

## Original Article

# The Effect of Apical Preparation Size on the Removal of Smear Layer and Organic Debris from Root Canal Wall: An In-vitro Scanning Electron Microscopic Study

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## ABSTRACT

**Introduction:** The study aims to evaluate the effect of apical preparation size on the removal of the smear layer and organic debris from the root canal wall.

**Methodology:** Thirty freshly extracted single-rooted teeth with comparable root canal morphology were selected. Chemo-mechanical preparation of root canals was done with hand K-files up to 20/0.02 and samples were randomly divided into three groups of 10 specimens each, Group-A (control group): Without further chemomechanical preparation; Group-B: Final chemomechanical preparation with rotary nickel-titanium files up to 25/0.04; Group-C: Final chemomechanical preparation with rotary nickel-titanium files up to 30/0.04. Copious irrigation was done with 17% ethylene diamine tetra acetic acid and 5.25% sodium hypochlorite. The data were analyzed via the Mann-Whitney U test with the help of SPSS 17 statistical software.

**Results:** A significant reduction of the smear layer and organic debris was observed when prepared with rotary nickel-titanium files to a final apical size of 30/0.04 compared to the final apical size of 25/0.04 in the apical third.

**Conclusion:** Increasing the size of the apical preparation to 30/0.04 greatly improved the cleanliness of the root canal walls.

**Keywords:** Root canal irrigants, Root canal preparation, Scanning electron microscopy, Smear layer.

## INTRODUCTION

Endodontic treatment's primary goal is to eliminate lesion etiology through effective chemo-mechanical shaping and

cleaning of the root canal space.<sup>1</sup> It facilitates the mechanical removal of necrotic tissue and biofilms, as well as the optimal flow of irrigant for chemical debridement and root canal disinfection.<sup>2</sup> The root structure morphology is not amenable to simple and effective biomechanical preparation. The organic debris and smear layer obstruct the apical seal, potentially leading to reinfection. Neither chemical nor mechanical debridement produces a clinically acceptable result. Inadequate microbial load removal may obstruct successful endodontic treatment, necessitating additional intervention, exacerbating complications, and a poor prognosis.<sup>1</sup>

To deliver irrigant solutions to the most inaccessible areas of the root canal space, the canal must be prepared to a specific apical size.<sup>1</sup> Mechanical instruments facilitate the shaping process, resulting in a wide range of instruments with varying sequences, apical diameters, and tapers. The latest generation of nickel-titanium rotary systems with super-elastic properties has been accepted for improved instrument centering within the root canal lumen.<sup>2</sup>

Apical preparation size influences root canal disinfection, microbial reduction, and healing outcomes.<sup>2</sup> The apical width of preparation is an important aspect of treating root canals and is critical in defining successful root canal debridement.<sup>3</sup> Various studies have shown that increasing the canal size to three files larger than the initial binding leads to efficient apical debridement.<sup>1</sup> However, the actual apical dimension does not always correlate to the first file that binds, and circumferential dentin removal is not possible because instrument designs do not conform to root canal anatomy.<sup>1</sup>

Despite numerous studies assessing the efficacy of various instrumentation systems and techniques in root canal disinfection, there is a paucity of evidence indicating the

efficient removal of the smear layer and organic residues in minimally prepared root canal systems. As a result, scanning electron microscope images of the root canal walls after in-vitro root canal treatment of extracted human teeth were used in this study. Open dentinal tubules in adequately debrided surfaces indicate that irrigant solutions can reach the apical third of the root canal system.

## METHODS

The study was approved by the Institutional Ethics Committee (MGDCH/IEC/2020-21/SS-01). The selection of teeth done by G\*Power 3.1.9.2 software (Heinrich-Heine-Universität Düsseldorf, Germany) was used to calculate the sample size. Samples were randomly divided into three groups of 10 specimens each. Group-A (control group): Without further chemo-mechanical preparation; Group-B: Final chemo-mechanical preparation with rotary nickel-titanium files up to 25/0.04; Group-C: Final chemo-mechanical preparation with rotary nickel-titanium files up to 30/0.04. Three groups of ten teeth were chosen for 80% power and a 0.05 alpha error probability. Thirty single-rooted mandibular premolar teeth with completely formed roots and comparable root canal morphology freshly extracted from people aged 20 to 40 years were chosen. The soft tissues attached to the external surface of the teeth were removed using a curette and stored in 5.25% NaOCl for 1 hour and then placed in a saline solution to be stored. The presence of a single canal (Vertucci's type I) with a mature apex and the absence of root resorption and calcification were confirmed using radiographs. The study excluded teeth with prior root canal treatment, cracks, fractures, and curved canals.

**Preparation of access cavity:** The access cavity was prepared in a standardized manner. The working length was determined by inserting a #10 K file (Dentsply Maillefer, Ballaigues, Switzerland) into the root canal and setting it to be one millimetre shorter than the anatomic apex. The root canals were chemo-mechanically prepared using hand K files (Dentsply Maillefer, Ballaigues, Switzerland) up to ISO size 20/0.02 and irrigated with 2 ml of 5.25% sodium hypochlorite solution (Novo Dental Product, India) between each file change, followed by rinsing with 5 ml of saline to continue the preparation of the apical segment until the apex was reached.

Each canal was shaped using the Hyflex-controlled memory (Coltene Whaledent, USA) rotary nickel-titanium system with the X Smart Endodontic Torque Control Motor (Dentsply Maillefer, Ballaigues, Switzerland) at 500 rpm

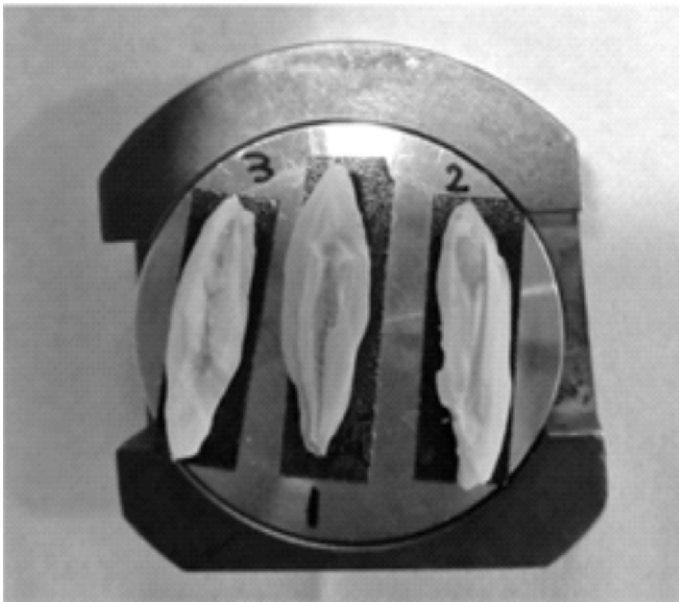
and 2.5 N cm torque according to the manufacturer's instructions in the following sequence: Coronal pre-flaring was done for all the samples using 25/0.08. In 'group B' the canals were instrumented using 20/0.04 followed by 25/0.04 and in 'group C' the canals were instrumented using 20/0.04 followed by 25/0.04 and then finally by 30/0.04.

In each group, the last instrument was considered the master apical file. Each rotary instrument was used for the preparation of five canals and applied for 5s to the working length using the crown-down technique. After each rotary file, the canal was rinsed with 2 ml of 1% NaOCl, delivered by a 30-gauge needle (RC Twents, Prime Dental Products, Mumbai, India) inserted deeply and passively from the coronal to the middle third.

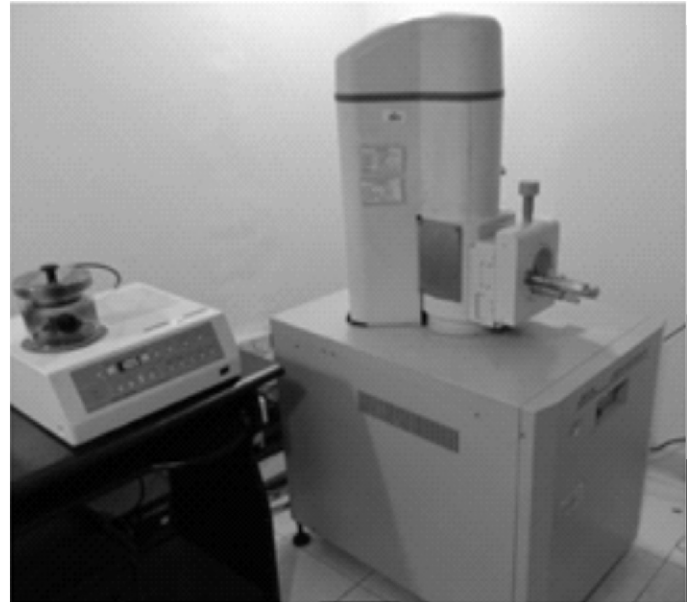
After the completion of instrumentation, the canals were irrigated with 5 ml of 17% ethylene diamine tetra acetic acid (Pulpdent Corporation, Watertown, MA, USA) for 3 minutes followed by rinsing them with 5 ml of 5.25% sodium hypochlorite solution for 5 minutes with a 30-gauge double side-vented endodontic irrigating needle (RC Twents, Prime Dental Products, Mumbai, India) placed 2 mm short of the working length and moved in short vertical strokes of 23 mm amplitude. The rate of irrigation used was 1 mL/min. The total volume of irrigation used in each group was standardized to 10 ml. After a final rinse of the canals with 5 ml of physiologic saline solution, the root canals were dried with 4% absorbent paper points (DiaDent, Europe), and the orifices were sealed with temporary cement.

**Root sectioning and scanning electron microscope imaging:** Each sample was divided into two halves for examination by making shallow longitudinal grooves bucco-lingually with a diamond disc (Toboom Dental Co., Shanghai, China). The roots were then split into mesial and distal halves of each canal with a chisel and mallet to prevent debris from entering during sectioning. These were then dehydrated in a series of ethanol solutions 30-100% for 10 minutes, 50% for 20 minutes, 90% for 30 minutes, and 100% for 30 minutes. The root sections were platinum-sputtered (JFC-1600 Auto Fine Coater, Jeol, Tokyo, Japan) to make the surface electrically conductive after being mounted on copper stubs with double-sided adhesive carbon tape (Figure 1).

To assess approximately the same area of each sample, two defined reference points were marked on each half of the apical third of the root. After that, the samples were evaluated qualitatively using a Scanning Electron Microscope (JSM 6010 LV, Jeol, Tokyo, Japan) at 10 kV



(a)



(b)

**Figure 1: (A) Sectioned tooth samples mounted on copper stubs (B) Scanning electron microscope along with platinum coater.**

**Table 1: Scoring system used to evaluate organic debris and smear layer**

<b>Organic debris</b>	Score 1	Clean canal wall, only very few debris particles
	Score 2	Few conglomerations
	Score 3	For many conglomerations, less than 50% of the canal wall covered
	Score 4	More than 50% of the canal wall covered
	Score 5	Complete or nearly complete coverage of the canal wall by debris
<b>Smear Layer</b>	Score 1	No smear layer and dentinal tubules open
	Score 2	Small amounts of scattered smear layer and dentinal tubules open
	Score 3	Thin smear layer and dentinal tubules partially open
	Score 4	Partial covering with a thick smear layer
	Score 5	Total covering with a thick smear layer

with magnifications of 27X, 1000X, and 2500X. By measuring visible tubules, several photomicrographs were obtained to assess the efficacy of debris and smear layer removal. Scoring was done independently by two different blinded evaluators using the five-point scale at 2500X as described by Hülsmann et al<sup>12</sup> (Table 1).

SPSS software was used to tabulate and analyze the data (SPSS version 18.0, SPSS, Chicago, IL, USA). The inter-evaluator, intra-evaluator agreement, and reproducibility were determined using Cohen's Kappa agreement coefficient. The Kruskal-Wallis and Mann-Whitney U tests were used to compare organic debris and smear layer removal scores across groups. A statistically significant p value of 0.02 was considered.

## RESULTS

The Cohen's kappa value for all groups and subgroups' interobserver agreement was 0.89. Groups B and C had significantly less organic debris and smear layer than group A. The summary scoring for organic debris and smear layer was done based on the image micrographs obtained in each group, which showed that increasing the diameter of the prepared canal reduces the amount of smear layer and organic debris left on the canal walls.

The root canal space in group A (control group), where the apical preparation was done only with 20/0.02, revealed a thick smear layer occluding the dentinal tubules. Group B has a thin smear layer and partially open dentinal tubules after final chemomechanical preparation with rotary

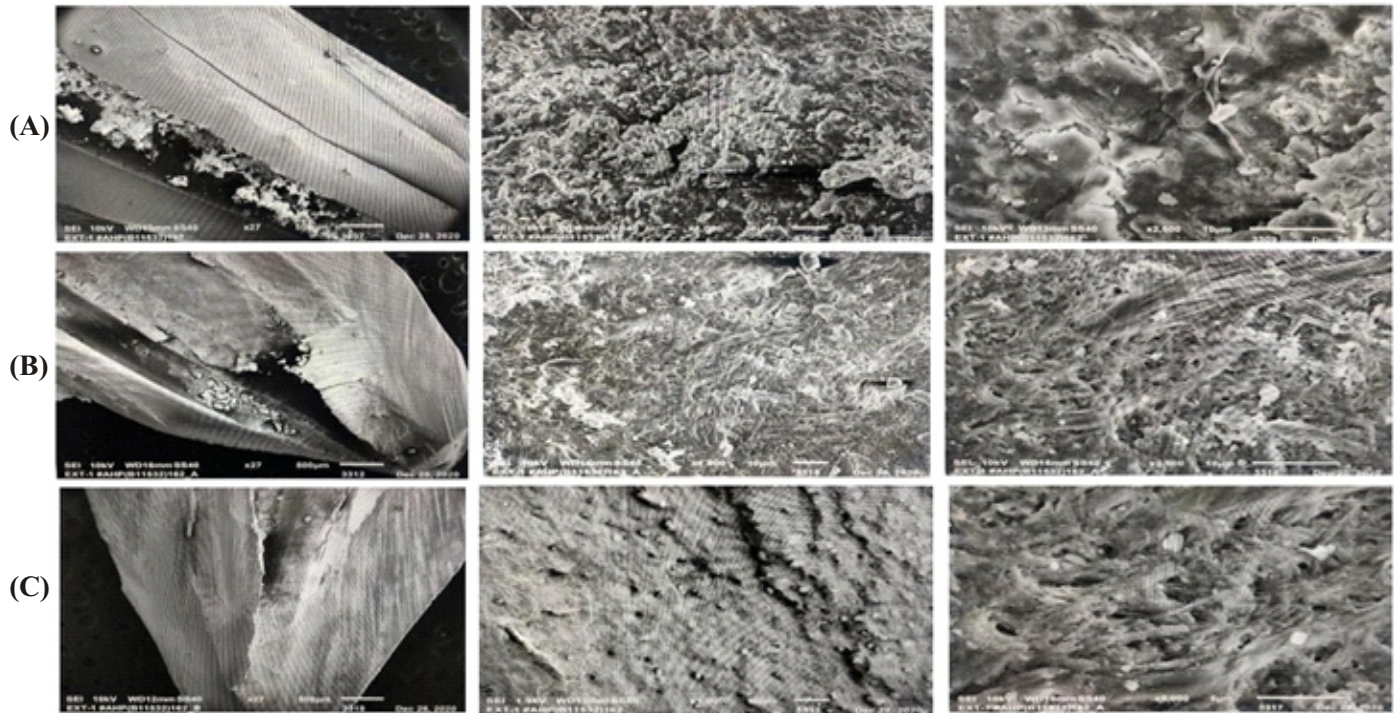


Figure 2: Scanning electron micrographs showing the cross-section of root canal space at 27X, 1000X, and 2500X, respectively.

Table 2: Intra group analysis of organic debris and smear layer scores

Parameter	Groups	Apical preparation size	Mean	S.D.	SE of mean	p value
<b>Organic debris scores</b>	Group A	Control	4.8	0.422	0.133	< 0.005
	Group B	25/0.04	3.6	0.516	0.163	< 0.005
	Group C	30/0.04	2.7	0.483	0.153	< 0.005
<b>Smear layer scores</b>	Group A	Control	4.7	0.483	0.153	< 0.005
	Group B	25/0.04	3.4	0.524	0.133	< 0.005
	Group C	30/0.04	2.3	0.483	0.153	< 0.005

SD: Standard deviation; SE: Standard error

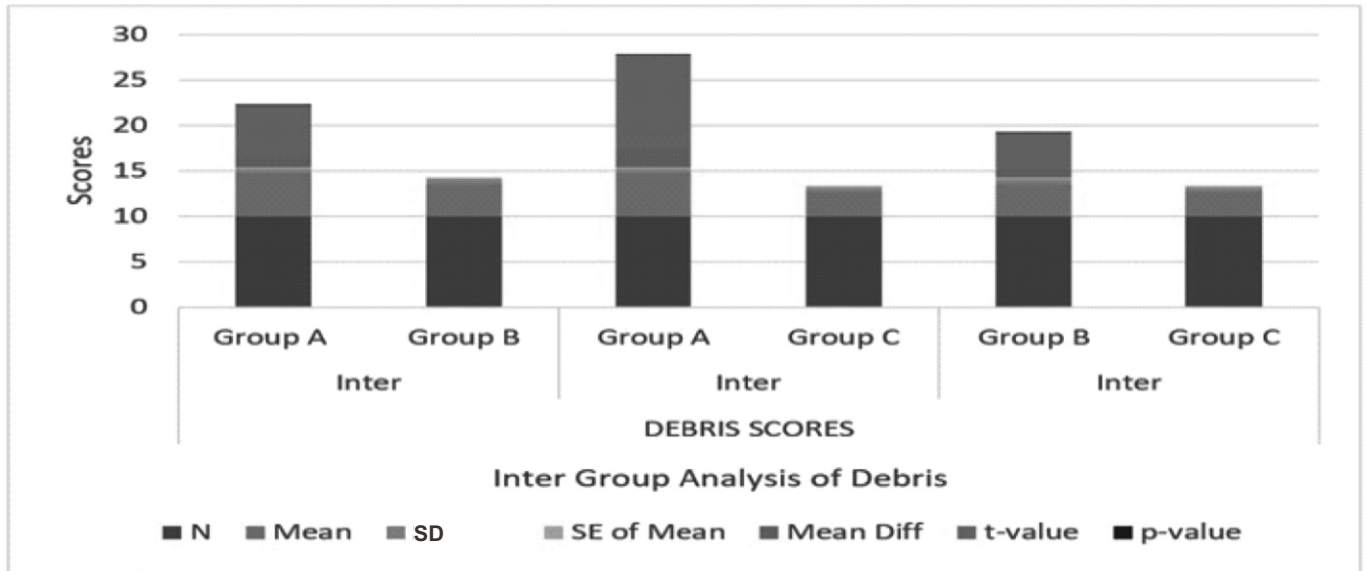
nickel-titanium files up to 25/0.04, whereas Group C has a clean root canal and open dentinal tubules after final chemomechanical preparation with rotary nickel-titanium files up to 30/0.04 (Figure 2).

Intergroup comparison analysis for organic debris and smear layer scores are represented in figure 2 and 3, respectively. Table 2 shows the intragroup comparison analysis. The Mann-Whitney U test revealed the following statistically significant differences between the two groups: scores for debris: group-C (30/0.04) differs significantly from group-B (25/0.04). Smear layer scores: group-C (30/0.04) differs significantly from group-B (25/0.04).

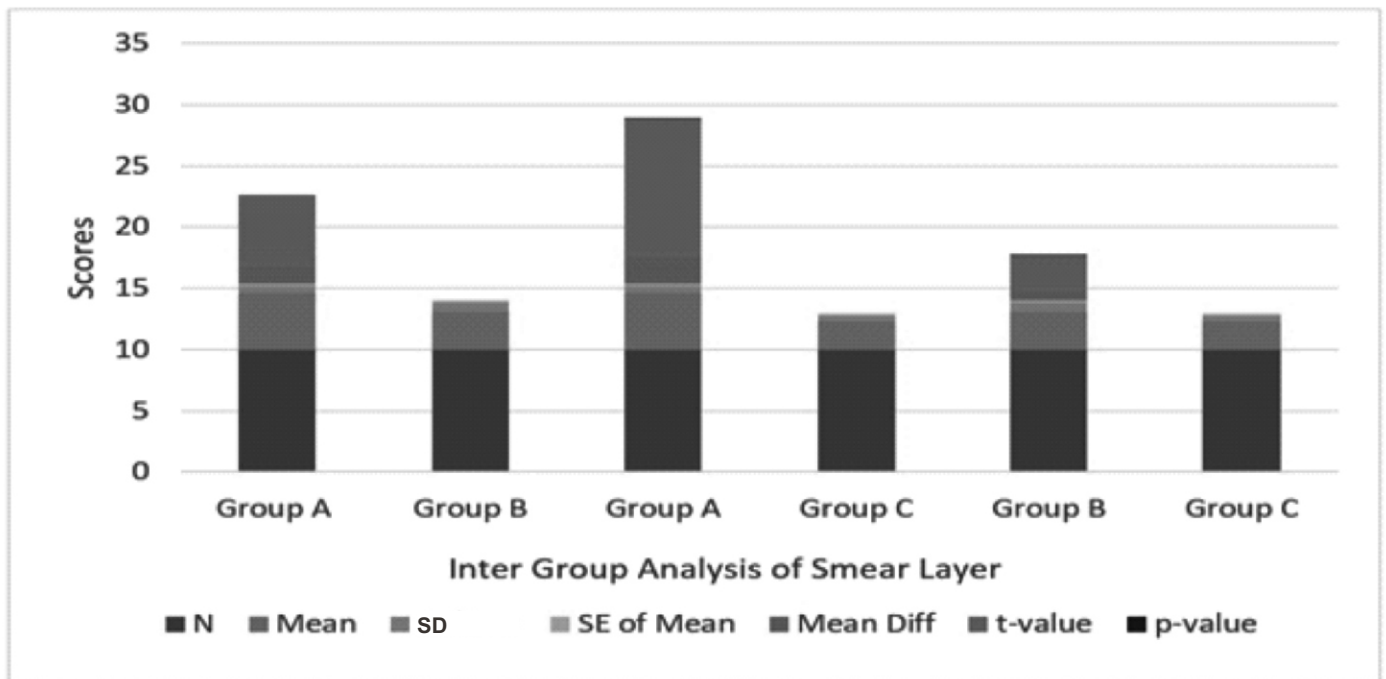
## DISCUSSION

A thorough shaping and cleaning of the root canal system is required for successful endodontic treatment. This is accomplished through mechanical shaping and chemical cleaning of the canal system. Mechanical preparation with manual or rotary tools produces a substantial amount of organic debris and smear layer.<sup>4</sup>

Debris is defined as dentine chips, tissue remnants, and particles that are loosely attached to the root canal wall, whereas the smear layer is a surface film of debris that is retained on dentine or other surfaces after instrumentation with rotary instruments or endodontic files.<sup>5</sup>



**Figure 3: Intergroup analysis of organic debris scores.**



**Figure 4: Intergroup analysis of smear layer scores.**

Pulp tissue residue, ground dentin, odontoblastic processes, and microbes comprise the smear layer. The majority of clinicians believe that the presence of the smear layer acts as a sealing barrier, causing leakage between the filling material and dentinal walls. It acts as a reservoir for microbial irritants and as a bacterial substrate. To remove the inorganic and organic components of the smear layer, the final irrigation sequence with ethylene diamine tetra acetic acid and sodium hypochlorite is currently

recommended.<sup>4</sup> The final size of the instrument used determines debris removal and irrigant penetration to the apical third of canals. Various results have been reported regarding the minimum master apical size of canals required for proper irrigant penetration.<sup>6</sup>

Pre-flaring of the coronal and middle thirds is recommended before determining the initial file that binds to determine the appropriate final diameter needed for complete apical enlargement. Apical gauging (AAE 2003)

is the measurement of the terminal diameter or shape of a canal after initial crown-down shaping to determine the size of the apical preparation. Although endodontists disagree on the ideal apical diameter of root canals, there is agreement that the ideal size varies from tooth to tooth and is determined by anatomical, microbiological, and mechanical factors.<sup>5</sup> In theory, preparing each canal to a specific apical diameter based on its initial apical size may assist clinicians in providing a more predictable root canal preparation.<sup>7</sup>

In this study cleaning efficacy at the apical third of root canals was evaluated using scanning electron microscopy after chemomechanical instrumentation with different sizes of the master apical file. A five-score evaluation index was used in previous studies to determine the organic debris and smear layer removal from the root canals, and magnifications of 27X, 1000X, and 2500X were used in the study because they provide a detailed image of the canal walls. The results revealed significant differences between group A (control group)- No further chemo-mechanical preparation, group B- Final chemo-mechanical preparation with rotary nickel-titanium files up to 25/0.04, and group C- Final chemo-mechanical preparation with rotary nickel-titanium files up to 30/0.04, which is most likely due to the improved irrigation in the apical third.

According to Salzgeber et al<sup>8</sup>, the removal of the smear layer and organic debris in the apical region was less predictable than in the coronal and middle thirds. This could be attributed to the apex's analogously smaller dimensions, which prevent irrigant penetration and cause limited contact between canal walls.<sup>8</sup> Bronnec et al<sup>9</sup> reported that improved root canal cleaning and shaping increased irrigant flow. Minimal apical size preparation, on the other hand, has been proposed to preserve tooth structure and limit obturating material extrusion.<sup>1</sup> Several researchers advocate for minimal apical size enlargement to preserve tooth structure and reduce endodontic complications.<sup>1</sup> Several studies have looked into the effect of apical preparation size on root canal cleanliness, microbial reduction, and healing outcomes following chemomechanical root canal wall preparation.<sup>2</sup>

There are various schools of thought regarding the ideal size and shape of root canal preparation. A common suggestion is to expand the root canal to at least three sizes beyond the initial file.<sup>10</sup> This recommendation is controversial because the initial file that binds does not always correspond to the true apical dimension, and it is

unclear whether enlarging by three sizes will adequately remove dentine circumferentially from the root canal walls.<sup>11</sup> In modern endodontic practice, there has been a shift toward the use of engine-driven rotary instrumentation with nickel-titanium instruments because they promote significantly less canal transportation than conventional files, provide more centered and tapered preparations, and emphasize the importance of additional irrigation as critical for adequate canal system disinfection.<sup>12</sup>

The apical size was critical during canal preparation in determining the successful debridement of the root canal system. According to Khademi et al<sup>6</sup> the smallest instrument size required for irrigant penetration to the apical third of the root canal is a #30/0.06 rotary file. This has been confirmed by research, which indicates that larger sizes are required for irrigating solutions to reach close to the apex and exhibit a greater microbial reduction.<sup>7</sup> Huang et al<sup>13</sup> proposed that increased apical size and taper allow for better irrigation throughout the root canal system. When the master apical size is less than 30, root canal taper can affect debridement, according to Arvaniti et al.<sup>14</sup> The results showed that increasing the taper size from 0.04 to 0.06 in file size #30 resulted in more smear layer removal but had no statistical significance in debris removal.<sup>14</sup>

Akhlaghi et al<sup>15</sup> reported that #30.04 maintains canal centering and minimal root thickness in the apical portion of curved canals. Sirekha et al<sup>16</sup> used two different needles to determine the effect of apical preparation size and taper on irrigant penetration into the apical third and discovered that an apical size of 30 allowed adequate irrigation with flat open-ended needles in the disto-buccal canal of mandibular molars. Boutsoukis et al<sup>17</sup> computational fluid dynamic model, an apical preparation size of 25 or greater improves the efficacy of conventional irrigation. This improves the antibacterial effects of intracanal medications and reduces late bacterial growth after shaping and cleaning the root canals.

Borges et al<sup>18</sup> investigated smear layer removal in curved canals using a preparation size of 30/0.02 versus 45/0.02. In group 45/0.02, they observed increased debris extrusion through the apical foramen, which resulted in postoperative periapical inflammation and delayed healing. As a result, this study compared apical preparation sizes of 25 and 30 with 4% taper that can be used in narrow and curved canals while avoiding iatrogenic complications like strip perforation, canal transportation, and ledging. As

a result, a master apical preparation size of 30 provided a good balance of tooth structure preservation and irrigation penetration depth, resulting in the efficient removal of organic debris and smear layer in the apical third of root canals.

**Limitations:** Cleaning efficacy was determined based on the number of opened dentinal tubules, but pathogens could exist in unobserved areas deeper within the dentinal matrix.

## CONCLUSION

The presence of non-occluded tubules aids but does not guarantee pathogen eradication within dentinal tubules via irrigant solutions. Further research using an activation protocol to incorporate the aforementioned elements is warranted and may be pursued in the future.

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