Original Article

Efficacy of Hand-free High-volume Evacuation Device in Reducing Bacterial Aerosol and Splatter During Endodontic Access Opening Procedure

Swaros Yuyuen[1], Soranun Chantarangsu[2], Ruchanee Salingcarnboriboon Ampornaramveth[3]

[1]Dental department, King Taksin Memorial Hospital, Bangkok Metropolitan Administration, Bangkok, Thailand

[2]Oral Pathology Department, Faculty of Dentistry, Chulalongkorn University, Bangkok, Thailand

[3]Center of Excellence on Oral Microbiology and Immunology, Microbiology Department, Faculty of Dentistry, Chulalongkorn University, Bangkok, Thailand

Abstract

To compare the effectiveness of aerosols reduction during endodontic access opening (OC) procedure using a hand-free high-volume evacuation device (EasyPrep) versus a rubber dam. Thirty-eight patients who required endodontic treatments of posterior mandibular teeth were recruited and divided into two groups which are rubber dams with high-volume evacuation (HVE) and EasyPrep. Air samples were collected by settle-plate technique from five different locations and at three different time points - before operation (baseline), at operation, and after operation. The number of colony-forming units (CFU) was enumerated. The genus and species of bacteria were identified by a proteomic fingerprint using a mass spectrometer. Statistical analysis was conducted using IBM SPSS Statistics version 29.0 (IBM). At and after operation, the total CFU count was significantly higher than the baseline in both groups across various locations ($p<0.05$). The total CFU in the EasyPrep group was slightly higher than the rubber dam group. However, there was no statistically significant difference in total CFU count at operation between the two groups at different locations. Gram-positive cocci species including *Micrococcus luteus* were the most found bacteria. A hand-free high-volume evacuation device provided comparable results to rubber dam in reducing aerosols during OC procedures.

Keywords: Rubber dam, EasyPrep, High-volume evacuation, Dental aerosol, Hand-free high-volume evacuation device

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Correspondence to:

Ruchanee Salingcarnboriboon Ampornaramveth, Center of Excellence on Oral Microbiology and Immunology, Microbiology Department, Faculty of Dentistry, Chulalongkorn University, 34 Herry-dunang Rd. Wangmai, Patumwan, Bangkok 10330 Thailand. Tel: 02-218-8682 E-mail: ruchanee.a@chula.ac.th

Introduction

When performing dental procedures such as restoration, ultrasonic scalers, and open access in endodontics, aerosols are inevitably created within the dental facility.[1] It has been widely discussed that these aerosols pose a significant risk to patients and dental professionals working in dental clinics. In 1969, Micik *et al.*[2] mentioned that droplets with a size greater than 50 micrometers tend to settle quickly onto surfaces near the source and cannot remain in the air due to their relatively large size. On the other hand, aerosols with a size of less than 50 micrometers can linger in the air for several hours before settling onto a surface. Aerosols produced during dental procedures, in particular, are usually small, with most of them being less than 5 micrometers in size, and can remain airborne for prolonged periods, even travelling long distances.[3] It should be noted that aerosols generated during dental procedures carry a variety of microorganisms from the oral cavity and dental unit waterlines. These microorganisms can spread in the air and settle on all surfaces in a dental room, increasing the risk of infection to dental professionals and patients. It is, therefore, crucial to take appropriate measures to minimize the generation and spread of aerosols in the dental clinic.[4-8] Studies have shown that oral microorganisms such as Staphylococcus spp., Streptococcus spp., Micrococcus spp., Bacillus spp., and others are present in aerosols generated during dental procedures.[9-11]

It is interesting to note that in 1862, Dr. Sanford Barnum invented the rubber dam to create a saliva-free field in the mouth during dental procedures. Later, Dr. G. A. Bowman developed the rubber dam clamp to stabilize the rubber dam to a tooth. The use of a rubber dam helps in tooth isolation and ensures that there is no microbial contamination during the dental procedure. The dam acts as a barrier against the saliva, tongue, and oral bacteria of the patient. It also provides several other advantages, such as enhancing visibility, providing a clean operation field, and preventing ingestion or aspiration of foreign objects.[12] Cochran and colleagues[13] indicated that the rubber dam is an effective measure to reduce microorganisms during restorative procedures.

The rubber dam is an essential tool in root canal treatment and is used from the beginning access opening in the first step all the way to the end. In addition, the use of a high-volume evacuator (HVE) can significantly reduce the amounts of aerosols generated during dental procedures by more than 90%.[14-15] While using a rubber dam and HVE effectively reduces aerosols during dental procedures, there are certain situations where its use may pose some challenges. Specifically, when working on teeth with crowns or rotated teeth that alter the crown-to-root relationship, the clinician may find it difficult to properly orient the tooth axis due to the dam's limiting effect. This can result in issues such as gouging, missed canals, and perforations, which usually occur due to faulty angulation of the bur with respect to the long axis of the root.[16] According to Dahlke *et al.*[17], a study was conducted to compare the effectiveness of the Hand-free high-volume evacuation device (the Isolite system, Santa Barbara, California) with that of the dental dam and HVE, in reducing spatter from a dental operative site during simulated occlusal surface preparations on three typodont teeth in a dental manikin. The study found no statistically significant difference between the Isolite system and the dental dam with HVE. It is important to note that this study primarily focused on restorative procedures and was conducted in a laboratory setting. Our study aimed to investigate the aerosolized bacteria that were generated during open access in endodontics. The focus was on comparing the efficacy of bacterial reduction between a rubber dam with HVE and a hand-free high-volume evacuation device (EasyPrep, Megaforce, Taiwan) during endodontic access opening.

Materials and methods

The Bangkok Metropolitan Administration Ethical Committee approved this study according to the Declaration of Helsinki, Belmont Report, CIOMS Guidelines, and ICH-GCP Guidelines (S007h/65).

Sample size calculation

According to the preliminary study investigation conducted by Dahlke *et al.*[17], to achieve a power of 0.80

with a significance level of alpha at 0.05, 17 trials in each group were necessary, based on the mean difference and standard error between the two groups. To account for a 10% potential attrition rate, 38 trials needed to be recruited.

Study design

The study involved performing an access opening procedure on healthy volunteers' posterior mandibular teeth before endodontic treatment, with informed consent. Thirty-eight endodontic cases were recruited and randomly (block-randomization: block of 4) assigned to either use rubber dam (19 cases) or EasyPrep (19 cases). The patients set up for each tooth isolation equipment were demonstrated in Figure 1A and B.

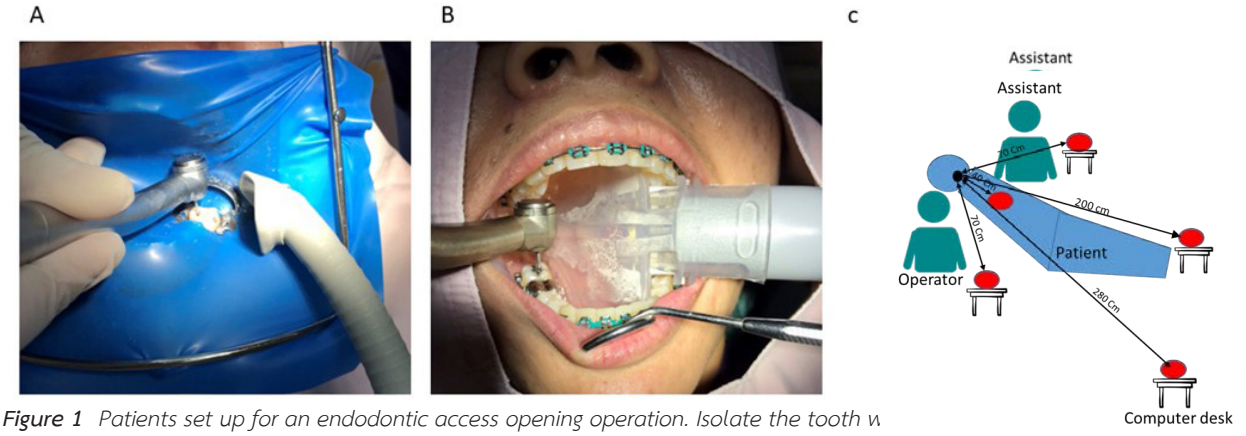

Figure 1 *Patients set up for an endodontic access opening operation. Isolate the tooth w (A) or EasyPrep (B). Positions where air samples were collected by settle plate technique (C).*

Patients with periodontitis, an upper respiratory tract infection, or on antibiotics, on an immunosuppressant, or undergoing chemotherapy, or have a seafood allergy (can't use 0.23% povidone mouthwash) and had an access opening procedure time of more than 45 minutes, were excluded. The treatment procedures were performed in a close dental operatory room size 4.16*4.30 square meters with one dental unit (Osada, model: smily). In-room air ventilation comprised one air conditioner (Carrier 13307 BTU) and one air purifier (Sharp model KI-E60TA). The air conditioner and air purifier were turned on 30 minutes before treatment. The room door was closed until the end of the procedure. The samples were collected as the first case in the morning and limited to one case per day. Microbial in the air were collected by passive air sampling or settle plates technique using sheep blood agar in 9 cm Petri dish plate at five different locations around the patient listed as follows (Fig. 1C);

1. The patient's chest (40 cm from the patient's mouth)
2. Dentist's mobile cabinet (70 cm from the patient's mouth)
3. Dental assistant's portable cabinet (70 cm from the patient's mouth)
4. On the table near patient's feet (200 cm from the patient's mouth)
5. On the computer table (280 cm from the patient's mouth)

Air samples were collected for 30 minutes at three different time points, which were at baseline (before operation), at and after operation. Prior to the procedure, patients were asked to rinse their mouth with 0.23% povidone mouthwash for 30 seconds. A fresh set of agar plates was opened at the beginning of the procedure. The timer was started from the initiation of the access opening and continued for up to 30 minutes. The access opening procedure included the removal of caries with a high-speed handpiece, followed by removing the roof of the pulp chamber. Then, all root canals were filed and irrigated with 2.5% Sodium hypochlorite (NaOCl). Calcium hydroxide ($Ca(OH)_2$) was applied in the pulp chamber, followed by a cotton pellet, Cavit, and IRM. After the patient left the room, a set of new agar plates was opened for 30 minutes.

All agar platesd were incubated at 37 degrees Celsius in an incubator (POL-EKO, model: CLW 750 STD, Poland) for 24-48 hours. The number of colony-forming units (CFUs) was counted and bacterial identification

(Genus and species) was performed by Mass spectrometer (Bruker, model: MALDI Biotyper, Germany) to determine a unique proteomic fingerprint of an organism.

Statistical analysis

All analyses were performed using IBM SPSS Statistics version 29.0 (IBM). Descriptive statistics were reported as frequencies and percentages for qualitative variables and means and standard deviations (SD) or median and interquartile range (IQR) for quantitative variables. The quantitative variables including age, number of decayed teeth and total CFU were tested for normal distribution using Shapiro-Wilk test. Then, the differences between RD and EP groups were analyzed by independent *t*-test (for age), Mann-Whitney U test (for number of decayed teeth), and analysis of covariance (ANCOVA) after controlling for baseline total CFU (for total CFU at operation and after operation). The differences of proportion in gender, tooth number, diagnosis and having underlying diseases between RD and EP groups were analyzed by Chi-square or Fisher exact tests. Difference of total CFU at baseline, at operation and after operation within each group were analyzed using the Friedman test followed by Dunn's post-hoc test. A p-value<0.05 was considered a statistically significant difference.

Results

All participants in this study consisted of 15 (39.5%) males and 23 (60.5%) females with an age range of 20-84 years old (mean 47.2 $\pm$ SD 19.0). Most of the tooth numbers included in this study were the first molars (68.4%), followed by the second premolars (23.7%) and the first premolars (7.9%). The pulp necrosis (52.6%) was the most frequent diagnosis, followed by the irreversible pulpitis (39.5%), chronic apical abscess (5.2%), and acute apical abscess (2.6%). Most of the participants in this study had no underlying disease (63.2%). Baseline demographic, characteristics and clinical data of the participants are shown in Table 1.

Table 1 *Baseline demographic, characteristics, and clinical data of the participants*

Variable	Total (n=38)	RD (n=19)	EP (n=19)	*p*-value
Age, mean (SD)	**47.2 (19.0)**	**45.1 (19.4)**	**49.3 (19.0)**	**0.503**[a]
Gender, n (%)				**0.319**[b]
Male	15 (39.5)	9 (47.4)	6 (31.6)	
Female	23 (60.5)	10 (52.6)	13 (68.4)	
Tooth number, n (%)				**0.509**[b]
34	1 (2.6)	0 (0)	1 (5.3)	
35	7 (18.4)	5 (26.3)	2 (10.5)	
36	14 (36.8)	7 (36.8)	7 (36.8)	
44	2 (5.3)	1 (5.3)	1 (5.3)	
45	2 (5.3)	0 (0)	2 (10.5)	
46	12 (31.6)	6 (31.6)	6 (31.6)	
Diagnosis, n (%)				**0.221**[b]
Irreversible pulpitis	15 (39.5)	9 (47.4)	6 (31.6)	
Pulp necrosis	20 (52.6)	8 (42.1)	12 (63.2)	
Chronic apical abscess	2 (5.2)	2 (10.5)	0 (0)	
Acute apical abscess	1 (2.6)	0 (0)	1 (5.3)	
Number of decayed teeth, median (IQR)	**1.0 (2.0)**	**1.0 (3.0)**	**1.0 (2.0)**	**0.318**[c]
Having underlying diseases, n (%)	**14 (36.8)**	**8 (42.1)**	**6 (31.6)**	**0.501**[b]
Underlying diseases, n (%)				
Hypertension	8 (21.1)	3 (15.8)	5 (26.3)	0.693[d]
Kidney disease	1 (2.6)	1 (5.3)	0 (0)	1.000[d]
Diabetes mellitus	1 (2.6)	1 (5.3)	0 (0)	1.000[d]
Gastric disease	2 (5.3)	1 (5.3)	1 (5.3)	1.000[d]

Table 1 *Baseline demographic, characteristics, and clinical data of the participants (cont.)*

Variable	Total (n=38)	RD (n=19)	EP (n=19)	*p*-value
Hypersensitivity	1 (2.6)	1 (5.3)	0 (0)	1.000[d]
Tachycardia	1 (2.6)	1 (5.3)	0 (0)	1.000[d]
Parkinson's disease	1 (2.6)	1 (5.3)	0 (0)	1.000[d]
Hyperlipidemia	1 (2.6)	0 (0)	1 (5.3)	1.000[d]

Abbreviations: SD, standard deviation; IQR, interquartile range; RD, rubber dam; EP, EasyPrep.

[a] *Difference between groups analyzed by independent t-test.*

[b] *Difference between groups analyzed by Chi-square test.*

[c] *Difference between groups analyzed by Mann-Whitney U test.*

[d] *Difference between groups analyzed by Fisher exact test.*

There was no statistically significant difference of baseline demographic, characteristics, and clinical data between RD and EP groups ($p>0.05$).

During access opening procedure with conventional aerosol control measure by utilizing rubber dam and HVE, the average bacteria in the air around operatory fields collected by settle plate technique were found to range from 3 to 9 CFU. While the average bacteria in the air from the EasyPrep group ranged from 6-16 CFU. (Fig. 2)

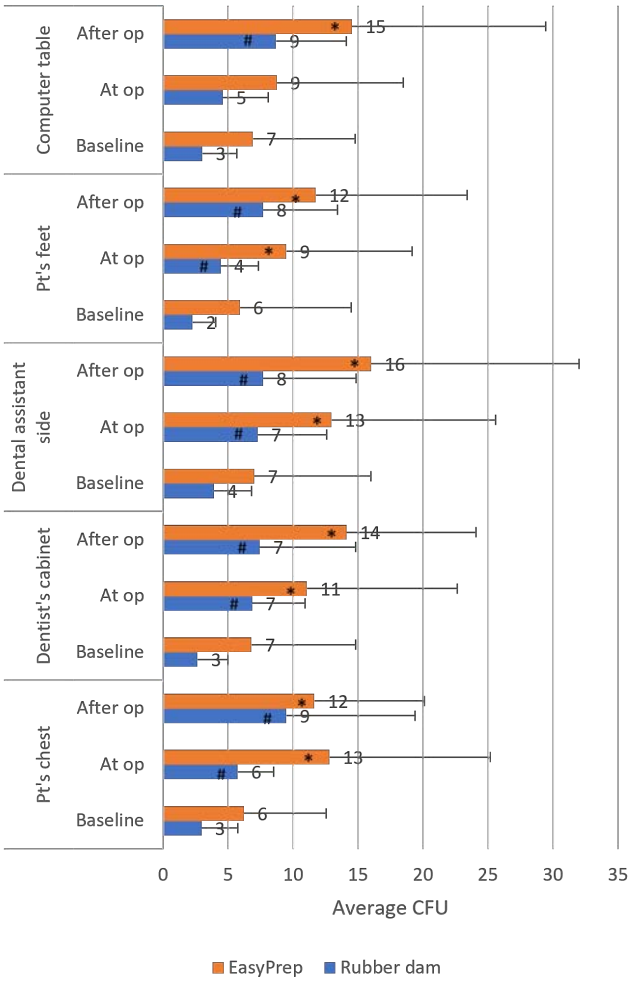

Figure 2 *Average total bacteria in the air collected by passive air sampling (settle plate technique) before, at and after endodontic access opening procedure. Blue bar indicates the rubber dam group. Orange bar indicates the EasyPrep group. For the same intervention at the same location, * and # showed a significant difference when compared with the baseline at p<0.05.*

Although the average CFU were higher in the EasyPrep group, there was no statistically significant difference observed between the two groups. (Fig. 2) Bacteria in the air at and after operation were found to be significantly higher than before operation in both rubber dam and EasyPrep groups.

Table 2 presents the ANCOVA results of total CFU at operation and after operation between RD and EP groups after controlling for baseline total CFU at different locations. There was no statistically significant difference of total CFU at operation between two groups at various locations including patients' chest, beside dentist, beside dental assistant, foot, and computer desk (p=0.239, 0.958, 0.285, 0.258 and 0.933, respectively). Additionally, there was no statistically significant difference of total CFU after operation between two groups at any of the locations (p>0.05). Total CFU at operation and after operation showed statistically significantly higher than baseline in both groups at various locations including patients' chest, beside dentist, beside dental assistant, and foot (p<0.05). However, no difference of total CFU in both groups was found between at operation and after operation at patients' chest, beside dentist, beside dental assistant, and foot. Additionally, total CFU at the computer desk after operation showed statistically significantly higher than both baseline and at operation in RD and EP groups (p<0.05), but no difference was found between baseline and at operation.

Table 2 *Comparison of total CFU between RD and EP groups according to locations and time points, analyzed using analysis of covariance (ANCOVA) after controlling for baseline total CFU*

Location	Time point	Mean (SD)		Adjusted mean (SE)[a]		Mean difference (RD – EP)[a]	p-value[a] (RD vs EP)
		RD (n=19)	EP (n=19)	RD (n=19)	EP (n=19)		
Patient chest	Baseline	2.95 (2.78)[A]	6.21 (6.33)[A]				
	Operation	5.74 (2.75)[B]	12.79 (12.38)[B]	8.21 (1.21)	10.32 (1.21)	-2.103	0.239
	After	9.47 (9.90)[B]	11.63 (8.49)[B]	11.56 (1.62)	9.54 (1.62)	2.023	0.396
	p-value[b] (within group)	<0.001	<0.001				
Beside dentist	Baseline	2.63 (2.36)[A]	6.79 (8.04)[A]				
	Operation	6.84 (4.06)[B]	11.05 (11.60)[B]	9.00 (1.47)	8.89 (1.47)	0.114	0.958
	After	7.42 (7.40)[B]	14.11 (9.96)[B]	9.18 (1.73)	12.34 (1.73)	-3.159	0.218
	p-value[b] (within group)	<0.001	0.001				
Beside dental assistant	Baseline	3.89 (2.90)[A]	7.00 (8.97)[A]				
	Operation	7.26 (5.30)[B]	12.95 (12.63)[B]	9.22 (1.13)	10.99 (1.13)	-1.764	0.285
	After	7.68 (7.18)[B]	16.00 (16.02)[B]	9.96 (1.81)	13.73 (1.81)	-3.765	0.155
	p-value[b] (within group)	0.003	<0.001				
Foot	Baseline	2.26 (1.76)[A]	5.89 (8.59)[A]				
	Operation	4.42 (2.89)[B]	9.47 (9.70)[B]	6.19 (0.91)	7.70 (0.91)	-1.507	0.258
	After	7.68 (5.72)[B]	11.74 (11.65)[B]	9.88 (1.26)	9.54 (1.26)	0.342	0.852
	p-value[b] (within group)	<0.001	<0.001				

Table 2 *Comparison of total CFU between RD and EP groups according to locations and time points, analyzed using analysis of covariance (ANCOVA) after controlling for baseline total CFU (cont.)*

Location	Time point	Mean (SD)		Adjusted mean (SE)[a]		Mean difference (RD – EP)[a]	p-value[a] (RD vs EP)
		RD (n=19)	EP (n=19)	RD (n=19)	EP (n=19)		
Computer desk	Baseline	3.00 (2.65)[A]	5.89 (8.59)[A]				
	Operation	4.58 (3.50)[A]	8.74 (9.75)[A]	6.71 (0.84)	6.61 (0.84)	0.103	0.933
	After	8.68 (5.40)[B]	14.53 (14.92)[B]	11.44 (1.81)	11.77 (1.81)	-0.332	0.900
	p-value[b] (within group)	0.001	<0.001				

Abbreviations: SD, standard deviation; SE, standard error; RD, rubber dam; EP, EasyPrep.

[a] *Analyzed by ANCOVA adjusted by baseline total CFU.*

[b] *Analyzed by Friedman test followed by Dunn's post-hoc test.*

Different capital letters in the same column in each location of each group indicated statistically significant difference (p<0.05).

During the access opening procedure, the most commonly found bacteria in air samples are gram positive cocci species, followed by gram positive bacilli and gram-negative bacilli. However, gram negative cocci and fungi are not found in these samples. (Fig. 3)

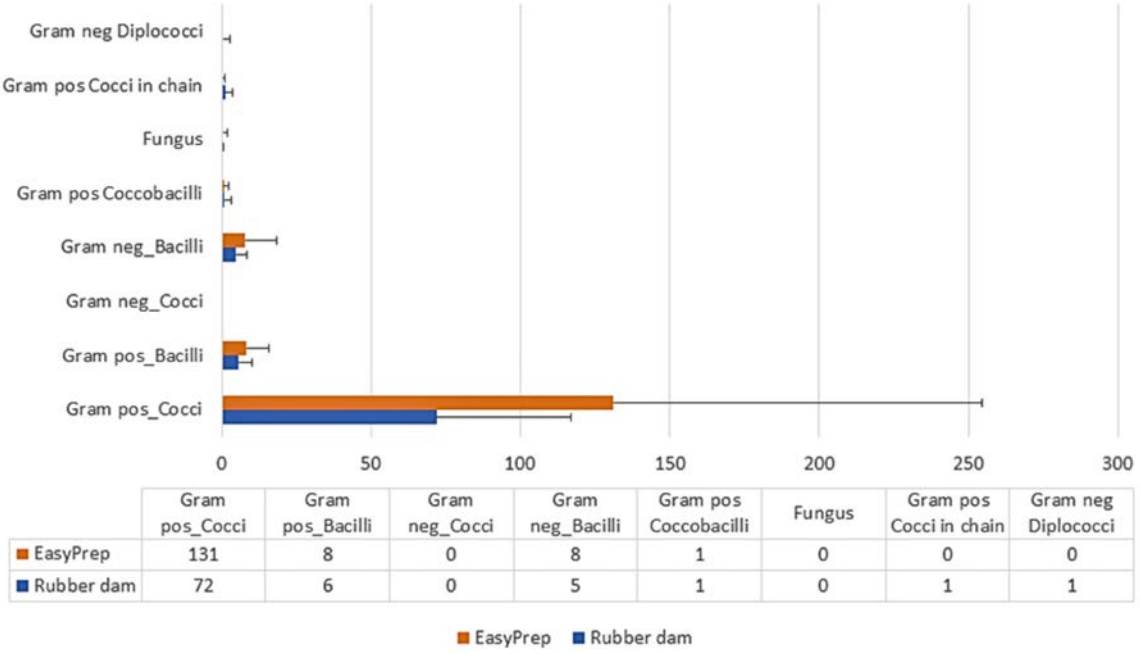

Figure 3 *Bacteria in air during OC procedure categorized by gram stain and morphology. Blue bar indicates bacterial number in Rubber dam group. Orange bar indicates bacterial number in EasyPrep group.*

Species identification by mass spectrometer (Bruker, model: MALDI-Biotyper, Germany). Seventy species were found in the samples. Among these, *Micrococcus luteus* were abundantly found in all samples. Most predominated bacterial species found in each experimental group were demonstrated in figure 4.

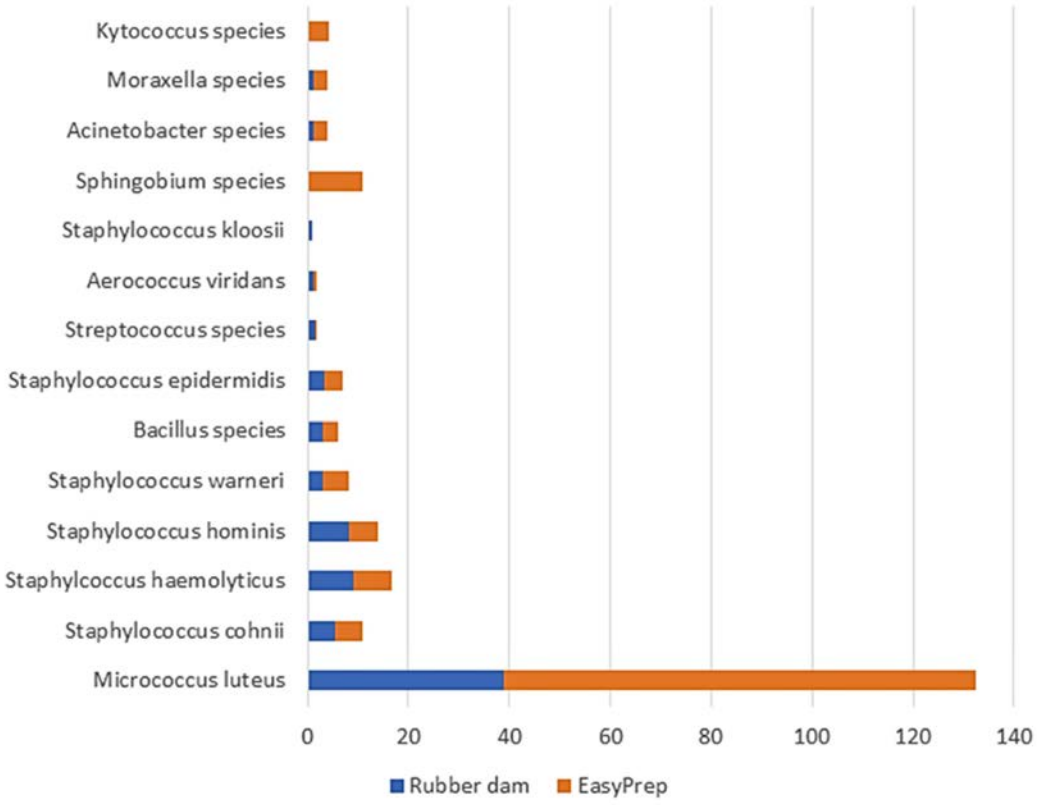

Figure 4 *Most abundant species of bacteria in air sampling during OC procedure identified by proteomic fingerprint. The blue bar indicates the rubber dam group. The orange bar indicates the EasyPrep group.*

Discussion

Our study shows that the endodontic access opening procedure can generate aerosols that disperse bacteria in the air. However, the use of conventional aerosol control measures such as rubber dam and HVE, or hand-free high-volume evacuation devices like EasyPrep, can significantly reduce the amounts of dental aerosols produced during the procedure. Although the average amount of bacteria in the air from the EasyPrep group was slightly higher than the rubber dam group, there was no statistically significant difference found between the two methods. This data corresponds to a study that utilized an intraoral hand-free high-volume evacuation suction device called Mr. Thirsty. [18] The study found that Mr. Thirsty performed similarly to HVE during the ultrasonic scaling procedure. This finding is consistent with a previous study by Kimmerle *et al.*[19], which compared the bacterial load in a multi-chair dental clinic, a single chair treatment room, and a public area. The study found no significant difference in microbial counts between the different locations in these dental treatment units. Another study by Mirhoseini *et al.,*[20] assessed the level of bacterial contamination of air and surfaces in different wards of the educational clinic. The study found no significant difference between the five active studied wards of the dental school clinics.

In endodontic procedures, rubber dams and HVE are the gold standard measures to control both aerosol and contamination in the field of operation. During the endodontic access opening procedure, in some atypical cases, the placement of a rubber dam cannot be done easily. In such situations, hand-free high-volume evacuation devices like EasyPrep can be an alternative to control aerosol. Our experience shows that EasyPrep is easy to use, even in situations where putting a rubber dam poses a difficulty. To use this device, the HVE of the dental unit

is directly connected to EasyPrep. The treatment can be performed by the operator alone, without the need for an assistant to hold the HVE. Due to the design of the device, water is drawn in from the superior and inferior edges of the backside, which exposes some of the gingival tissue and oral mucosa. It's interesting to note that the small leakage of oral fluid in EasyPrep may have contributed to the higher detection of *Micrococcus luteus*, which is a predominant oral flora species, in comparison to the rubber dam group. However, when a rubber dam is used, the tooth is completely isolated from other soft tissue and an HVE is placed in front, close to the tooth to prevent contamination. It should be noted that EasyPrep has a limitation in that it is not suitable for patients who have a limited of jaw opening.

Regarding the standard infection control protocol in post COVID-19 pandemic, during data collection in the clinic, it was recommended that patients rinse their mouth with antiseptic mouthwash. As part of this study, all patients rinsed their mouths with 0.23% povidone mouthwash for 30 seconds before the procedure. It is believed that this step may reduce the viability of micro-organisms collected from air samples during and after the operation to be less than the actual number.

This study utilized the settle plate technique to collect air samples and used nonselective bacterial growth media (blood agar) and cultured the samples in an aerobic condition. This technique is advantageous due to its low cost and not requiring special equipment. However, one should consider its limitations, such as the number of bacteria on the plate not being an exact representation of the number in the air, as only the bacteria that drop off by velocity above the plate are counted. Despite this limitation, the settle plate technique is an appropriate measure to determine the likelihood of surface contamination by bacteria in the air.

In this study, the air samples were collected by settle plate technique for 30 minutes at three different time points by following a previous study by Zemouri *et al.*[21] This technique can collect predominantly particles larger than 5 micrometers due to their high setting velocities.[22] Particles less than 5 micrometers are less likely to settle onto the plate within the limited time. The settle plate technique seems like an effective method for collecting air samples, although it does have some limitations in terms of the size of particles it can collect. It is important to acknowledge these limitations and consider them when interpreting the results of the study.

In this study, MALDI-TOF MS was used to identify bacteria instead of conventional methods due to the equipment being available at King Taksin Memorial Hospital. Advantages of using MALDI-TOF MS include high throughput, low reagent cost, ease of use, ability to analyze as little as a portion of a single colony, and potential to identify organisms that are difficult and laborious to identify by conventional technique. It provides highly specific results within minutes of preparation, while the conventional method takes about 18-24 hours. The limitation of the MALDI-TOF MS technique is that identification of new isolates is possible only if the spectral database contains peptide mass finger-prints of the type strains of specific genera/species/subspecies/strains. These may improve over time as spectral databases expand. The limitation also includes the high-cost of maintenance and of service.[23-24] The comparative study about the advantage of MALDI-TOF MS over biochemical-based phenotyping for microbial identification showed that at the genus level, both MALDI-TOF MS and based systems showed the lowest number of false (4%) and approximately 60% correct identifications. In contrast, the biochemical-based systems assigned 25% of the genera incorrectly. The differences were even more apparent at the species level. [25]

It was found that *Micrococcus luteus* was abundant in all air samples. This finding agrees with a previous study, which reported that *Micrococcus luteus* was also the most prevalent microbe detected in a multi-chair dental clinic, a single-chair treatment room, and a non-dental public area.[19] These bacteria are gram-positive cocci, usually considered non-pathogenic bacteria, and are part of the normal microbial flora on human skin. Additionally, they

have been detected in water, soil and mucous membranes, including the oral cavity.[26-27] It is not typically associated with causing diseases in healthy individuals. However, under certain circumstances, *Micrococcus luteus* can become an opportunistic pathogen and cause infections with indwelling catheters.[28] Bacteria that was found next in order in the rubber dam group was *Streptococcus haemolyticus*, one of the coagulase-negative staphylococci. It is increasingly implicated in opportunistic infections in immunocompromised patients, particularly in hospitalized patients and those with medical implants. It causes severe infections such as meningitis, endocarditis, prosthetic joint infections and bacteremia and is frequently in a hospital environment.[29]

Conclusion

This study concludes that there was no statistically significant difference between the EasyPrep and HVE in the amount of aerosol reduction during endodontic access opening procedures in mandibular posterior permanent teeth. Therefore, dentists can utilize hand-free high-volume evacuation devices like EasyPrep to reduce bacterial aerosols during the endodontic access opening procedure, especially in cases where the clinician may find it difficult to properly orient the tooth axis due to the limiting effect of the dam. Once the root canal is located, it is still recommended to place the rubber dam immediately to prevent any further contamination and ensure a successful root canal procedure.

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Conflict of interest

All authors declare no conflicts of interest directly relevant to the content of this article.

Data availability statement

The authors confirm that the data supporting the findings of this study are available within the article. Raw data that support the findings are available from the corresponding author, upon reasonable request.

Author contributions statement

SY, RSA, conceptualization, design, data acquisition and interpretation; SC, statistical analyses; SY, SC, drafted the manuscript; SY, SC, RSA, data curation, critically revised the manuscript. All authors have read and given final approval to the published version of the manuscript.

Reference

1. Harrel SK, Molinari J. Aerosol and splatter in dentistry: A brief review of the literature and infection control implications. ***J Am Dent Assoc*** 2004;135(4):429-37.

2. Micik RE, Miller RL, Mazzarella MA, Ryge G. Study on dental aerobiology:I. Bacterial aerosols generated during dental procedures. ***J Dent Res*** 1969;48(1):49-56.

3. Gralton J, Tovey E, McLaws ML, Rawlinson WD. The role of particle size in aerosolised pathogen transmission: a review. ***J Infect*** 2011; 62(1):1-13.

4. Kumar S, Atray D, Paiwal D, Balasubramanyam G, Duraiswamy P, Kulkarni S. Dental unit waterlines: source of contamination and cross-infection. ***J Hosp Infect*** 2010;74(2):99-111.

5. Yamada H, Ishihama K, Yasuda K, Hasumi-Nakayama Y, Shimoji S, Furusawa K. Aerial dispersal of blood-contaminated aerosols during dental procedures. ***Quintessence Int*** 2011;42(5):399-405.

6. O'Donnell MJ, Boyle MA, Russell RJ, Coleman DC. Management of dental unit waterline biofilms in the 21st century. ***Future Microbiol*** 2011;6(10):1209-26.

7. Zemouri C, de Soet H, Crielaard W, Laheij A. A scoping review on bio-aerosols in healthcare and the dental environment. ***PLoS One*** 2017;12(5): e0178007.

8. Monteiro PM, Carvalho A, Pina C, Oliveira H, Manso MC. Air quality assessment during dental practice: Aerosols bacterial counts in an universitary clinic. ***Rev Port Estomatol Med Dent Cir Maxillofac*** 2013;54(1):2-7.

9. Ahmed MA, Jouhar R. Dissemination of aerosol and splatter in clinical environment during cavity preparation: An *in vitro* study. ***Int J Environ Res Public Health*** 2021;18(7).

10. Bahador M, Alfirdous RA, Alquria TA, Griffin IL, Tordik PA, Martinho

FC. Aerosol generated during endodontic treatment: A special concern during the coronavirus disease 2019 pandemic. ***J Endod*** 2021;47(5):732-9.
11. Boccia G, Di Spirito F, D'Ambrosio F, De Caro F, Pecora D, Giorgio R, *et al.* Microbial Air Contamination in a Dental Setting Environment and Ultrasonic Scaling in Periodontally Healthy Subjects: An Observational Study. **Int J Environ Res Public Health** 2023;20(3).
12. Gulabivala K, Ng Y-L. Endodontics. 4th ed. London: Mosby; 2014. P. 159-160.
13. Cochran MA, Miller CH, Sheldrake MA. The efficacy of the rubber dam as a barrier to the spread of microorganisms during dental treatment. ***J Am Dent Assoc*** 1989;119(1):141-4.
14. Ravenel TD, Kessler R, Comisi JC, Kelly A, Renne WG, Teich ST. Evaluation of the spatter-reduction effectiveness and aerosol containment of eight dry-field isolation techniques. ***Quintessence Int*** 2020;51(8):660-70.
15. Jacks ME. A laboratory comparison of evacuation devices on aerosol reduction. ***J Dent Hyg*** 2002;76(3):202-6.
16. Glickman GN, Vogt MW. Preparation for Treatment. *In Cohen's Pathways of the pulp.* 10th ed. St.Louis: Mosby; 2011 P. 109-110.
17. Dahlke WO, Cottam MR, Herring MC, Leavitt JM, Ditmyer MM, Walker RS. Evaluation of the spatter-reduction effectiveness of two dry-field isolation techniques. ***J Am Dent Assoc*** 2012;143(11):1199-204.
18. Finnerty D. Aerosol and spatter reduction efficacy of Mr.Thirsty and alternative products. Dent Advisor. 2020 Available at www.dentaladvisor.com/pdf-download/?pdf_url=wp-content/uploads/2020/09/RR-140-Mr.-thirsty-Zirc.pdf
19. Kimmerle H, Wiedmann-Al-Ahmad M, Pelz K, Wittmer A, Hellwig E, Al-Ahmad A. Airborne microbes in different dental environments in comparison to a public area. ***Arch Oral Biol*** 2012;57(6):689-96.
20. Mirhoseini SH, Bayani M. Evaluation of the bacterial contamination of air and surfaces in different dental environments. ***Int J Environ Health Engineering*** 2022;11(1) doi:10.4103/ijehe.ijehe_14_21.
21. Zemouri C, Volgenant CMC, Buijs MJ, Crielaard W, Rosema NAM, Brandt BW. *et al.* Dental aerosols: microbial composition and spatial distribution. ***J oral microbiol*** 2020;12 doi:10.1080/20002297.2020.1762040.
22. Manibusan S, Mainelis G. Passive bioaerosol samplers: a complementary tool for bioaerosol research. a review. ***J Aerosol Sci*** 2022;163 doi:10.1016/j.jaerosci.2022.105992.
23. Branda JA, Kiss K, Fritsche TR, Kus J, Burnham C-A, Mingle L, *et al.* M58 Methods for the identification of cultured microorganisms using Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry. 1st ed. Pennsylvania; 2017. P. 4-5.
24. Singhal N, Kumar M, Kanaujia PK, Virdi JS. MALDI-TOF mass spectrometry: an emerging technology for microbial identification and diagnosis. ***Front Microbiol*** 2015;6 doi:10.3389/fmicb.2015.00791.
25. Urwyler SK, Glaubitz J. Advantage of MALDI-TOF-MS over biochemical-based phenotyping for microbial identification illustrated on industrial applications. ***Lett Appl Microbiol*** 2016;62(2):130-7.
26. Hetem DJ, Rooijakkers SH M, Ekkelenkamp MB. Staphylococci and Micrococci. In Infectious Diseases. 4th ed. Amsterdam: Elsevier; 2017. P. 1522.
27. Kocur M, Kloos WE, Schleifer KH. The genus Micrococcus. In The Prokaryotes. 3rd ed. pp 961-971. New York: Springer, 2006.
28. Zhu M, Zhu Q, Yang Z, Liang Z. Clinical characteristics of patients with Micrococcus luteus bloodstream infection in a Chinese tertiary hospital. ***Polish J Microb*** 2021;70(3) DOI:10.33073/pjm-2021-030.
29. Czekaj T, Ciszewski M, Szewczyk EM. Staphylococcus haemolyticus – an emerging threat in the twilight of the antibiotics age. ***Microbiology*** 2015;161(11):2061-8.