using SPSS 22. Descriptive analysis was performed to determine the frequency of contamination in different groups and bur sites and chi-square test was applied to determine the association of different pre-cleaning methods and site of bur with the frequency of contamination. Level of significance was kept at $p \leq 0.05$.

Results

There were 210 burs which were divided into 6 groups of 35(16.66%) each. In the negative control group, only 1(2.8%) dental bur showed contamination. In positive control group, all (100%) the burs showed contamination (Table-1).

Among the test groups, 27(77.1%) dental burs in Group-1, 29(82.8%) in Group-2 and 27(77.1%) in Group-3 demonstrated contamination. In Group-4, 24(68.5%)

Table-1: Frequency of contamination in the Control Groups.

Control Group n=35/group	Staining n (%)
Negative (new and unused)	1 (2.8%)
Positive (new and used)	35 (100%)

*n= 70 (35 per group).

Table-2: Frequency of contamination in the used burs subjected to different pre-cleaning methods.

Test Groups n= 35/Group	Staining n (%)	p-value
Group-1 (Manual scrubbing)	27 (77.14%)	0.57
Group-2 (Ultrasonic)	29 (82.85%)	
Group-3 (Manual + Enzyme)	27 (77.14%)	
Group-4 (Manual +Ultrasonic +Enzyme)	24 (68.57%)	

* Chi-square test was applied at ≤ 0.05 level of significance.

*n= 140 (35/group).

because it was convenient, easily available, with no need for expensive and delicate instrument and results could be achieved in shorter time.

The present study used two control groups. The primary reason was to confirm the efficacy of the dye to stain only contaminated surfaces, and, secondly, to assess the sterility of brand new burs. We found that only one unsterilized brand-new bur in the negative control group showed contamination that can be due to handling error. A study also reported contamination in new burs assessed using bacterial culture.¹¹ On the contrary, a study showed no contamination in new dental burs assessed using bacterial culture.¹⁵ These results indicate that in theory, brand-new burs can be used without sterilisation but at

Table-3: Association between bur sites and frequency of contamination subjected to different pre-cleaning methods.

Contamination	Bur Sites n (%)			Total n = 420	Head vs neck	p-value Head vs shank	Neck vs shank
	Head n =140	Neck n =140	Shank n =140				
Yes	99 (70.7%)	19 (13.6%)	1 (0.7%)	119 (28.3)	0.003	1.00	0.136
No	41 (29.3%)	121 (86.4%)	139 (99.3%)	301 (71.7%)			
Total	140 (100%)	140 (100%)	140 (100%)	420 (100%)			

* Chi-square test / Fisher's Exact Test was applied at ≤ 0.008 level of significance.

dental burs showed contamination after staining with the dye. There was no association ($p=0.57$) between the type of pre-cleaning and contamination on burs (Table-2).

The frequency of contamination was significantly associated with the sites of burs ($p<0.05$), and among the different sites, the head portion of the dental burs was the most frequently contaminated site ($p\leq 0.003$) (Table-3).

Discussion

Dental burs are recognised as potential source of cross infection due to their contact with teeth, blood, saliva and bone during various dental procedures.¹⁶⁻¹⁸ Studies have shown that reusing of dental instruments is a common practice and there are serious concerns regarding the cleanliness of these instruments before use on the patients.¹⁹ Studies have also demonstrated that effective cleaning of dental instruments before sterilisation reduces the risk of cross-infection.²⁰ While most of the dental instruments are effectively cleaned after use, the diamond bur is often neglected and only brushed or immersed in a mild disinfectant prior to any reuse.²¹ Different studies have been conducted using various methods of contamination assessment.^{1,8,9} In the present study, the method for contamination assessment used was staining using Phloxine B dye. We used this method

the same time there is a chance of contamination. As such, it is recommended to sterilise the new burs before use on patients.

Manual cleaning, is simple and cost-effective but can potentially serve as a source of contamination due to the aerosols produced during the process.^{12,13} Manual scrubbing also produce unpredictable results, as it is very operator-sensitive.⁶ In our study, 77.14% dental burs in Group-1 demonstrated contamination. Studies have reported that manual cleaning is unreliable and ineffective in removing contamination from the dental instruments.^{6,22,23}

Ultrasonic cleaning has been found to be effective in removing saliva and dried blood from the dental instruments and increases safety of dental personnel during handling of instruments.²⁴ In the present study, 82.85% burs in Group-2 showed contamination. A study reported contamination in 58.3% diamond burs when subjected to ultrasonic cleaning method and assessed using scanning electron microscopy (SEM).²⁵ The difference in the frequency of contamination might be due to different variables like type of ultrasonic bath, processing time, cleaning solutions, temperature of the ultrasonic bath and the difference in the assessment

method. Studies have also shown that ultrasonication is insufficient to remove contamination.^{22,26}

Enzymatic agents help to digest organic debris, including bacteria due to presence of amylases, lipases and proteases, but their use is operator-dependent and if manufacturer's guidelines are not exactly followed regarding dilution of enzyme solution, its immersion time, reuse of solution greatly affects the cleaning efficacy and might lead to recontamination of dental instruments.⁶ In the present study, the burs in group-3 were treated with a combination of manual cleaning followed by immersion in the enzyme solution. Nearly, 77.14% burs showed contamination with this approach. This demonstrates that regardless of the pre-cleaning method, once contaminated, dental burs are very difficult to clean.

The burs in the test group-4 were subjected to a combination of pre-cleaning methods (manual + ultrasonic + enzyme) and yet 68% diamond dental burs exhibited contamination. A study reported contamination in 45% burs after combination of pre-cleaning methods (manual + ultrasonic + washer disinfectant) followed by gas sterilisation.¹⁵

Our results suggest that current methods of pre-cleaning of dental burs might be insufficient to ensure complete cleaning of contaminants. This is probably attributed to complex geometrical design of the bur head and inability of cleaning agent to adequately access the surface of the contaminated instrument.

The most commonly contaminated site in the present study was the head portion (working end) of the bur regardless of the pre-cleaning method used. Similar results were described in another study.⁶ This is probably attributed to the surface roughness of the head portion; bur head is the part which mostly encounters the infected tooth.

In the present study, none of the pre-cleaning methods were 100% effective in removing the contamination even after steam autoclave because remaining organic debris if not removed properly will protect the pathogens from the effect of steam autoclave. Our results are similar to other studies that showed contamination of different dental instruments even after pre-cleaning and sterilisation procedures.^{11,15}

The strengths of the present study are that multiple pre-cleaning methods were compared and contamination at each part of a bur was separately evaluated. The limitations of the study include its inability to identify the nature of remaining contaminants that can be either bacterial contaminants, food debris or any other host cell

adherent material. Therefore, we recommend more sensitive testing like culture and genomic testing for exact identification of contaminating source, multicentre study with incorporating additional variables such as time of usage of burs, burs of different materials, appropriate education and training for the staff responsible for sterilisation and all instruments should be considered single-use if they cannot be cleaned properly. Our future research plan is to perform more sensitive and quantitative method to assess the effectiveness of decontamination methods.

Conclusions

None of the pre-cleaning methods were effective against decontamination of the diamond burs. However, combination of multiple methods was found to be relatively better although not statistically significant to decontaminate the burs prior to sterilisation. Head of the dental bur was the most contaminated site and was least affected by any pre-cleaning method.

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Conflict of Interest: None.

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