



## RESEARCH ARTICLE

### A COMPARATIVE EVALUATION OF SMEAR LAYER FORMATION FOLLOWING ROOT CANAL INSTRUMENTATION USING FIVE DIFFERENT INSTRUMENTATION TECHNIQUES: A SCANNING ELECTRON MICROSCOPE STUDY

\*Dr. Shivani Varma, Dr. Dildeep Bali, Dr. Vijaya Dhar Bhatt, Dr. Prashant Bhasin and Dr. Era Arora

Department of Conservative Dentistry and Endodontics, Santosh Dental College, PratapVihar, Ghaziabad, Uttar Pradesh, India

#### ARTICLE INFO

##### Article History:

Received 22<sup>nd</sup> May, 2016  
Received in revised form  
28<sup>th</sup> June, 2016  
Accepted 15<sup>th</sup> July, 2016  
Published online 31<sup>st</sup> August, 2016

##### Key words:

Hyflex CM files,  
ProTaper Next files,  
Smear layer,  
Twisted Files,  
WaveOne Files.

#### ABSTRACT

**Aim and objectives:** The aim of this study was to compare evaluate the quantitative formation of smear layer at the coronal, middle and apical thirds of root canals after using five different systems under the scanning electron microscope.

**Material and Methods:** The study was conducted on 75 extracted single-rooted human premolars. The teeth were divided into five experimental groups of 15 each. Group 1 was Control Group (Manual NiTi K-flexfile), Group 2 was Hyflex CM Files, Group 3 was ProTaper Next Files, Group 4 was Twisted Files and Group 5 was WaveOne Files. Evaluation of the photomicrographs was done to grade the smear layer with a 5 score index for each using reference photographs. Photomicrographs at X4000 (for the smear layer) were taken in the apical, middle and coronal thirds of the canal.

**Results:** Lower scores were found to be more among Group 1 and Group 3 whereas higher scores were found to be significantly more among Group 4 in Coronal, Middle and Apical third. The mean smear layer remaining value was significantly ( $p\text{-value}\leq 0.05$ ) lower among GROUP 1 in comparison to groups 2, 4 and 5 in coronal and middle third. The mean smear layer remaining value was significantly ( $p\text{-value}\leq 0.05$ ) lower among GROUP 1 in comparison to group 2 and 4 in apical third.

**Conclusion:** Manual NiTi K-flexfiles were found to have the maximum cleaning efficiency in the root canal at the coronal third as well as the middle third whereas at the apical third WaveOne files gave excellent results.

Copyright©2016, Dr. Shivani Varma et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Citation: Dr. Shivani Varma, Dr. Dildeep Bali, Dr. Vijaya Dhar Bhatt, Dr. Prashant Bhasin and Dr. Era Arora, 2016. "Assessing sterilization and infection control practices: the case of private dental practitioners in east Delhi region", *International Journal of Current Research*, 8, (08), 37104-37111.

## INTRODUCTION

The fundamental goals of endodontic treatment involve proper cleaning and shaping of the root canals, removal of all the pulp tissue, bacteria and their products, as well as provision of a suitable conformation for subsequent sealing.<sup>3</sup> Biomechanical preparation, disinfection and obturation together constitute as the important phases of the treatment. However, common to all types of cutting instruments used during cleaning and shaping a layer of material composed of organic and inorganic substances covers the instrumented walls which has a granular and an amorphous appearance under the scanning electron microscope. This is called as smear layer.<sup>9</sup> The smear layer was first described by McComb & Smith as a layer of

microcrystalline and organic particle debris that is found spread on root canal walls after root canal instrumentation.<sup>33</sup> Insufficient removal of smear layer increases the chance for bacterial contamination by acting as a source of nutrients for them and also induces stresses on the cutting segments of endodontic instruments.<sup>9</sup> Therefore, the importance of smear layer removal cannot be undermined. The different instruments used along with the irrigation regime that is followed during cleaning and shaping have a major role to play in the amount of smear layer formed. EDTA, sodium hypochlorite and saline amongst others are the different solutions used for irrigation during preparation. However, according to a study carried out by Ahmetoglu et al, sodium hypochlorite alone failed to remove smear layer effectively from the canals, but when it was used in combination with EDTA solution better results were obtained.<sup>2</sup> Newer methods have also been introduced to tackle smear layer formation. Moogi PP and Rao NR

\*Corresponding author: Dr. Shivani Varma,

Department of Conservative Dentistry and Endodontics, Santosh Dental College, PratapVihar, Ghaziabad, Uttar Pradesh, India.

advocated the use of laser beam (Nd: YAG) as a method to reduce formation of smear layer when compared with conventional filing technique.<sup>24</sup> Although when Takeda FH et al compared the effects on different lasers (argon, Nd:YAG, and Er:YAG) on the smear layer, it was found that Er:YAG laser was the most effective in removing smear layer on root canal walls.<sup>32</sup> Different instruments produce different amounts of debris and smear layer depending upon their design. For this, Poggio et al studied the effect of Mtwo, Revo-S and HyFlex CM on smear layer removal where HyFlex CM seem to be not very effective in promoting cleanliness of root canal walls when compared to Mtwo and Revo-S.<sup>26</sup> Similarly, according to a study by Sharma et al, when Twisted, Mtwo, and ProTaper rotary nickel titanium instruments were compared, Mtwo instruments were found to produce significantly cleaner dentin wall surfaces throughout the canal length in comparison to Twisted and ProTaper rotary files.<sup>31</sup>

Recently, HyFlex CM (where CM stands for Controlled Memory) (Coltene-Whaledent, Allstetten, Switzerland) NiTi files have been introduced. These instruments with shape memory are produced by an innovative methodology that uses a complex heating and cooling treatment that controls the material's memory.<sup>26</sup> It has the ability to respond to excessive resistance with straightening of their spirals, which avoids binding to the walls and thereby increases its fracture resistance. This straightening can be quickly reversed by heat treatment during autoclaving or with a glass bead sterilizer, making the instrument return to its original shape. This strengthens the files making it more resistant to cyclical fatigue. The new ProTaper Next (Dentsply Maillefer) is a recently introduced rotary file (Dentsply Maillefer) made of M-Wire NiTi material that improves its flexibility while still retaining the cutting efficiency. It has a rectangular cross-section that gives the file a snake-like "swaggering" movement as it moves through the canal. This results in optimization of the root canal as there are only two contact points at any one time between the file and the canal wall. Twisted files have been manufactured by SybronEndo (Orange, CA, USA). These files have a triangular cross section with constant tapers of 0.04, 0.06, 0.08, 0.10 and 0.12. These files are developed by twisting a triangular piece of nickel titanium (NiTi) wire. According to the manufacturer a thermal process allows twisting during phase transformation into the so called "R phase" of NiTi which increases the cyclic fatigue resistance and improves the flexibility.<sup>31</sup>

Wave one NiTi single file system is used in a reciprocal motion that needs special automated device. The system consists of 3 single use files made of alloy called M-Wire that is created by thermal-treatment process. The benefits of M-Wire NiTi are increased flexibility of the instruments and improved resistance to cyclic fatigue. The file is used in a reciprocal motion that needs special automated device.<sup>17</sup> Different methodologies can be undertaken to assess the presence of smear layer on the canal walls like stereoscopic microscope or scanning electron microscope. The advantage of electron microscopes over optical ones lies in the fact that they have a higher resolution and are therefore also able of a higher magnification that ranges from 20X to approximately 30,000X (SEM). They allow for the visualization of structures, which

would normally be not visible by optical microscopy. Therefore, scanning electron microscope was chosen for quantitative evaluation of smear layer formation at the coronal, middle and apical regions of the canals using Hand NiTi K-flex files, Hyflex CM, ProTaper Next, Twisted, and Wave One files, keeping the irrigation regime constant.<sup>17</sup> This study was undertaken to compare evaluate the quantitative formation of smear layer at the coronal, middle and apical thirds of root canals after using five different systems namely- Hand NiTi K-flex files, Hyflex CM, ProTaper Next, Twisted, and Wave One files under the scanning electron microscope.

## MATERIALS AND METHODS

The study was conducted on 75 extracted single-rooted human premolars, which were collected from Department of Oral and Maxillofacial Surgery, Santosh Dental College and Hospital and fixed in 10% formalin following extractions. The root surfaces were debrided with hand scalers, washed and stored in distilled water till the study was carried out.

### Preparation of specimen

Crowns of each of the specimens were removed with diamond discs placed at the cemento-enamel junction. A conventional access preparation was only done in teeth in which the canal opening was compromised. A small barbed broach was used to extirpate the pulp before using orifice shapers or the subsequent instruments. A 10 K- flexofile (Dentsply, Maillefer) was placed into the canal until it was visible at the apical foramen and the working length was established. The procedure was done under magnification using operating microscope at 15X.

### Materials used

The teeth were divided into five experimental groups of 15 each. Group 1 was Control Group (Manual NiTi K-flexfile), Group 2 was Hyflex CM Files, Group 3 was ProTaper Next Files, Group 4 was Twisted Files and Group 5 was WaveOne Files.

### Preparation of Canal

Group 1 was shaped using "watch-winding" manipulation of Hand NiTi K-flex files (20-40) with a step-back technique. Final apical preparation size was done till file of size 25. Group 2 were prepared using Hyflex CM according to manufacturers' instructions. The shaping procedure commenced with Hyflex CM file size 25 taper 0.08. File size 20 taper 0.04 was then inserted and shaping continued with file size 25 taper 0.04 and completed with file size 20 taper 0.06. Group 3 were prepared using Protaper Next according to manufacturers' instructions. The shaping commenced with Protaper Next X1 file size 17 taper 0.04 followed by X2 (size 25 taper 0.06) to the working length. Group 4 were prepared using Twisted Files according to the manufacturers' instructions. The shaping procedure commenced with Twisted Files size 25 taper 0.08. The coronal one third or two thirds of the root canal was shaped if passive penetration was possible. TF size 25 taper 0.06 was inserted and used until 2mm short of

WL. Shaping continued with TF size 25 and taper 0.04 and completed with size 25 taper 0.06 file to the working length. Group 5 were prepared using WaveOne Primary File, having a size 25 and a taper of 0.08, according to manufacturers' recommendation. The shaping procedure commenced with PathFile #1 followed by #2 to WL. WaveOne Primary reciprocating files used in motion till the WL followed this. Canals were irrigated with 1ml of 5.25% NaOCl followed by 0.5ml of 10% EDTA after each change of the instrument. Canals were flooded with irrigating agent throughout the procedure using passive irrigation. After canal preparation a final 1ml of 17% EDTA solution was left in situ for 2 minutes, then replaced by 1ml of 5.25% NaOCl for 3 minutes. Irrigation was performed using 5ml syringe with 27-gauge needle. Immediately after canal preparation, two longitudinal grooves were made on each sample using diamond discs (buccal and lingual surface). The grooves made were not deep enough to enter the canals. This was followed by final splitting which was done by mallet and a stainless steel chisel, to avoid contamination of the canals during the separation process. The root halves were cleaned from the grinding material and dried using water and air blasting for three seconds. The specimens were left to dry overnight. One half of each tooth was selected for further SEM analysis.<sup>20</sup>

### Scanning & Imaging

The specimens were then dehydrated, mounted on brass stubs. The stubs were marked with marking pen and gold sputtered in an ion sputtering machine. The specimens were examined under SEM (Zeiss EVO 40) for assessment of microscopic pattern of magnification X4000 for smear layer. A standardized series of 3 photomicrographs were taken for each pulp space (one in the apical third, one in the middle third and one in the coronal third) for comparative purposes. Evaluation of the photomicrographs was done by 2 evaluators to grade the smear layer with a 5 score index for each using reference photographs. Photomicrographs at X4000 (for the smear layer) were taken in the apical, middle and coronal thirds of the canal. Consensus reached on the scoring criteria was taken as the final score.

### Criteria for evaluation of Smear layer

The smear layer was evaluated according to the criteria by Manjunatha et al.<sup>20</sup> According to Manjunatha et al.<sup>20</sup> two different terminologies were used- Smear layer and debris to evaluate the cleaning efficacy of different file systems. Smear layer scoring was given according to the number of dentinal tubules that were open.

### Statistical analysis

The data was recorded and transferred from assessment form to the computer and master chart was created in the excel sheet using Microsoft Excel Software. The statistical analysis was performed using SPSS version 16 software. The data was summarized in the form of mean and standard deviation for numerical data, and frequencies and percentages for categorical data. Presentation of summarized data was done using Tables and Graphs.

The normality of the data was checked using Shapiro Wilk test. Non-Parametric tests were used for the inferential statistics of those variables which failed to achieve normal distribution and/or which were ordinal in nature. Mann Whitney U test was used for comparison of the mean values between two groups. Kruskal Walli's test was used for comparison of the mean values between three or more groups. The distribution of the categorical variables among the different age groups and gender was compared using Chi-square test. All the statistical tests were set at a 95% confidence level and p-value of less than 0.05 was considered to be statistically significant.

## RESULTS

The distribution of scores for smear layer formed at the Coronal Third of the root showed that score 1 was significantly ( $p\text{-value}\leq 0.05$ ) more among Manual NiTi K-flexofile (73.3%) and ProTaper Next (46.7%) groups. The score 2 was significantly more among WaveOne File (66.7%) and HyFlex CM File (40.0%) groups. The score 3 was significantly more among HyFlex CM File (40.0%) and ProTaper Next (20.0%) groups. The score 4 and 5 was significantly more in Twisted File group (80.0% and 20.0% respectively). (Table 1) The distribution of scores for smear layer formed at the Middle Third of the root showed that the score 1 was significantly ( $p\text{-value}\leq 0.05$ ) more among Manual NiTi K-flexofile group (66.7%) followed by ProTaper Next group (40.0%) and score 2 was significantly ( $p\text{-value}\leq 0.05$ ) more among WaveOne File (66.7%) and ProTaper Next (46.7%) groups.

The score 3 was significantly more among HyFlex CM File group (53.3%) followed by ProTaper Next group (13.3%). Score 4 and 5 were significantly more among Twisted File group (93.3% and 6.7% respectively). (Table 2) The distribution of scores for the smear layer formed at the Apical Third of the root showed that Score 1 was significantly ( $p\text{-value}\leq 0.05$ ) more among WaveOne File (33.3%) and ProTaper Next (20.0%) groups whereas score 2 was significantly ( $p\text{-value}\leq 0.05$ ) more among ProTaper Next (60.0%) followed by Manual NiTi K-flexofile group (53.3%). The score 3 was significantly more among HyFlex CM file (66.7%) and Manual NiTi K-flexofile (33.3%) groups.

The score 4 and 5 was significantly more among Twisted file group (80.0% and 20.0% respectively). Lower scores were found to be significantly ( $p\text{-value}\leq 0.05$ ) more among WaveOne File and ProTaper Next file groups followed by Manual NiTi K-flexofile and HyFlex CM file groups whereas the higher scores were found to be significantly ( $p\text{-value}\leq 0.05$ ) more among Twisted File group. (Table 3) The mean smear layer remaining values at Coronal third among Manual NiTi K-flexofile group was  $1.27\pm 0.46$ , Hyflex File group was  $2.20\pm 0.77$ , ProTaper next group was  $1.73\pm 0.80$ , Twisted file group was  $3.93\pm 0.46$  and Waveone File group was  $1.80\pm 0.56$ . The mean smear layer remaining values were compared using the kruskal-wallis test and were found to have a statistically significant ( $p\text{-value}\leq 0.05$ ) difference between these groups. (Table 4) The inter-group comparison of the mean smear layer remaining values at Coronal third among the various groups was done using the Mann-whitney U test.

**Table 1. The distribution of smear layer scores among the different groups in coronal third of the teeth**

| Coronal | GROUP 1 - Manual NiTi K-flexofile | GROUP 2- HYFLEX FILE | GROUP 3- ProTaper next | GROUP 4 - Twisted | GROUP 5- WAVEONE FILE | p-value  |
|---------|-----------------------------------|----------------------|------------------------|-------------------|-----------------------|----------|
| Score 1 | 11<br>73.3%                       | 3<br>20.0%           | 7<br>46.7%             | 0<br>.0%          | 4<br>26.7%            | 0.000*** |
| Score 2 | 4<br>26.7%                        | 6<br>40.0%           | 5<br>33.3%             | 0<br>.0%          | 10<br>66.7%           |          |
| Score 3 | 0<br>.0%                          | 6<br>40.0%           | 3<br>20.0%             | 2<br>13.3%        | 1<br>6.7%             |          |
| Score 4 | 0<br>.0%                          | 0<br>.0%             | 0<br>.0%               | 12<br>80.0%       | 0<br>.0%              |          |
| Score 5 | 0<br>.0%                          | 0<br>.0%             | 0<br>.0%               | 1<br>6.7%         | 0<br>.0%              |          |
| Total   | 15<br>100.0%                      | 15<br>100.0%         | 15<br>100.0%           | 15<br>100.0%      | 15<br>100.0%          |          |

Chi-square test  
\*\*\* Very Highly significant difference (p-value≤0.001)

**Table 2. The distribution of smear layer scores among the different groups in middle third of the teeth**

| Middle  | GROUP 1 - Manual NiTi K-flexofile | GROUP 2- HYFLEX FILE | GROUP 3- ProTaper next | GROUP 4 - Twisted | GROUP 5- WAVEONE FILE | p-value      |
|---------|-----------------------------------|----------------------|------------------------|-------------------|-----------------------|--------------|
| Score 1 | 10<br>66.7%                       | 3<br>20.0%           | 6<br>40.0%             | 0<br>.0%          | 4<br>26.7%            | 0.000**<br>* |
| Score 2 | 5<br>33.3%                        | 4<br>26.7%           | 7<br>46.7%             | 0<br>.0%          | 10<br>66.7%           |              |
| Score 3 | 0<br>.0%                          | 8<br>53.3%           | 2<br>13.3%             | 0<br>.0%          | 1<br>6.7%             |              |
| Score 4 | 0<br>.0%                          | 0<br>.0%             | 0<br>.0%               | 14<br>93.3%       | 0<br>.0%              |              |
| Score 5 | 0<br>.0%                          | 0<br>.0%             | 0<br>.0%               | 1<br>6.7%         | 0<br>.0%              |              |
| Total   | 15<br>100.0%                      | 15<br>100.0%         | 15<br>100.0%           | 15<br>100.0%      | 15<br>100.0%          |              |

Chi-square test  
\*\*\* Very Highly significant difference (p-value≤0.001)

**Table 3. The distribution of smear layer scores among the different groups in apical third of the teeth**

| Apical  | GROUP 1 - Manual NiTi K-flexofile | GROUP 2- HYFLEX FILE | GROUP 3- ProTaper next | GROUP 4 - Twisted | GROUP 5- WAVEONE FILE | p-value  |
|---------|-----------------------------------|----------------------|------------------------|-------------------|-----------------------|----------|
| Score 1 | 2<br>13.3%                        | 0<br>.0%             | 3<br>20.0%             | 0<br>.0%          | 5<br>33.3%            | 0.000*** |
| Score 2 | 8<br>53.3%                        | 5<br>33.3%           | 9<br>60.0%             | 0<br>.0%          | 7<br>46.7%            |          |
| Score 3 | 5<br>33.3%                        | 10<br>66.7%          | 3<br>20.0%             | 0<br>.0%          | 3<br>20.0%            |          |
| Score 4 | 0<br>.0%                          | 0<br>.0%             | 0<br>.0%               | 12<br>80.0%       | 0<br>.0%              |          |
| Score 5 | 0<br>.0%                          | 0<br>.0%             | 0<br>.0%               | 3<br>20.0%        | 0<br>.0%              |          |
| Total   | 15<br>100.0%                      | 15<br>100.0%         | 15<br>100.0%           | 15<br>100.0%      | 15<br>100.0%          |          |

Chi-square test  
\*\*\* Very Highly significant difference (p-value≤0.001)

**Table 4. The comparison of mean smear layer scores among the different groups in coronal third of the teeth**

| Coronal third                     | Mean | S.D.                         | Mean Rank |
|-----------------------------------|------|------------------------------|-----------|
| GROUP 1 - Manual NiTi K-flexofile | 1.27 | 0.46                         | 19.67     |
| GROUP 2- HYFLEX FILE              | 2.20 | 0.77                         | 40.40     |
| GROUP 3- ProTaper next            | 1.73 | 0.80                         | 30.03     |
| GROUP 4 -Twisted                  | 3.93 | 0.46                         | 67.33     |
| GROUP 5- WAVEONE FILE             | 1.80 | 0.56                         | 32.57     |
| Critical value                    |      | 44.563                       |           |
| p-value <sup>a</sup>              |      | < 0.001*                     |           |
| Post-hoc comparisons <sup>b</sup> |      | Gp 1 <Gp 2,4<br>Gp 3,5 <Gp 4 |           |

<sup>a</sup>Kruskal-wallis test  
<sup>b</sup>Mann-whitney U test  
\* Significant difference

**Table 5. The comparison of mean smear layer scores among the different groups in middle third of the teeth**

| Middle third                      | Mean | S.D.            | Mean Rank |
|-----------------------------------|------|-----------------|-----------|
| GROUP 1 - Manual NiTi K-flexofile | 1.33 | 0.49            | 20.17     |
| GROUP 2- HYFLEX FILE              | 2.33 | 0.82            | 41.47     |
| GROUP 3- ProTaper next            | 1.73 | 0.70            | 29.17     |
| GROUP 4 –Twisted                  | 4.07 | 0.26            | 68.00     |
| GROUP 5- WAVEONE FILE             | 1.80 | 0.56            | 31.20     |
| Critical value                    |      | 46.49           |           |
| p-value <sup>a</sup>              |      | < 0.001*        |           |
| Post-hoc comparisons <sup>b</sup> |      | Gp 1 < Gp 2,4,5 |           |
|                                   |      | Gp 2,5 < Gp 4   |           |
|                                   |      | Gp 3 < Gp 2, 4  |           |
| <sup>a</sup> Kruskal-wallis test  |      |                 |           |
| <sup>b</sup> Mann-whitney U test  |      |                 |           |
| * Significant difference          |      |                 |           |

**Table 6. The comparison of mean smear layer scores among the different groups in apical third of the teeth**

| Apical third                      | Mean | S.D.              | Mean Rank |
|-----------------------------------|------|-------------------|-----------|
| GROUP 1 - Manual NiTi K-flexofile | 2.20 | 0.68              | 30.73     |
| GROUP 2- HYFLEX FILE              | 2.67 | 0.49              | 41.67     |
| GROUP 3- ProTaper next            | 2.00 | 0.65              | 26.10     |
| GROUP 4 –Twisted                  | 4.20 | 0.41              | 68.00     |
| GROUP 5- WAVEONE FILE             | 1.87 | 0.74              | 23.50     |
| Critical value                    |      | 45.55             |           |
| p-value <sup>a</sup>              |      | < 0.001*          |           |
| Post-hoc comparisons <sup>b</sup> |      | Gp 1 < Gp 2,4     |           |
|                                   |      | Gp 3,5 < Gp 2     |           |
|                                   |      | Gp 2, 3, 5 < Gp 4 |           |
| <sup>a</sup> Kruskal-wallis test  |      |                   |           |
| <sup>b</sup> Mann-whitney U test  |      |                   |           |
| * Significant difference          |      |                   |           |

The mean smear layer remaining value was significantly ( $p\text{-value}\leq 0.05$ ) lower among Group 1 in comparison to groups 2, 4 and 5, group 2 and 3 had significantly ( $p\text{-value}\leq 0.05$ ) lower mean smear layer remaining value in comparison to group 4 and group 4 had significantly ( $p\text{-value}\leq 0.05$ ) higher mean smear layer remaining value in comparison to group 5. (Table 4) The mean smear layer remaining values among Manual NiTi K-flexofile group was  $1.33\pm 0.49$ , Hyflex file group was  $2.33\pm 0.82$ , ProTaper next group was  $1.73\pm 0.70$ , Twisted file group was  $4.07\pm 0.26$  and Waveone file group was  $1.80\pm 0.56$ . The mean smear layer remaining values were compared among these groups using the Kruskal-wallis test and were found to have a statistically significant ( $p\text{-value}\leq 0.05$ ) difference. (Table 5) The inter-group comparison of the mean smear layer remaining values at CORONAL third among the various groups was done using the Mann-whitney U test. The mean smear layer remaining value was significantly ( $p\text{-value}\leq 0.05$ ) lower among Group 1 in comparison to Group 2, 3 and 5, group 2 and 3 had significantly ( $p\text{-value}\leq 0.05$ ) lower mean smear layer remaining value in comparison to group 4, group 2 had significantly ( $p\text{-value}\leq 0.05$ ) higher mean smear layer remaining value in comparison to group 3 and 5 whereas group 4 had significantly ( $p\text{-value}\leq 0.05$ ) higher mean smear layer remaining value in comparison to group 5. (Table 5) The mean smear layer remaining values in the Apical third among Manual NiTi K-flexofile group was  $2.20\pm 0.68$ , Hyflex file group was  $2.67\pm 0.49$ , ProTaper next file group was  $2.00\pm 0.65$ , Twisted file group was  $4.20\pm 0.41$  and Waveone file

group was  $1.87\pm 0.74$ . The mean smear layer remaining values were compared between these groups using the Kruskal-wallis test and were found to have a statistically significant ( $p\text{-value}\leq 0.05$ ) difference. (Table 6) The inter-group comparison of the mean smear layer remaining values at CORONAL third among the various groups was done using the Mann-whitney U test. The mean smear layer remaining value was significantly ( $p\text{-value}\leq 0.05$ ) lower among Group 1 in comparison to group 2 and 4, group 2 and 3 had significantly ( $p\text{-value}\leq 0.05$ ) lower mean smear layer remaining value in comparison to group 4, group 2 had significantly ( $p\text{-value}\leq 0.05$ ) higher mean smear layer remaining value in comparison to group 3 and 5 whereas group 4 had significantly ( $p\text{-value}\leq 0.05$ ) higher mean smear layer remaining value in comparison to group 5. (Table 6)

## DISCUSSION

McComb & Smith<sup>21</sup> were the first researchers to describe the smear layer formed on the surface of instrumented root canals. They suggested that it consisted of dentine along with the remnants of odontoblastic processes, pulp tissue and bacteria. This study was closely followed by Lester & Boyde<sup>18</sup> who described smear layer as the 'organic matter trapped within translocated inorganic dentine.' They were able to conclude that smear layer was primarily composed of inorganic dentine as irrigation done with sodium hypochlorite proved ineffective.

During early stages of instrumentation, it has been generally observed that the smear layer present on the walls of canals is higher in organic content than inorganic due to presence of necrotic and/or viable pulp tissue.<sup>5</sup> However, increased centrifugal forces resulting from the movement and the proximity of the instruments to the dentinal wall can cause creation of a thicker layer which is more resistant to removal with chelating agents.<sup>13</sup> One of the most important factors affecting the outcome of root canal treatment is the seal created between the restoration and the walls of the canal. Therefore, it was important to understand the effect of the smear layer on the apical and coronal seal.<sup>33</sup> There were two schools of thought relating to the creation of smear layer. While on one hand, it was believed to limit the effective disinfection of dentinal tubules by preventing sodium hypochlorite, calcium hydroxide and other intracanal medicaments from penetrating into the dentinal tubules thus providing an avenue for leakage.<sup>19,22</sup> On the other hand, it was also believed that it blocked the dentinal tubules, limiting bacteria and their toxins from penetrating into the tubules by altering dentinal permeability altogether.<sup>23,28</sup> However, the removal of smear layer is advocated by factors such as its unpredictable thickness and volume as water is considered as its main component.<sup>7</sup> In addition, it contains bacteria, their by-products and necrotic tissue, which could serve as a reservoir of microbial irritants, allowing their deeper penetration into the dentinal tubules.<sup>8,11,12</sup> It limits the optimum penetration of disinfecting agents by acting as a barrier between filling materials and the canal wall thereby compromising the formation of a satisfactory seal.<sup>25,32,34,35</sup> There have been various methods that have been employed in order to remove smear layer over the years. A number of chemicals have been investigated in the process. Kaufman & Greenberg<sup>15</sup> stated that 'a working solution is the one which is used to clean the canal, and an irrigation solution the one which is essential to remove the debris and smear layer created by the instrumentation process.' One of the most popular irrigants, which had a long lasting antibacterial effect, was Chlorhexidine, but it was found incapable of removing smear layer, as it could not dissolve its organic contents. Studies done also concluded that even sodium hypochlorite, alone, was unable to remove smear layer from the instrumented walls. It was able to disinfect them only superficially.<sup>4</sup> It has generally been observed that the removal of smear layer as well as soft tissue and debris can be achieved effectively by the alternate use of EDTA and NaOCl.<sup>4,6,35</sup> Various combinations of EDTA and NaOCl have been examined in studies but the most effective one is found to be a final rinse with 10 mL of 17% EDTA followed by 10 mL of 5.25% NaOCl.<sup>35</sup> In the present study, scanning electron microscope was used to compare the quantitative formation of smear layer using Hand NiTi K-files, Hyflex CM files, ProTaper Next files, Twisted files and WaveOne files.

The goals of instrumentation include a continuously tapered preparation that maintains the canal anatomy, facilitates optimal irrigation, allows complete debridement and placement of local medicaments and permanent root filling. Thus, mechanical instrumentation plays a major role in the cascade of treatment procedures that aim at the eradication of microbes from the root canal. The objective of canal shaping as stated by Schilder,<sup>30</sup> included cleaning and shaping of the root canal i.e.

the canals should be cleaned of their organic remnants and shaped to receive a three dimensional hermetic filling throughout the canal space. In order to achieve complete removal of debris and microbes from the canal, instrumentation must be supplemented with appropriate irrigation. Residual bacteria and other microorganisms usually exist in areas that are inaccessible like the crevices, fins and ramifications of a root canal as well as the dentinal tubules. These areas cannot be cleaned mechanically; therefore, this makes chemical disinfection or canal cleaning an important adjunct to canal shaping.<sup>30</sup> The result of root canal preparation is dependent upon various factors, out of which, canal anatomy, is of prime importance. Keeping this in mind, over the years a variety of instruments and techniques have also been introduced in order to obtain a clean and debris free canal fit for obturation.<sup>30</sup> In the present study, Manual NiTi K-flexofile was found to be best in debris removal in the coronal and middle third as compared to the other file systems under study. It was also found to be effective in the apical third as well third best after Waveone and Protaper Next. Similarly, Reddy<sup>27</sup> compared the amount of smear layer and debris removal from canal walls following the use of manual nickel-titanium files and rotary ProTaper NiTi files and observed that manual instrumentation produced significantly less smear layer and debris when compared with the rotary instruments. So, the findings similar to the present study were found. In a study by Dhanyakumar et al,<sup>9</sup> evaluation of smear layer formation was done after using three different nickel-titanium rotary instruments-Endowave, K3 and ProTaper files.

This study demonstrated that Endowave system produced cleaner walls but the results were not statistically significant when compared with K3 and ProTaper. Almost, similar results were reported in the present study with no significant difference between protaper and waveone system. In a study carried out by Kocak et al,<sup>16</sup> comparing the amount of extruded debris after canal preparation using ProTaper Universal and ProTaper Next files concluded that the ProTaper Universal group produced a significantly greater amount of debris extrusion ( $P < 0.001$ ) when compared to ProTaper Next files. So, protaper next is considered to be better than its predecessor. Soumya et al<sup>29</sup> compared the cleaning efficacy of the following NiTi instruments- Hand ProTaper, HERO shaper and Twisted files and found that HERO shaper files caused maximum amount of smear layer which was followed by the Twisted rotary instruments and finally by hand instruments that produced least amount of smear layer. However, in the present study, twisted file groups was found to produce the maximum smear layer when compared to the protaper next. Sharma et al<sup>31</sup> evaluated the smear layer formed on root canal walls of extracted human teeth using three rotary nickel-titanium instruments: Twisted, Mtwo, and ProTaper concluded that Twisted Files resulted in less smear layer formation in the apical thirds of the canal when compared to ProTaper rotary instruments but the results were statistically insignificant. It was however, the Mtwo rotary file system that produced significantly less smear layer ( $p < 0.001$ ) in the apical portion. Contrasting to this study, our study showed that the Twisted files produced maximum amount of smear layer as compared to the protaper next and other file systems. The studies carried out by Foschi et al,<sup>10</sup> Soumya et al<sup>29</sup> and Reddy<sup>27</sup> have

concluded that all type of instrumentation showed some amount of smear layer with inefficiency of cleaning at the apical third. In our study, Twisted Files were most inefficient in removing the smear layer from the instrumented canals. It could be due to its constant taper of a maximum 8% that is able to remove smaller amounts of dentin from the canals thereby compromising irrigation control and upward debris removal.<sup>14</sup> In our study, the three regions namely- coronal, middle and apical third were also compared in respect of their cleanliness and disinfection post instrumentation and a strict irrigation regime. The results obtained demonstrated that the coronal third of the root canal was much cleaner than the middle region. However, this protocol was unable to create a clean and a smear layer free apical region. This could be attributed to the reduced dimension of the root canal at the apical third causing entrapment of air bubbles and prevention of total wetting with the irrigant.<sup>32</sup>

### Conclusion

In the present study Manual NiTi K-flexofiles were found to have the maximum cleaning efficiency in the root canal at the coronal third as well as the middle third. This was closely followed by ProTaper Next files and WaveOne files. However, at the apical third WaveOne files gave excellent results which were followed by results of ProTaper Next and Manual NiTi K-flexofiles. Twisted files were found to be least effective in removing smear layer from the root canals at all the levels. Within the limitations of this study it may be concluded that none of the canal preparation instruments used left completely clear root canal walls. However, further in vitro studies and clinical trials using different methodologies are warranted to establish the cleaning efficacy of the files used in this study design.

### REFERENCES

1. A comparative evaluation of cleaning efficacy (debris and smear layer removal) of hand and two rotary instrumentation systems (K3 and Protaper): a SEM study. *J Contemp Dent Pract* 2013;14(6):1028-35.
2. Ahmetoglu F, Keles A, Yalcin M, Simsek N. Effectiveness of different irrigation systems on smear layer removal: A scanning electron microscopic study. *Eur J Dent* 2014;8(1): 53-7.
3. Amaral P, Forner L, Llana C. Smear Layer removal in canals shaped with reciprocating rotary systems. *J Clin Exp Dent* 2013; 5(5): e227-30.
4. Baumgartner JC, Mader CL. A scanning electron microscopic evaluation of four root canal irrigation regimens. *J Endod* 1987;13(4):147-57.
5. Cameron JA. The use of ultrasound for the removal of the smear layer. The effect of sodium hypochlorite concentration; SEM study. *Aust Dent J* 1988;33(3):193-200.
6. Cengiz T, Aktener BO, Piskin B. Effect of dentinal tubule orientation on the removal of smear layer by root canal irrigants. A scanning electron microscopic study. *Int Endod J* 1990;23(3):163-71.
7. Cergneux M, Ciucchi B, Dietschi JM, Holz J. The influence of the smear layer on the sealing ability of canal obturation. *Int Endod J* 1987;20(5):228-32.
8. Cunningham WT, Martin H, Forrest WR. Evaluation of root canal débridement by the endosonic ultrasonic synergistic system. *Oral Surg Oral Med Oral Pathol* 1982;53(4):401-4.
9. Dhanyakumar N M, Vasundhara Shivanna, Saurabh Garg. SEM evaluation of smear layer formation after using three different nickel titanium rotary instruments- Endowave, K3 and Protaper- an invitro study. *Endodontology* 2010;22(1):26-36.
10. Foschi F, Nucci C, Montebugnoli L, Marchionni S, Breschi L, Malagnino VA, Prati C. SEM evaluation of canal wall dentine following use of Mtwo and ProTaper NiTi rotary instruments. *Int Endod J* 2004;37(12):832-9.
11. George S, Kishen A, Song KP. The role of environmental changes on monospecies biofilm formation on root canal wall by *Enterococcus faecalis*. *J Endod* 2005;31(12):867-72.
12. Goldberg F, Abramovich A. Analysis of the effect of EDTAC on the dentinal walls of the root canal. *J Endod.* 1977 Mar;3(3):101-5.
13. Jodaikin A, Austin JC. Smear layer removal with chelating agents after cavity preparation. *J Prosthet Dent* 1981;46(2):171-4.
14. Kadhom TH, Al-Hashimi WN. A study to compare the efficiency of different instrumentation systems for cleaning oval-shaped root canals (An in vitro study). *J Bagh Coll Dentistry* 2013;25(1):49-55.
15. Kaufman AY, Greenberg I. Comparative study of the configuration and the cleanliness level of root canals prepared with the aid of sodium hypochlorite and bis-dequalinium-acetate solutions. *Oral Surg Oral Med Oral Pathol* 1986;62(2):191-7.
16. Koçak MM, Çiçek E, Koçak S, Sağlam BC, Yılmaz N. Apical extrusion of debris using ProTaper Universal and ProTaper Next rotary systems. *Int Endod J* 2015;48(3):283-6.
17. Kocak MM, Kocak S, Sağlam BC, Turker SA. Smear layer and debris removal following use of WaveOne and OneShape single-file systems- SEM study. *International Journal of Experimental Dental Science* 2014; 3(2):77-83.
18. Lester KS, Boyde A. Scanning electron microscopy of instrumented, irrigated and filled root canals. *Br Dent J* 1977;143(11):359-67.
19. Mader CL, Baumgartner JC, Peters DD. Scanning electron microscopic investigation of the smeared layer on root canal walls. *J Endod* 1984;10(10):477-83.
20. Manjunatha M, Annapurna K, Sudhakar V, Sunil Kumar V, Hiremath VK, Shah A. Smear layer evaluation on root canal preparation with manual and rotary techniques using EDTA as irrigant: a scanning electron microscopy study. *J Int Oral Health* 2013;5(1):66-78.
21. McComb D, Smith DC. A preliminary scanning electron microscopic study of root canals after endodontic procedures. *J Endod* 1975;1(7):238-42.
22. Meryon SD, Brook AM. Penetration of dentine by three oral bacteria in vitro and their associated cytotoxicity. *Int Endod J* 1990;23(4):196-202.
23. Michelich VJ, Schuster GS, Pashley DH. Bacterial penetration of human dentin in vitro. *J Dent Res* 1980;59(8):1398-403.

24. Moogi PP, Rao RN. Cleaning and shaping the root canal with an Nd: YAG laser beam: A comparative study. *J Conserv Dent* 2010;13(2):84-8.
25. Outhwaite WC, Livingston MJ, Pashley DH. Effects of changes in surface area, thickness, temperature and post-extraction time on human dentine permeability. *Arch Oral Biol* 1976;21(10):599-603.
26. Poggio C, Dagna A, Chiesa M, Beltrami R, Bianchi S. Cleaning Effectiveness of Three NiTi Rotary Instruments: A Focus on Biomaterial Properties. *J. Funct. Biomater* 2015;6:66-76.
27. Reddy JM, Latha P, Gowda B, Manvikar V, Vijayalaxmi DB, Ponangi KC. Smear layer and debris removal using manual Ni-Ti files compared with rotary Protaper Ni- Ti files - An In-Vitro SEM study. *J Int Oral Health* 2014;6(1):89-94.
28. Safavi KE, Spangberg LS, Langeland K. Root canal dentinal tubule disinfection. *J Endod* 1990;16(5):207-10.
29. Soumya S, Aggarwal S, Borkar A, Tandale A, Shetty P, Bhosale S. A scanning electron microscopic study of smear layer remaining following use of Greater Taper rotary instruments. *J Int Clin Dent Res Organ* 2011;3:37-42.
30. Schilder H. Cleaning and shaping of the root canal. *Dent Clin North Am* 1974;18:269-96.
31. Sharma G, Kakkar P, Vats A. A comparative SEM investigation of smear layer remaining on dentinal walls by three rotary NiTi with different cross-sectional designs in moderately curved canals. *J Clin Diagn Res* 2015;9(3):ZC43-7.
32. Takeda FH, Harashima T, Kimura Y, Matsumoto K. Comparative study about the removal of smear layer by three types of laser devices. *Clin Laser Med Surg* 1998;16(2):117-22.
33. Violich DR, Chandler NP. The smear layer in endodontics- a review. *Int Endod J* 2010;43(1):2-15.
34. Wayman BE, Kopp WM, Pinero GJ, Lazzari EP. Citric and lactic acids as root canal irrigants in vitro. *J Endod* 1979;5(9):258-65.
35. Yamada RS, Armas A, Goldman M, Lin PS. A scanning electron microscopic comparison of a high volume final flush with several irrigating solutions: Part 3. *J Endod* 1983;9(4):137-42.

\*\*\*\*\*