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Fluorescent and Stereomicroscopic Evaluation of Splatter and Settled Aerosols Generated during Three Different Orthodontic Procedures: An in Vitro Study

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ABSTRACT

Objectives: This study evaluated the magnitude of generated aerosols and the extent and intensity of splatter during three orthodontic procedures: orthodontic bonding, motorized interproximal reduction, and debonding and clean-up of adhesive remnants

Materials and Methods: Ten extracted teeth were mounted in a phantom jaw. Acridine orange dye was injected into the water irrigation reservoir. For each procedure, 24 grade-I filter paper discs at 2,4,6,8,10 and 12 o'clock positions at 1, 2,3, and 4ft distances, and 5 additional discs were placed on the operator's face shield and right arm. After each procedure, the filter papers were left in place and the operator remained in his position for 30 minutes. The filter papers were analyzed for the amount and concentration of acridine orange dye using a stereomicroscope and a fluorescent microscope.

Results: Maximum contamination occurred at the 4 o'clock position at 1ft and 2ft. Minimum was at the 10 o'clock position at 1ft distance in all procedures. Contamination of filter papers was found to be maximum on the operator's face shield and minimum on the operator's right arm for all three procedures. The intensity of contamination was similarly maximum at 1ft. distance at the 4 o'clock and 6 o'clock positions for the first procedure and the at 4 o'clock position for the second and third procedures. It was equally minimum at 12, 8 and 10 o'clock positions at 1ft distance in all 3 procedures.

Conclusion: Orthodontic procedures produce localized contamination, highlighting the need for protective equipment for the operator, assistant and patient.

Keywords: Orthodontics; Dental Debonding; Aerosol

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INTRODUCTION

Aerosol generation is inevitable during routine dental procedures such as tooth preparation, oral prophylaxis, and oral surgery that involve the use of high-speed handpieces and ultrasonic scalers. The heat generated due to the friction between the device and tooth surface can cause pathological changes in dental pulp [1]. Therefore, to prevent overheating, it is a general consensus to use water coolant when performing dental procedures. The coolant used with highspeed dental instruments generates aerosols. When combined with body fluids in the oral cavity, such as blood and saliva, bioaerosols are created, which are commonly contaminated with the bacteria, fungi, and viruses, and have the potential to

float in the air for a considerable amount of time and be inhaled by the dentist or other patients [2]. Therefore, along with the hazardous effects of aerosols generated by the water coolant, the potential infection threat of the splatter droplets must also be considered.
The coronavirus disease-2019 pandemic has posed unique challenges to orthodontic profession by adversely impacting provision of in-office orthodontic care due to the risks of infection transmission through splatter and aerosols in aerosol-generating procedures (AGPs) [3]. Aerosols remain in the air for a long period of time even after the completion of dental procedure, and have the potential risk of entering the respiratory passages. Splatter evaporates, leaving smaller particles called droplet nuclei, which can carry bacteria and viruses and transmit various diseases such as the severe acute respiratory syndrome (SARS) and tuberculosis [4]. The risk of infection is not only high for the dentist, but also for the dental team and the patient.
Studies have shown that SARS-CoV-2, similar to SARS coronavirus, can go airborne in the laboratory settings [4]. If the same is true in non-laboratory settings, it means that aerosols of the SARS-CoV-2 suspend in the air for hours, and may be inhaled before sitting on surfaces [5]. Guidelines of the Center for Disease Control and Prevention necessitates the need for social distancing, personal protective equipment (PPE), N-95 respirators, hand hygiene, and other equipment-based considerations as means of transmission-based precautions from SARS Cov-2. In this light, assessment of the efficacy of these measures in protecting the dental personnel from the risk of infection through AGPs and splatter is important.
Several methodologies have been used to evaluate dental aerosols and splatter. These include the use of tracer dyes, measurement of bacterial contamination, and the use of optical particle-counting instruments [6]. AGPs in the field of orthodontics involve the use of 3-in-1 air and water spray after conventional etching for bonding, motorized interproximal reduction, and removal of adhesive after bracket debonding. Considering the importance of infection transmission prevention, especially in the current era of SARS-Cov 2 infection, the present study aimed to evaluate the amount of aerosol generation and the extent of splatter in different directions during orthodontic bonding with the conventional etching system, motorized interproximal reduction, and removal of adhesive after debonding.

MATERIALS AND METHODS

Ethical approval for this study was obtained from the institutional ethics committee (ITSCDSR/L/2019/156).
The study was conducted in a closed room without any ventilation. Ten extracted teeth (second premolar to second premolar teeth) were mounted in a phantom jaw simulating the maxillary arch, which was attached to a dental mannequin. The mannequin was placed on a dental chair inclined at a 45-degree angle relative to the floor. Acridine orange dye in the form of an orange red odorless powder (Himedia Laboratories, India) was injected into the water irrigation reservoir supplying the 3-way air-water spray and the air rotor unit.
Twenty-four grade-I qualitative white filter paper discs made from cotton cellulose fibers with 9.0cm diameter and 0.2mm thickness were used for each procedure (Fig 1).

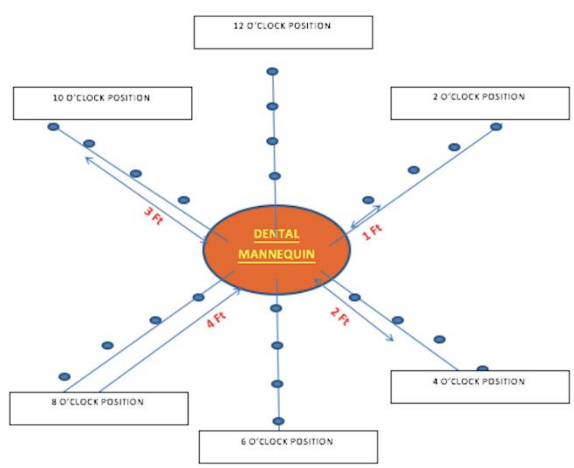

Fig 1. Placement of filter papers for capturing splatter and settled aerosols

They were placed at 12 o'clock, 2 o'clock, 4 o'clock, 6 o'clock, 8 o'clock and 10 o'clock positions in relation to the dental mannequin. At each position 4 filter papers were placed at the distances of 1, 2,3, and 4ft respectively. The study methodology was adopted from a study by Allison et al, [7] on placement of filter papers in the immediate environment. Five additional filter papers were placed on the face shield, wrist and shoulder on both sides of the operator. The operator performed the procedures sitting at the 9 o'clock position with a high-vacuum suction placed in the mannequin's oral cavity at all times. After completion of each procedure, the filter papers were left in place and the operator remained in his position for a period of 30 minutes to allow the aerosols to settle down (Fig 2). The filter papers were then replaced with new ones for the next procedure. The collected filter papers were then analyzed for the absorbed amount and concentration of acridine orange dye using a stereomicroscope (Olympus SZX7, Olympus Corporation, Japan) and a fluorescent microscope (Olympus U-CTR30-2, Olympus Corporation, Japan), respectively.

Fig 2. Set-up for the study depicting position and placement of filter papers

The procedure 1 included orthodontic bonding using conventional acid etching involving the use of a 3-way water spray for etchant removal with a high-vacuum suction in place. The procedure 2 included motorized interproximal reduction where water spray was used intermittently for 5 seconds after performing each interproximal reduction of 0.5mm between two teeth. The procedure-3 included debonding of orthodontic brackets and clean-up of adhesive remnants from the tooth enamel surface using tungsten carbide bur under continuous water spray from the air rotor handpiece.

Analysis of filter papers:

Quantitative assessment: Each filter paper was divided into 4 areas: upper left, upper right, lower left, and lower right to be individually visualized under a stereomicroscope (Olympus SZX7, Olympus Corporation, Japan) for the acquired spots of acridine orange dye. A photograph of the visualized area was taken using the microscope camera (Fig 3).

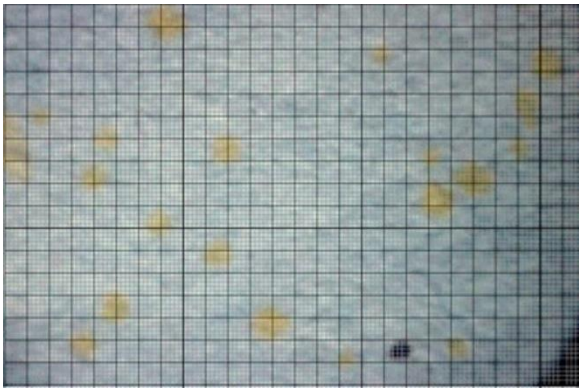

Fig 3. Filter paper visualized under a stereomicroscope with a transparent grid placed digitally

The contamination area was calculated after superimposing a transparent grid with $1cm^2$ squares digitally over the acquired images of the filter paper. If a square had at least one orange area, it was counted as contaminated. The area of contamination was measured by counting the number of $1cm^2$ contaminated squares and the sum of contaminated squares from all the visualized areas of a particular filter paper was calculated to obtain the total contaminated area. This method of calculating the area of contamination of filter papers was adopted from a study by Veena et al, [8] to evaluate the dissemination of aerosols and splatter during ultrasonic scaling.

Qualitative assessment: The areas containing maximum contamination were later analyzed under a fluorescent microscope (Olympus U-CTR30-2, Olympus Corporation, Japan) for estimation of the intensity of the acridine dye absorbed by the filter paper (Fig 4). Each contaminated area was classified as mild, moderate, or severe in terms of intensity of fluorescence visualized under the fluorescent microscope.

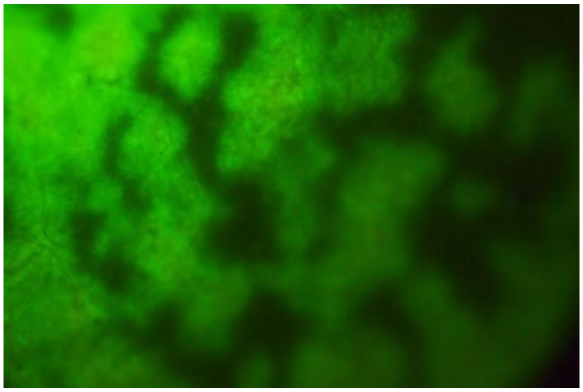

Fig 4. Filter paper visualized under a fluorescent microscope

Statistical analysis:

The data were collected, compiled, tabulated, and subjected to statistical analysis using SPSS 16.0 (SPSS Inc., Chicago, IL, USA). The normality of the numerical data was checked using the Shapiro-Wilk test. The data for surface area contamination of filter papers under the stereomicroscope were found to be normally distributed ($P>0.05$) while the data for the intensity of contamination were not ($P<0.05$). The data for surface area contamination of filter papers for each procedure at different positions and distances were analyzed by ANOVA followed by a post-hoc test for intergroup comparison. As the data for intensity of contamination of filter papers under the fluorescent microscope were not normally distributed, the Kruskal Wallis non-parametric test was used to compare the mean rank for intensity of contamination of filter papers for various positions at varying distances. The Mann-Whitney U test was used to compare each two different positions at a particular distance for each procedure.

RESULTS

Table 1 presents the descriptive findings and comparison of surface area of contamination of filter papers under a stereomicroscope during different procedures at different distances and positions. The maximum surface area of contamination was observed at the 4 o'clock position for all procedures at 1ft. and 2ft. distances. For all three procedures, the mean surface area of contamination of filter papers at 1ft. distance was found to be maximum at the 4 o'clock position. There was a statistically significant difference between the 4 o'clock and 6 o'clock positions with all other positions ($p\leq0.001$) for all three procedures. The mean surface area of contamination for all three procedures at 1ft. distance was found to be minimum at the 10 o'clock position. The difference for this position was statistically significant with the 4 o'clock and 6 o'clock positions for the procedures 1 and 2 ($p\leq0.001$), and with the 2 o'clock($p\leq0.001$), 4 o'clock ($p\leq0.001$) and 6 o'clock ($p\leq0.05$) positions for the procedure 3. Intragroup comparison of the mean surface area of contamination of filter papers at 1ft. distance showed a statistically significant difference among all the positions for all the procedures.

At 2ft. distance, contamination of filter papers was found only at the 4 o'clock and 6 o'clock positions for all three procedures, and the difference was statistically significant between these positions for the procedures 1($p=0.005$) and 3($p=0.004$), and insignificant for the procedure 2($p=0.152$).

At 3ft. distance, contamination of filter papers was found only for the procedure 3 at the 4 o'clock and 6 o'clock positions. The difference in the mean area of contamination of filter papers between these positions was statistically significant ($p\leq0.001$).

Table 2 shows the frequency distribution and comparison of the intensity of contamination of filter papers under the fluorescent microscope during different procedures at different distances and positions. For the procedure 1, the mean rank for the intensity of contamination of filter papers under the fluorescent microscope at 1ft. distance was found to be maximum at the 4 o'clock and 6 o'clock positions with the difference being statistically significant when compared with the 12 o'clock, 8 o'clock, and 10 o'clock positions($p\leq0.001$). There was low intensity observed at the 12, 8 and 10 o'clock positions at 1ft. distance. For both procedure 2 and procedure 3, the mean rank for the intensity of contamination of filter papers at 1ft. distance was found to be maximum at the 4 o'clock position with the difference being statistically significant when compared with the 12 o'clock, 8 o'clock, and 10 o'clock positions.

Table 1. descriptive statistics and comparison of the surface area of contamination of filter papers under a stereomicroscope during different procedures at different distances and positions

Procedure	Distance (ft.)	Position (o'clock)	Mean ± SD surface area (cm²)	95% Confidence interval		P value (distance × position)	Pairwise comparison (o'clock)
				Lower bound	Upper bound		
1	1	12	6.00±3.00	1.43	10.56	0.00	4, 6
		2	11.00±2.00	6.43	15.56		4, 6, 10
		4	41.33±5.85	36.76	45.89		12, 2, 6, 8, 10
		6	26.00±5.29	21.43	30.56		12, 2, 4, 8, 10
		8	10.33±1.15	5.76	14.89		4, 6
		10	2.33±1.52	-2.23	6.89		4, 6
	2	4	14.66±3.05	11.14	18.19	0.005	6
		6	4.33±0.57	0.80	7.85		4
2	1	12	5.00±1.00	-0.03	10.03	0.00	4, 6
		2	7.00±2.00	1.96	12.03		4, 6
		4	35.00±7.54	29.96	40.03		12, 2, 6, 8, 10
		6	22.66±5.68	17.63	27.69		12, 2, 4, 8, 10
		8	7.66±1.15	2.63	12.69		4, 6
		10	2.66±0.57	-2.36	7.69		4, 6
	2	4	3.33±1.52	1.48	5.18	0.152	None
		6	1.66±0.57	-0.18	3.51		None
3	1	12	8.00±1.00	4.38	11.61	0.00	2, 4, 6
		2	15.00±1.00	11.38	18.61		12, 4, 6, 8, 10
		4	48.66±6.42	45.04	52.28		12, 2, 6, 8, 10
		6	38.66±2.08	35.04	42.28		12, 2, 4, 8, 10
		8	9.00±1.00	5.38	12.61		2, 4, 6
		10	7.00±1.00	3.38	10.61		2, 4, 6
	2	4	21.66±2.51	18.33	25.00	0.004	6
		6	11.33±1.52	7.99	14.67		4
	3	4	7.33±1.52	5.48	9.18	0.006	6
		6	2.33±0.57	0.48	4.18		4

*significant differences

Intragroup comparison for the mean surface area of contamination of filter papers under a stereomicroscope showed statistically significant differences among all positions for each procedure (Table 3). For different positions of filter papers, the mean surface area of contamination of filter papers under the stereomicroscope was found to be maximum on the operator's face shield for all three procedures and had a statistically significant difference with the rest of the positions (p≤0.001).

The mean surface area of contamination of filter papers under the stereomicroscope was found to be minimum at the operator's right arm for all three procedures. There was an intragroup statistically significant difference for this position with the rest of the positions in the procedure 3 (Face shield and left hand wrist (p≤0.001)) (Right hand wrist p=0.041) (left arm (p≤0.001)). However, the difference for this position with the operator's right-hand wrist was not statistically significant for the procedure 1 and procedure 2.

Table 2. Frequency distribution and comparison of the intensity of contamination of filter papers under a fluorescent microscope during different procedures at different distances and positions

Procedure	Distance (ft.)	Position (o'clock)	Intensity grade (% within position)			Mean Rank	P value	Pairwise comparison* (o'clock)
			Grade-1	Grade-2	Grade-3			
1	1	12	3 (100)	0	0	5.50	0.012	2, 4, 6
		2	1 (33.3)	2 (66.7)	0	10.17		12, 8, 10
		4	0 (0)	1 (33.3)	2 (66.7)	15.17		12, 8, 10
		6	0 (0)	1 (33.3)	2 (66.7)	15.17		12, 8, 10
		8	3 (100)	0	0	5.50		2, 4, 6
		10	3 (100)	0	0	5.50		2, 4, 6
	2	4	1 (33.3)	2 (66.7)	0	3.50	1.00	none
		6	1 (33.3)	2 (66.7)	0	3.50		none
	3	4	0	1(100)	0	1.50	1.00	none
		6	0	1 (100)	0	1.50		none
2	1	12	3 (100)	0	0	5.50	0.012	4, 6
		2	1 (33.3)	2 (66.7)	0	10.83		4, 6
		4	0 (0)	1(33.3)	2 (66.7)	16.17		12, 2, 8, 10
		6	0 (0)	3 (100)	0	13.50		12, 2, 10
		8	3 (100)	0	0	5.50		4
		10	3 (100)	0	0	5.50		4
	2	4	1 (33.3)	2 (66.7)	0	4.50	0.114	none
		6	3 (100)	0	0	2.50		none
	3	4	1 (100)	0	0	1.50	1.00	none
		6	1 (100)	0	0	1.50		none
3	1	12	3 (100)	0	0	5.50	0.012	2, 4, 6
		2	1 (33.3)	2 (66.7)	0	10.83		12, 8, 10
		4	0 (0)	1 (33.3)	2 (66.7)	16.17		12, 8, 10
		6	0 (0)	3 (100)	0	13.50		12, 8, 10
		8	3 (100)	0	0	5.50		2, 4, 6
		10	3 (100)	0	0	5.50		2, 4, 6
	2	4	2 (66.7)	1 (33.3)	0	4.00	0.317	none
		6	3 (100)	0	0	3.00		none
	3	4	1 (100)	0	0	1.50	1.00	none
		6	1 (100)	0	0	1.50		none

*significant differences

The intensity of contamination of filter papers under the fluorescent microscope was found to be equally high on the operator's face shield and left-hand wrist for the procedure 1 (12.50) and procedure 3 (12.0) while it was maximum on the operator's left-hand wrist for the procedure 2. It was equally minimum for the rest of the positions for the procedure 1 (5.0), equally minimum for the rest of the positions for the procedure 2 (7.0) and similarly minimum on the operator's right-hand wrist and right arm for the procedure 3 (4.50).

Intragroup comparison of different positions of filter papers for the intensity of contamination showed a statistically significant difference for the procedures 1 (P=0.007) and 3 (P=0.021) and an insignificant difference for the procedure 2 (P=0.071; Table 4).

Table 3. Comparison of surface area of contamination of filter papers under a stereomicroscope during different procedures for different operator positions

Procedure	Position	Mean ± SD area(cm^2)	95% Confidence interval Lower Bound	Upper Bound	Pairwise comparison*	P value
1	OFS	21.0±3.60	12.04	29.96	ORHW, ORLW, ORA, OLA	0.000
	ORHW	9.0±1.73	4.70	13.30	OFS, OLHW	
	OLHS	15.33±3.21	7.35	23.32	OFS, ORHW, ORA	
	ORA	6.33±1.52	2.54	10.13	OFS, OLHW. OLA	
	OLA	11.33±2.51	5.08	17.58	OFS, ORA	
2	OFS	15.00±1.73	10.70	19.30	ORHW, OLHW, ORA, OLA	0.000
	ORHW	3.00±1.00	0.52	5.48	OFS, OLHW, ORA, OLA	
	OLHW	10.67±1.52	6.87	14.46	OFS, ORHW, ORA, OLA	
	ORA	1.67± 0.57	0.23	3.10	OFS, OLHW, OLA	
	OLA	6.00±2.00	1.03	10.97	OFS, ORHW, ORA, OLHW	
3	OFS	31.00±4.58	19.62	42.38	ORHW, ORLW, ORA, OLA	0.000
	ORHW	7.67±1.15	4.80	10.54	OFS, ORHW, ORA, OLA	
	OLHW	18.00±1.00	15.52	20.48	OFS, OLHW, ORA, OLA	
	ORA	3.33±1.15	0.46	6.20	OFS, OLHW, ORHW, OLA	
	OLA	12.00±1.00	9.52	14.48	OFS, OLHW, ORHW, ORA	

*significant difference; SD: standard deviation; OFS: operator's face shield; ORHW: Operator's right-hand wrist; ORLW: Operator's left-hand wrist; ORA: Operator's right arm; OLA: Operator's left arm

Table 4. Frequency distribution and comparison of the intensity of contamination of filter papers under a fluorescent microscope during different procedures for different operator positions

Procedure	Position	Intensity Grade (% within Position) Grade -1	Grade -2	Grade -3	Mean Value	P Value	Pairwise comparison*
1	OFS	0	3 (100)	0	12.50	0.007	ORHW, OLHW, ORA, OLA
	ORHW	3 (100)	0	0	5.00		OFS, OLHW
	OLHW	0	3 (100)	0	12.50		OFS, ORHW, ORA, OLA
	ORA	3 (100)	0	0	5.00		OFS, OLHW
	OLA	3 (100)	0	0	5.00		OLHW
2	OFS	3 (100)	0	0	7.00	0.071	none
	ORHW	3 (100)	0	0	7.00		none
	OLHW	1 (33.3)	2 (66.7)	0	12.00		none
	ORA	3 (100)	0	0	7.00		none
	OLA	3 (100)	0	0	7.00		none
3	OFS	0	3 (100)	0	12.00	0.021	ORHW,ORA
	ORHW	3 (100)	0	0	4.50		OFS, OLHW
	OLHW	0	3 (100)	0	12.00		ORHW, ORA
	ORA	3 (100)	0	0	4.50		OFS, OLHW
	OLA	2 (66.7)	1 (33.3)	0	7.00		OFS

*significant difference; OFS: operator's face shield; ORHW: Operator's right-hand wrist; ORLW: Operator's left-hand wrist; ORA: Operator's right arm; OLA: Operator's left arm

DISCUSSION

Dental procedures have always been under scrutiny for high risk of disease transmission. While there are a lot of dental procedures through which cross infection can occur, AGPs have been well documented in the literature to cause both viral and bacterial infections [9]. Settled aerosols, and in turn the splatter, contain contaminated droplets which later become the so called “droplet nuclei” composed of saliva, dried serum, and microorganisms [10]. Tran et al. [11] suggested that some procedures, potentially capable of generating aerosols, are associated with increased risk of SARS transmission to healthcare workers. In the era of SARS CoV-2 pandemic, assessment of aerosol generation during various dental procedures holds importance both for the healthcare workers and patients as well.

The present in vitro study was conducted aiming to assess aerosol generation during three different orthodontic procedures both qualitatively and quantitatively. Orthodontic bonding, interproximal reduction, and debonding of orthodontic brackets were assessed as these procedures are likely to generate aerosols.

Mirhoseini et al. [12] suggested that microbiological assessment of air in indoor environments is time-consuming and labor-intensive. Thus, the potential dispersion of bacterial and fungal aerosols in the indoor air of dental offices can be estimated via monitoring of particulate matter concentrations. Methods to measure particle sizes have consistently found pathogens in small particles (i.e., <5 μm) [13]. Passive sampling techniques have been proven to be effective to quantify the settled aerosols and splatter. Holliday et al, [14] in their study used filter papers at various positions as a method to assess settled aerosol contamination. The same methodology was followed by Llandro et al, [6] to assess splatter and settled aerosol particles. The present study relied on this passive sampling technique using placement of cotton-cellulose filter papers around a dental mannequin and over various parts of the operator wearing PPE, which was consistent with the study by Allison et al [7]. Tang et al. [15] stated that dental virtual simulators provide a great way to enhance dental education by simulating the natural oral environment. Similarly, a phantom jaw mounted in a dental mannequin was used in the current study to assess the amount of splatter and settled aerosols. But on the contrary, Roy et al. [16] assessed the reliability of dental mannequin to simulate the natural environment and reported slightly significant differences.

The filter papers placed around the mannequin aimed to determine the distance of splatter and settled aerosols, while the ones placed over the operator aimed to highlight the importance of PPE. The filter papers were quantitatively assessed for the area of settled aerosol particles and splatter at the microscopic level by using a stereomicroscope and qualitatively by using a fluorescent microscope. However, Allison et al. [7] analyzed samples using photographic image analysis and spectrofluorometric analysis. Llandro et al. [6] analyzed samples using digital image analysis and spectrofluorometric analysis. Visualization of the particles under fluorescent microscope by using fluorescein dye with the passive sampling technique has been well documented in the literature. Acridine orange is the fluorescent dye routinely used for histological analysis [17] and was thus used in the present study to visualize contamination of filter papers by the settled aerosols and splatter.

Llandro et al. [6] evaluated aerosol and/or splatter contamination during an orthodontic debonding procedure and concluded that orthodontic debonding procedures are low risk for aerosol generation, but localized splatter is likely. There has been a debate on the use of coolant during interproximal reduction of teeth to gain space. Omer and Sanea [18] concluded that interproximal reduction with/without a coolant is a safe procedure for dental pulp in teeth with medium dentin thickness. On the contrary, Sehgal et al. [19] stated that frictional heat produced with different stripping techniques increased the pulpal temperature; therefore, caution is advised during this procedure. A coolant spray can limit the pulpal temperature rise. Thus, taking into account the worst-case scenario for aerosol generation, water spray was used intermittently with interproximal reduction of tooth surfaces in the present study.

The present results showed that the mean surface area of contamination at 1ft. distance was found to be maximum at the 4 o'clock position during adhesive removal after bracket debonding, followed by bonding and interproximal reduction. This finding was consistent with high production of fine aerosols during the use of air rotor for any routine dental procedure. Similar findings were reported by Micik et al, [20] who showed that rate of aerosol production during the use of air rotor handpiece was the highest. As the procedure was done by a right-handed operator, these results may be attributed to close proximity to the right hand of the operator. According to a similar study by Veena et al, [8] contamination was found to be the highest at 1ft. in the 4 o'clock position, similar to the present results. But on the contrary, a study by Revathi and Muralidharan [21] reported that maximum contamination was seen at 2ft. away followed by 1ft. away and 1ft. height. Although the present study did not evaluate other procedures requiring high-speed rotary instruments, Rautemaa et al. [22] showed that contamination was less intense during periodontal and orthodontic treatments (598CFU/m^2/h at >1.5m from the patient) when high-speed rotary and ultrasonic instruments were not used.

For different positions of filter paper over the operator, the mean surface area of contamination of filter papers under the stereomicroscope was found to be maximum on the operator's face shield. Maximum contamination was observed during debonding and residual adhesive removal and minimum during interproximal reduction. The mean surface area of contamination of filter papers under a stereomicroscope was found to be minimum at the operator's right arm for all three procedures. The contamination on the operator's face shield, arms and wrist highlights the need for suitable PPE and face shield with a face mask. Similar findings were observed by Allison et al, [7] who noted heavy contamination of the operator's visor along with the non-dominant side of the body. On the contrary, Llandro et al. [6] evaluated settled and spattered aerosols during orthodontic debonding procedure. They found maximum spatter on the clinicians' right leg and minimum on their face shield. One possible explanation could be that they added fluorescein to the saliva rather than the water supply. We did not evaluate the leg region for contamination in the present study.

Not much evidence exists in the literature regarding qualitative measurement of contamination. This was evaluated by measuring the intensity of contamination of filter papers by acridine orange dye and visualization of the intensity using a fluorescent microscope.

For all 3 procedures, heaviest aerosol contamination was observed at the 4 o'clock and 6 o'clock positions at 1ft. distance with the difference being statistically significant when compared with the rest of the positions. It was minimum at the 10 o'clock position at 1ft. distance. Thus, a clinician operating at this position is less likely to be exposed to splatter and settled aerosols generated during the procedures when compared with the rest of the positions. Similar findings were observed by Han et al, [23] who visualized aerosol contamination in a similar manner to the present study. The fluorescence intensity was maximum at the positions which would represent the 4'oclock and 6 o'clock positions in the present study.

In the current study, the maximum intensity of contamination was observed on the operator's face shield and operator's left-hand wrist during the bonding and debonding procedures. Minimum contamination was observed on the operator's right-hand wrist and right arm during the debonding procedure. These findings correlate to the heavy splatter observed in the form of increased surface area.

These results highlight the importance of using barriers in the form of gowns or PPE to cover the operator's body, and face shield to reduce the risk of infection transmission. A study conducted by Nagraj et al. [24] indicated the use of high-volume evacuator, dental isolation combination system, rubber dam, air cleaning systems, disinfectants-antimicrobial coolants as interventions to reduce contaminated aerosols generated during dental procedures to prevent infectious diseases. In the field of orthodontics, use of water-spray syringe for rinsing should be minimized in bonding-related procedures, use of 0.2% chlorhexidine mouthwash should be prioritized to reduce bacterial count, use of tungsten carbide burs should be considered for enhancement of cutting efficiency, and restriction of the duration of the procedure should be practiced as well. It should be noted that efficient sanitization procedures combined with the correct use of these methods and physical barriers can significantly reduce the probability of SARS-CoV-2 being transmitted during dental practice.

A major limitation of this study was the use of a passive technique that measured the amount of splatter and aerosol generation only based on aerosols settled on the filter papers. The number of particles that remained suspended in the air and did not settle down was not taken into account in this study. Van Doremalen et al. [4] reported that SARS-CoV-2 can survive in the air for many hours, causing potential aerosolized transmission. In conjunction to this finding, air samplers/particle counting instruments may be used as a viable option for future research as means of counting the total number of particles generated during dental procedures.

CONCLUSION

The conclusions drawn from this study were:

Maximum surface area of contamination was observed at the 4 o'clock position for all procedures at 1ft. and 2 ft. distances while it was minimum at the 10 o'clock position at 1 ft. distance for all procedures.

Contamination of filter papers was found to be maximum on the operator's face shield and minimum on the operator's right arm for all three procedures.

For all procedures, the mean rank for the intensity of contamination of filter papers under the fluorescent microscope at 1ft. distance was found to be equally maximum at the 4 o'clock and 6 o'clock positions while it was equally minimum at the 12, 8, and 10 o'clock positions at 1ft. distance.

The intensity of contamination of filter papers under the fluorescent microscope was found to be equally maximum on the operator's face shield and operator's left-hand wrist for the procedure 1 and procedure 3 while it was maximum on the operator's left-hand wrist for the procedure 2.

CONFLICT OF INTEREST STATEMENT

None declared.

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