Dentsply Maillefer) possessing the program for WaveOne files instrumentation was used. The flutes of the file were cleaned after every three pecks. On meeting obstruction, the file was removed, the canal irrigated, recapitulated, and the file reintroduced into the canal again. The instrumentation was done until the WaveOne primary file reached the apex.

- *Group 3 (iRace):* Ten specimens were instrumented using iRace rotary files. The root canal was flared using SX file from the universal Protaper (Dentsply Tulsa Dental, Tulsa, OK). For iRace group, an endodontic motor (X-Smart, Dentsply Maillefer) was set at 600 rpm and a torque of 1.5 Ncm. R1 file (15/0.06) followed by R2 file (25/0.04) was used in a sequential manner up to the working length. A 15 K-file was used for recapitulating between each file to prevent the apical blockage.

Measurement of the Extruded Irrigant

Following canal preparation, the solution that was collected in the collecting vials was thoroughly mixed in a Vortex mixer (CM 101 Cyclomixer, Remi, India) for 5 minutes and debris allowed to sediment for 10 minutes. Micropipettes were used to measure the volume of extruded irrigant.

Measurement of the Bacteria

About 0.01 mL of the extruded irrigant was pipetted out of the collecting vial and using a sterile cotton swab the suspension was plated on Mueller Hinton agar plates at 37°C and incubated for 24 hours (Fig. 2). The counting of bacteria was done using surface plating method.[19] In this method, the viable count of the bacteria was calculated by taking the average colony count per plate.

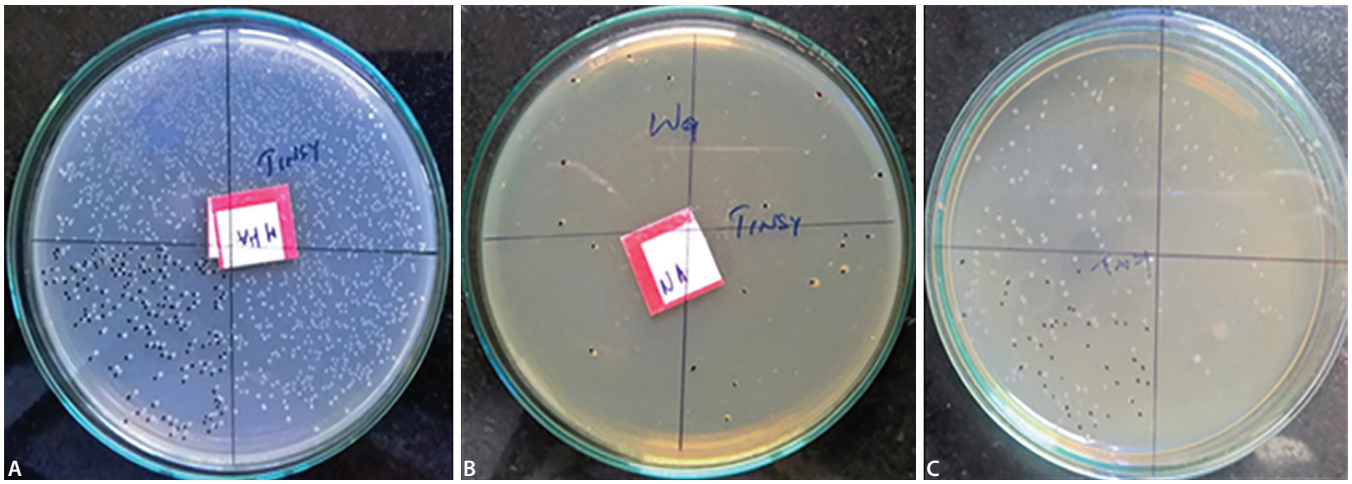
Measurement of the Debris

The root apex was washed using 1 mL distilled water into the collecting vial to procure any debris sticking to the external root surface. The collecting vials were then incubated at 37°C till the irrigant evaporated. The collecting vials were weighed using a 10^{-5} precision electronic balance (MYA 5.3Y microbalance, RADWAG balances, and scales, USA). Three subsequent readings were taken for every sample recording the mean. The amount of apically extruded debris was obtained by subtracting the postoperative weight of the collecting vial from the preoperative weight of the collecting vial.

Data analysis was carried out using statistical package for the social sciences (SPSS) version 16 software. $p < 0.05$ was considered significant. Extrusion of irrigant, debris and bacteria were assessed using the independent Kruskal–Wallis test. Intergroup comparison was done using Mann–Whitney tests to test for significant differences.

Results

The mean value of debris, bacteria, and irrigant extruded (in mg) obtained in each group are shown in Table 1. The maximum debris extrusion was seen in Protaper NEXT group (1.21 mg) followed by iRace group (0.21 mg) and the least in WaveOne group (0.05 mg). Maximum irrigant extrusion was in WaveOne (0.62 mg) and Protaper Next groups (0.62 mg) and least was by iRace rotary system (0.20 mg). The maximum bacteria extruded was in the Protaper Next group (246.40 CFU/mL) followed by iRace group (158.80CFU/mL), while WaveOne group had the least amount of bacteria extrusion (14.60 CFU/mL).



Figs 2A to C: *E. faecalis* growth on Mueller Hinton agar plates. (A) ProTaper NEXT rotary system; (B) WaveOne rotary system; (C) iRace rotary system

Table 1: Descriptive statistics and association between extrusion of irrigant, debris, and bacteria (using independent Kruskal–Wallis test)

	Debris (in mg)	*Volume of irrigant (in mL)*	*Bacteria (CFU/mL)*
ProTaper Next	1.21 ± 2.21	0.62 ± 0.06	246.40 ± 41.41
WaveOne	0.05 ± 0.03	0.62 ± 0.15	14.60 ± 2.76
iRace	0.21 ± 0.09	0.20 ± 0.05	158.80 ± 22.47
Test statistic	25.351	19.561	25.864
Degree of freedom	2	2	2
*p value	<0.001	<0.001	<0.001

*$p < 0.05$ is significant

Table 2: Comparison of apical extrusion of irrigant, bacterial, debris using Mann Whitney analysis between group I (ProTaper NEXT) and group II (WaveOne) and group III (iRace)

		Mann–Whitney statistics	*Wilcoxon W*	*Test statistic*	*Standard error*	*p value*
Group I and II	Debris (mg)	0.000	55	3.790	13.194	0.000
	Irrigant (mL)	44	99	0.457	13.13	0.684
	Bacteria (CFU/mL)	0.000	55	3.791	13.19	0.000
Group II and III	Debris (mg)	100	155	3.791	13.19	0.000
	Irrigant (mL)	0.000	55	3.791	13.19	0.000
	Bacteria (CFU/mL)	100	155	3.787	13.20	0.000
Group I and III	Debris (mg)	2	57	2	3.790	0.000
	Irrigant (ml)	0.000	55	0.000	3.805	0.000
	Bacteria (CFU/mL)	0.000	55	0.000	3.633	0.000

*p <0.05 is significant

Intergroup comparison using Kruskal–Wallis test revealed that the amount of debris, volume of irrigant and bacteria was statistically significant in all the three groups (Table 1). The p value was kept as ≤ 0.05. Mann–Whitney analysis (Table 2) revealed there was a significant difference in the extruded debris and bacteria between Protaper NEXT and WaveOne groups (p = 0.000). However, no significant difference was noted in the amount of irrigant extruded (p =0.684) between ProTaper NEXT and WaveOne. There was a significant difference (p = 0.000) in the amount of debris, bacteria, and irrigant between WaveOne and iRace. A significant difference (p = 0.000) was also noted in the amount of debris, irrigant, and bacteria extruded between Protaper NEXT and iRace group.

Discussion

Mandibular premolars having a single root and the single canal was clinically evaluated and selected using operating loupes. This was done as factors like the presence of more than one canal, and the canal curvature have been known to affect the amount of apical extrusion.[20-22]

Several methods have been cited in the literature to quantify the extrusion of debris and irrigant; however, the method given by Myers and Montgomery[18] in 1991 has been most widely used.[9,23] This method simulates real clinical conditions as it does not allow the operator to visually perceive the apical foramen whilst canal preparation, by making use of amber colored glass vial in which the tooth is suspended, thereby reducing operator bias. However, it has some inherent disadvantages like being unable to replicate periapical resistance. The canals were preflared using an Sx file as coronal flaring has been reported to improve the control over instrument while preparing the apical third of the canal.[24]

The apical patency of the premolars was maintained by using a 10 K-file to achieve standardization of apical diameters. Tinaz et al.[25] have shown that as the apical diameter increases, the debris extrusion also increases, while Lambrianidis et al.[26] has paradoxically reported that greater amount of extrusion with intact apical constriction. Apical preparation was standardized in all the groups by keeping the master apical file size the same (25 mm).

In order to further decrease variability the amount of irrigant was kept constant (3 mL) for all specimens, the depth of irrigation needle insertion was determined at an established level (3 mm from working length); and the irrigant passively injected to minimize any undue forces on the irrigating syringe.[27]

Enterococcus faecalis was used as a bacteriological marker in the present study as they have been found in the oral cavity.[28] They have been implicated in persistent root canal infections owing to their facultative anaerobic nature and have been retrieved from root canals of teeth with post-treatment disease.[29]

Over the years, a variety of NiTi instruments have emerged all aimed at overcoming the limitations encountered in earlier systems. The differences obtained in various studies may be attributed to variation in following characteristics of the instruments: (i) the preparation technique (ii) the varied cross-sectional design of the file and (iii) the apical tapers of the instruments.[14]

The descriptive results of the present study revealed extrusion of debris, bacteria, and irrigant in all the groups regardless of the file design or kinematic motion involved confirm the findings of previous studies. Many earlier studies have affirmed that all instrumentation techniques produce apical extrusions[7,14] reinforcing the fact that it is impossible to clean and shape canal chemo mechanically without causing apical extrusions. Subsequent studies in endodontic literature[9,21,25] have also shown that all engine-driven instruments extruded lesser intracanal bacteria than manual instruments. Reddy and Hicks suggested that the rotational motion produced by engine-driven instruments tended to pack dentinal debris into the flutes of the file and direct them towards the orifice away from the apical foramen thereby reducing their extrusion.[21]

The results from the existing studies are controversial in regard to the type of engine-driven system causing lesser apical extrusions. While some indicated that continuous rotational movement extruded a lesser amount of debris[30-32] others demonstrated a higher amount of debris extrusion with continuous rotation compared to reciprocation motion.[16,33,34]

Hence the null hypothesis regarding WaveOne files extrude lesser debris and bacteria was accepted.

The reason for such a finding can be attributed to the following reasons:

WaveOne files are designed to work in an unequal reciprocating motion which is similar to a mechanized balanced force pressure less technique while the balanced force technique apparently allows better control over the apically extruded debris as suggested by Yared in 2008.[35]

The reciprocating motion squeezes the debris and bacteria into the flutes and carries it out of the canal reducing debris extrusion.

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