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# Comparison of smear layer removing efficacy of Cold Atmospheric Pressure (CAP) Plasma Jet with different chelating agents. An ex-vivo study

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**ABSTRACT**
The present study aimed to assess the effectiveness of different final irrigation regimens (Cold Atmospheric Pressure Plasma Jet, MTAD, and EDTA) in removing the smear layer from intra-radicular dentin using a Scanning Electron Microscope (SEM). Eighty-four mandibular premolars were prepared with ProTaper Universal hand files and were equally divided into four groups i.e. Normal saline (control), EDTA, MTAD and CAP Plasma Jet. Prepared samples in the control, EDTA and MTAD groups were irrigated with 5 milliliters of the irrigant, and it was retained for 2 min. In the CAP Plasma Jet group, the plasma plume was directed towards the canal lumen for 2 min. The smear layer removal of all the groups was evaluated at the coronal, middle and apical thirds. Statistical analysis was performed using Kruskal–Wallis test followed by Dunn's test. Evaluation by SEM showed that the smear layer removal ability of MTAD and EDTA were significantly better than CAP Plasma Jet ($p < 0.05$). While CAP Plasma Jet showed results comparable to EDTA in the coronal third. In the middle and apical third of the canal, its effect was comparable to the control group ($p > 0.05$). MTAD and EDTA aided in better smear layer removal than the CAP Plasma Jet in the coronal, middle, and apical third of the test samples. CAP Plasma jet performed better in the coronal third.

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Smear layer removal; endodontic irrigation; cold atmospheric pressure plasma jet; BioPure MTAD; eDTA

## Introduction

Root canal instrumentation causes an amorphous 'smear layer' to form on the canal walls, comprised of dentin particles, pieces of the viable or necrotic pulp tissue, bacterial components, and leftover irrigant [1,2]. This layer inhibits root canal irrigant, medicaments, and obturation materials from accessing dentinal tubules, increasing the risk of bacterial infection and microleakage [3]. Despite some debate regarding whether it should be preserved or removed, most studies suggest that removing it without causing erosive damage will enhance the root canal system's 'fluid-tight seal' [4]. Currently, decalcifying agents like ethylenediaminetetraacetic acid (EDTA), doxycycline, citric acid, and BioPure MTAD remove the inorganic part while the dissolution of the organic part is by sodium hypochlorite (NaOCl) [5]. As a final rinse, 17% EDTA forms soluble calcium chelates when it reacts with the divalent calcium ions on the dentinal surfaces, thereby removing the smear layer [6]. Similarly, MTAD, which contains 4.25% citric acid as well as an aqueous solution of 3% doxycycline and 0.5% polysorbate 80 detergent (Tween 80), has been shown to be effective in removing the smear layer, eliminating microbes, and antibacterial action that is maintained due to doxycycline's binding affinity for hard tissues [7]. Combined use of NaOCl and MTAD had better smear layer removal ability and caused minimal damage to root dentin as compared to the use of NaOCl and EDTA [5,8]. Plasma, the fourth state of matter, is a quasi-neutral collection consisting of neutral species and charged particles [9]. Cold Atmospheric Pressure (CAP) Plasma sources have

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emerged as a viable clinical alternative for various medical and dental purposes. This highly reactive plasma can be delivered using plasma pencils, plasma needles, plasma brushes, and Dielectric Barrier Discharge (DBD) plasma jets to produce a clean, etched surface and improve bonding to various substrates, or it can be combined to produce a thin layer of plasma coating, altering the surface properties [10]. Numerous studies have shown that CAP Plasma can cause surface alterations on dentin, cause opening of dentinal tubules and also make it more hydrophilic [11,12]. Based on these studies CAP Plasma Jet could be a innovative chemical free alternative for smear layer removal in the intra-radicular dentin. Hence the research question was does a cold atmospheric pressure plasma jet remove smear layer efficiently enough on intra-radicular dentine surfaces, when compared to conventional methods? The null hypothesis for the study was that there is no significant difference in the efficacy of smear layer removal on intra-radicular dentinal surfaces using Cold Atmospheric Pressure (CAP) Plasma Jet, when compared with MTAD and EDTA.

## Materials and methods

### Sample size estimation

A priori sample size analysis was conducted using PWR2 package in R software to test the difference between twelve independent groups. Two-way ANOVA was used with effect size of 0.4 and 0.2 for treatment factor and location factor, respectively. The statistical power assumed is 0.8 at significance level of 0.05. The calculation using the aforementioned criteria provided that a minimum of 252 test samples (21 per sub-group) are required to achieve a significance of 0.05 at 80% statistical power of the study. So, total 84 teeth were collected.

### Sample collection and storage

Ethical approval was obtained from our Institutional Ethical Committee. Eighty-four single rooted mandibular 1st and 2nd premolar teeth with single canal, freshly extracted for orthodontic or periodontal reasons were collected. The selected teeth were devoid of caries, cracks, endodontic treatments, restorations, or any calcification, extra canals, internal and external resorptions, root caries and open apices. Teeth with intact and mature root apices, having a single root canal and single canal orifice and apical foramen, with a degree of root curvature ≤15°, as determined by the Schneider method [13], were included. The samples were anatomically matched based on radiographs by exposing to mesiodistal and buccolingual angulated intraoral periapical radiograph to find samples with circular canal anatomy and to understand the similarity of the samples. The samples that had unusual anatomy, calcification, resorption and severe curvature were not included. The tooth samples were immersed in 3% NaOCl for 24 h, cleaned using an ultrasonic scaler, autoclaved (Dental autoclave SW PLUS series) at 121 °C, 15 lbs, and eventually stored in 0.9% normal saline at room temperature until used.

### Root canal preparation

The samples were decoronated with a diamond disc (DFS, Germany), under cooling with distilled water to prevent the crack generation. The root length was standardised to 12 mm and mounted on wax blocks, ensuring apical 2 mm of the root was visible. The samples were then divided into one control and three experimental groups, with twenty-one samples per group. The canals were enlarged up to 15k file (Dentsply M access K files), then pulp extirpation was done with a barbed broach (Dentsply, Maillefer, India) and patency was established using a 15k file.

Estimation of working length was done by subtracting 0.5 mm from the length recorded when the tip of 15k file was just visible at the apical foramen. Chemo-mechanical preparation was done by a crown-down technique using hand ProTaper (Dentsply, Maillefer, India) files up to F3. After each file sequence during preparation, the canals were rinsed with 5 millilitres of 5.25% NaOCl) (SafeEndo Dental India Pvt. LTD) using a 30 G side-vented irrigation needle (ENDO-TOP Endo irrigation needles) that was positioned 2 millimetres short of the apex within 50-60 s with the flow rate of 0.1 ml/s approximately. The apical foramen was sealed with sticky wax to prevent the irrigating solution from passing from the apical foramen. The samples were irrigated with 5 ml Normal Saline (B|Braun group company, India), 5 ml EDTA (Neelkanth Healthcare (P.) LTD, India) and 5 ml MTAD solution (Biopure, Dentsply Sirona) for a period of 2 min constituting the control, EDTA and MTAD group, respectively. The samples in the CAP Plasma Jet group were exposed to the plasma plume, keeping the nozzle 1 cm from the canal lumen for a period of 2 min. The canals were dried with paper points (Dentsply, Maillefer, India), and the canal orifice was sealed with sticky wax.

### Specimen preparation

Longitudinal grooves were made on the buccal and lingual aspects of each sample by using a diamond disk at low speed, under cooling with distilled water, without penetrating the canal. The osteotome was used to split the teeth along the grooves into two halves. The half containing the major part of the root canal was prepared for SEM evaluation. The sectioned parts were immersed for tissue fixation in 2% glutaraldehyde with phosphate buffer (pH = 7.3) for 12 h. The specimens were then washed with 20 ml of phosphate buffer for 15 min, followed by dehydration in a graded ethanol series. The specimens were then dried in a Zero-point desiccator. The specimens were then sputter coated with a Platinum film, followed by observation under an SEM (Carl Zeiss, EVO18 Special Edition) at 20.00 kV.

### SEM evaluation

The total test samples in each group ($n = 21$) were further subdivided into three different sub-groups based on the location (coronal, middle and apical third) for SEM evaluation at 3000x magnification. Each sample was divided into thirds, after marking using vernier callipers, having 4 mm of root surface in each third (longitudinally). So, SEM image was taken at 2, 6 and 10 mm from the apex in the canal lumen. The central zone of each third was focused under 10x magnification and then it was imaged under 3000x magnification.

Each photograph captured of the coronal, middle and apical third of the canal was then divided into 16 equal subareas by overlaying a grid. Each subarea of an image was evaluated using the scoring system suggested by Prado et al. (2011) [14], which are as follows:-

Score 1: No smear layer and debris at all, with all tubules clean and open.

Score 2: A few areas are covered by smear layer and debris, with most tubules clean and open.

Score 3: Smear layer and debris covering almost all the surfaces, with few tubules open.

Score 4: Smear layer and debris covering all the surfaces.

The scoring of each subarea was done by two trained and experienced examiners, and any disagreement between the two examiners was solved after a discussion with the third examiner (Principal Investigator). The number of subareas with scores 1 and 2 were further combined to get a final score representing the area with either no smear layer/debris or a few areas covered by smear layer/debris. The final score of each test sample in different groups and subgroups was used for statistical analysis (Figure 1).

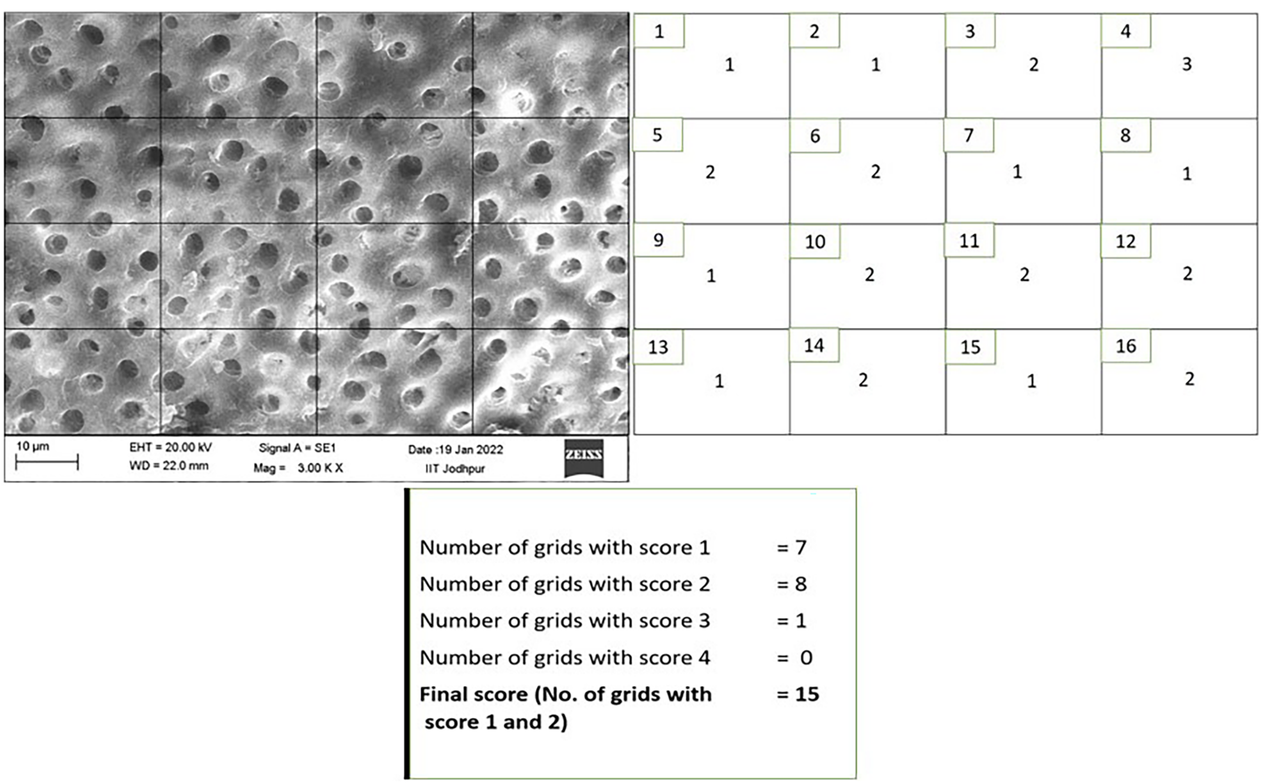


**Figure 1.** Sample scoring of the SEM image.

**Table 1.** Inter-group comparison of median values of the final scores in control, EDTA, MTAD and CAP Plasma Jet.

| | | P value (Pairwise groups) | | |
|---|---|---|---|---|
| Group | Median (IQR) | EDTA | MTAD | CAP |
| Control | 0.0 (0.0, 0.0) | $2.32\times10^{-10}$ | $3.64\times10^{-15}$ | 0.06 |
| EDTA | 12.0 (0.0, 16.0) | | 0.83 | $3.27\times10^{-4}$ |
| MTAD | 16.0 (0.5, 16.0) | | | $2.1\times10^{-7}$ |
| CAP | 0.0 (0.0, 8.0) | | | |

IQR- Interquartile Range, P-value < 0.05 is significant.

### *Statistical analysis*

Inter-examiner reliability by Cohen's Kappa statistics was found to be 0.85. Kolmogorov–Smirnov test was applied to assess the normality of the data. The data followed a Non-normal distribution. Data were analyzed by Kruskal–Wallis test and Dunn's test for pairwise comparison between groups in R studio (version = 4.2.1) data analysis software. A p-value of less than 0.05 was considered statistically significant.

## Results

Table 1 shows the comparison of smear layer removal from the canal lumen amongst the groups. The two experimental groups, EDTA and MTAD, had the maximum median values of the final scores and they were significantly better than the control and CAP Plasma Jet group in smear layer removal (P value < 0.05). There was no discernible difference between the EDTA and MTAD groups (P value > 0.05), nor between the CAP Plasma Jet and control groups (P value > 0.05).

Table 2 shows the comparison of smear layer removal within the groups comparing the coronal, middle and apical third of the canal surface. In the MTAD group, maximum smear layer removal was seen in the coronal third, followed by a middle and apical third, with a statistically significant difference observed only between coronal v/s apical third ($p<0.05$). EDTA showed maximum smear layer removal in the coronal and middle third, both being statistically significant when compared with that in the apical third ($p<0.05$). In the CAP Plasma Jet group, maximum smear layer removal was seen in the coronal third, followed by the middle third. The smear layer removal ability of CAP Plasma Jet was statistically significant when a comparison was made between the coronal v/s middle third and the coronal v/s the apical third ($p<0.05$).

Table 3 shows intergroup comparison of median values of final scores in the coronal, middle and apical third. All three experimental groups were significantly better than the control group in the coronal third (P value < 0.05). MTAD and EDTA had the highest median value of final scores indicating maximum smear layer removal ability was shown by them. The above two groups showed comparable results in the coronal, middle and apical third (P value > 0.05). The smear layer removal ability of CAP Plasma Jet group was comparable to EDTA in the coronal third (P value > 0.05), while in the middle and apical third, it was as good as the control group (P value > 0.05).

**Table 2.** Intra group comparisons of the median values of the final scores in coronal, middle and apical third of the samples analyzed.

| Group | Region | Median (IQR) | P-value (All sub-groups) | P value | |
|---|---|---|---|---|---|
| | | | | Middle | Apical |
| Control | Coronal third | 0.0 (0.0, 0.0) | 0.05 | 0.0933 | 0.0933 |
| | Middle third | 0.0 (0.0, 0.0) | | | 1 |
| | Apical third | 0.0 (0.0, 0.0) | | | |
| EDTA | Coronal third | 15.0 (7.0, 16.0) | 0.0001 | 1 | 0.0005 |
| | Middle third | 16.0 (6.0, 16.0) | | | 0.0007 |
| | Apical third | 0.0 (0.0, 5.0) | | | |
| MTAD | Coronal third | 16.0 (16.0, 16.0) | 0.0009 | 0.4125 | 0.0006 |
| | Middle third | 16.0 (5.0, 16.0) | | | 0.0805 |
| | Apical third | 0.0 (0.0, 16.0) | | | |
| CAP | Coronal third | 8.0 (0.0, 14.0) | $8.12\times10^{-6}$ | $5.59\times10^{-3}$ | $5.57\times10^{-3}$ |
| | Middle third | 0.0 (0.0, 0.0) | | | 0.292 |
| | Apical third | 0.0 (0.0, 0.0) | | | |

IQR- interquartile range, P-value < 0.05 is significant.

## Discussion

The chemo-mechanical preparation of the root canal inevitably results in the formation of an irregular amorphous layer, smear layer that covers the root canal walls [15]. In a review, Shahravan et al. (2007) determined that removing the smear layer improves the fluid-tight seal following root canal obturation. According to Violich and Chandler's recent assessment of the smear layer, removal of the smear layer resulted in more thorough disinfection of the root canal system and better adaptation of filling materials to the canal walls [3].

The chemomechanical preparation generally involves the use of Sodium hypochlorite, which is the most widely recommended irrigant in endodontics on the basis of its unique capacity to dissolve necrotic tissue remnants and excellent antimicrobial potency [16]. However, sodium hypochlorite even at concentrations of 5.25% was only

**Table 3.** Inter-group comparison of median values of the final scores in coronal, middle and apical third regions.

| Region | Group | Median (IQR) | P value (All groups) | P value (Pairwise groups) EDTA | MTAD | CAP |
|---|---|---|---|---|---|---|
| Coronal third | Control | 0.0 (0.0, 0.0) | | $1.09 \times 10^{-4}$ | $3.80 \times 10^{-8}$ | $3.46 \times 10^{-2}$ |
| | EDTA | 15.0 (7.0, 16.0) | $5.02 \times 10^{-8}$ | | 0.7683 | 0.7633 |
| | MTAD | 16.0 (16.0, 16.0) | | | | 0.0138 |
| | CAP | 8.0 (0.0, 14.0) | | | | |
| Middle third | Control | 0.0 (0.0, 0.0) | | $4.00 \times 10^{-6}$ | $1.04 \times 10^{-6}$ | 1.00 |
| | EDTA | 16.0 (6.0, 16.0) | $4.19 \times 10^{-9}$ | | 1.00 | $1.00 \times 10^{-3}$ |
| | MTAD | 16.0 (5.0, 16.0) | | | | $3.54 \times 10^{-4}$ |
| | CAP | 0.0 (0.0, 0.0) | | | | |
| Apical third | Control | 0.0 (0.0, 0.0) | | 0.0254 | 0.0020 | 1.0000 |
| | EDTA | 0.0 (0.0, 5.0) | $9.27 \times 10^{-5}$ | | 1.0000 | 0.0254 |
| | MTAD | 0.0 (0.0, 16.0) | | | | 0.0020 |
| | CAP | 0.0 (0.0, 0.0) | | | | |

IQR- interquartile range, P-value < 0.05 is significant.

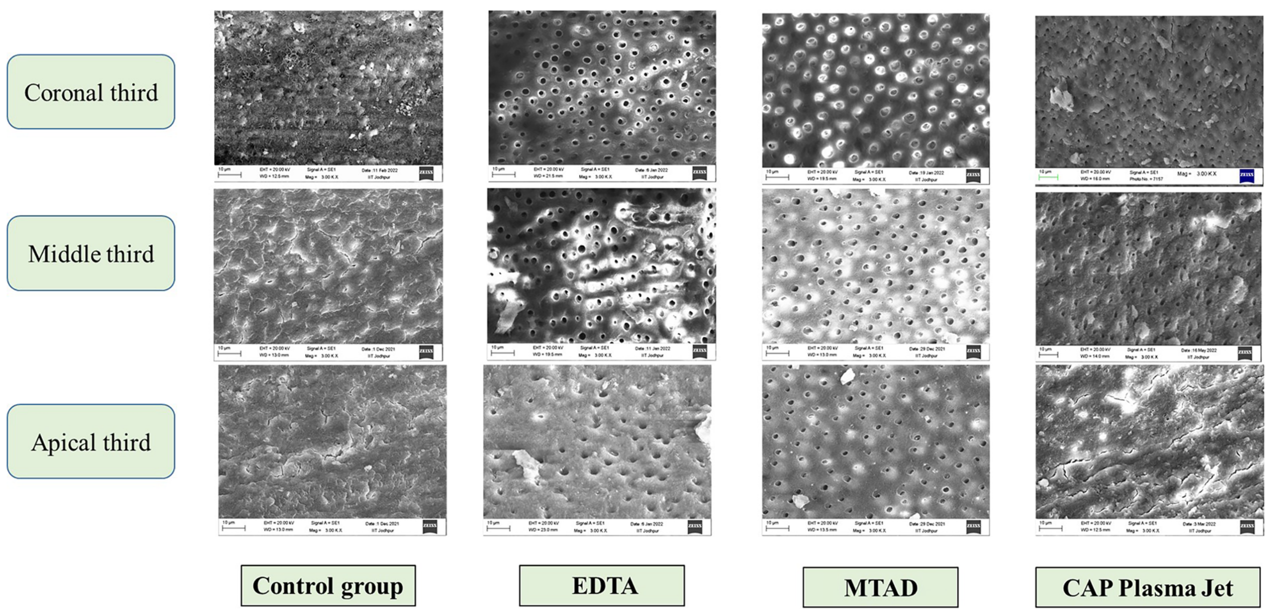


**Figure 2.** Representative SEM images of the samples analyzed.

able to remove the organic component of the smear layer [17]. So, there is a need for chelating agents to remove the inorganic component. The presently employed techniques for smear layer removal include chemical methods, i.e. the use of chelating agents.

Lehmann A. et al. (2013) and Dong X et al. (2015) observed surface changes in dentin after CAP-Plasma treatment presenting as opening of the dentinal tubules and smear layer removal from the dentinal surface [12,18]. Plasma treatment can generally change surfaces in two ways: modification and etching [10]. Plasma consists of many energetic and chemically reactive species including high energy electrons, ionic species, electronically excited neutrals, and free radicals, etc. which can react with the treated surface and thus modify the surface chemistry [19].

In the present study, the effectiveness of smear layer removal by CAP Plasma Jet as a final treatment regimen was evaluated when compared to 17% EDTA, MTAD and a control group (normal Saline) when used as a final rinse, following irrigation with 5.25% NaOCl during instrumentation (Figure 2).

MTAD in addition to its smear layer removal ability, has the added advantage of being able to eliminate microorganisms resistant to conventional irrigating solutions, intracanal medication and capable of effective antimicrobial activity due to the affinity of doxycycline in binding to dental hard tissues [20,21]. In comparison to EDTA, MTAD cause minimal erosion of the dentinal tubules when used as a final irrigant in conjunction with 5.25% NaOCl as a root canal irrigant [5].

Scanning electron microscopy (SEM), which has been used to assess the effectiveness of various chemicals in removing the smear layer, is still an effective method to study the morphology of the dentin surface [5]. In the coronal third, maximum smear layer removal was seen with the MTAD group followed by the EDTA, CAP

Plasma Jet and Normal saline group. Interestingly, the EDTA and CAP Plasma Jet groups showed comparable results in the coronal third, i.e. similar cleaning ability with no statistically significant difference in their values. These results may be attributed to greater surface wetting in the coronal third owing to a larger reservoir in the region allowing for extended irrigant contact with the canal walls. CAP Plasma plume was also in close proximation to the coronal third of the canal orifice. The free radicals and chemically reactive species could cause the surface modifications thereby removing the smear layer [19].

In the middle third and apical third, MTAD and EDTA has comparable smear layer removal. CAP Plasma Jet could not produce significant smear layer removal and showed comparable results as the control group. This could be attributed to the geometry of the plasma device and the working gases. Previous studies showing surface alterations in dentin used oxygen gas in conjunction with Helium/Argon which produced better surface alterations in dentin owing the increased production of chemically reactive species and free radicals by $He/O_2$ or $Ar/O_2$ combination.

The findings of the present study are in conjunction with the study by Mancini M. et al. (2009) where efficacy of smear layer removal by MTAD and EDTA showed no significant difference in the coronal, middle and apical third of the root canal [7].

Contrary to our results, study by Mozayeni MA et al. (2009) and Torabinejad M. et al. (2003) showed that efficacy of smear layer removal by MTAD and EDTA showed no significant difference in the coronal and middle third [5,22], while MTAD showed statistically significant better results than EDTA in the apical third. This difference in result from our study could be attributed to the different methodology in the above-mentioned studies. In the study by Mozayeni MA et al. the total exposure time to the final solution was approximately 5 min, while in our study it was 2 min. Dai L. et al. (2011) also concluded that MTAD showed better smear layer removal ability vis a vis EDTA, when the contribution from different thirds of the root canal was taken into consideration [23]. They used 1.3% sodium hypochlorite before final rinse with MTAD, whereas in our study 5.25% sodium hypochlorite was used to maintain standardization amongst the groups. Similar results were seen later in studies by Paul M.L. et al. (2013) and Vemuri S. et al. (2016) [24,25].

On the other hand, research by Lotfi M et al. (2012) and Wu L et al. (2012) demonstrated that EDTA had superior smear layer removal ability when compared to MTAD [26,27]. This could be attributed to a different methodology. Lotfi M et al. used 1.3% Sodium hypochlorite during instrumentation of the canals. While in the study by Wu L et al. they used 1 ml of the chelating agent for a duration of 1 min which could be the factor producing different results. MTAD and EDTA show comparable results in the smear layer removal when the entire canal lumen was considered. CAP plasma Jet showed results comparable to the control group.

This was one of the few studies where the efficacy of plasma plume penetration directly into the root canal was assessed. The results of the study showed deeper action of CAP plasma jet upto complete coronal third (i.e. 4 mm) of the root canals, which was comparatively better result than the previous study by Jiang C et al. wherein the effect of plasma was visible only upto the coronal 1 mm. The present study aimed to remove the observer subjectivity in scoring the SEM images by overlaying each image with 16 grids and scoring each subarea. This would ensure that each subarea (entire SEM image) was considered equally for the scoring purpose.

The present study aimed to assess the smear layer removal ability of the CAP Plasma Jet, which could provide a chemical free method in the form of a device. The major shortcomings of the present study may be attributed to the design of the CAP Plasma Jet which affects the distance of plasma plume where it acts on a surface. Also, the gas used in the plasma jet was Helium, which produces less free radical, reactive oxygen and nitrogen species compared to Helium/Argon-Oxygen based plasma jet, which could have affected the efficacy of CAP Plasma Jet. Hence the present device produced less efficient results even after as long an exposure as 2 min. Further research on the optimization of the plasma device is warranted for efficient smear layer removal. Jet length, jet power, jet volume, and gas flow rate should all be taken into account when modifying the CAP Plasma jet to increase its efficacy. Thus, modification of the Plasma Jet on the above mentioned parameters is necessary so as to be better able to cause surface modifications in the middle and the apical third of the root canal, thereby removing smear layer.

Finally, this study evaluated the smear layer removal using SEM. Future longitudinal examinations of root canal smear layers should be performed with micro-CT to obtain more reproducible, dependable results.

## Conclusion

Both MTAD and EDTA had the highest smear layer removal ability in all thirds of the root canal when the samples were irrigated with 5 ml of the solution for a period of 2 min. CAP Plasma Jet and EDTA showed comparable smear layer removal ability in the coronal third, while in the middle and apical third, CAP Plasma Jet was as good as the control group. Further optimization of the jet and more clinical studies are warranted to incorporate CAP Plasma Jet as a smear layer removal technology.

## Disclosure statement

No potential conflict of interest was reported by the authors.

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## References

[1] McComb D, Smith DC. A preliminary scanning electron microscopic study of root canals after endodontic procedures. J Endod. 1975;1(7):1–9. doi:10.1016/S0099-2399(75)80226-3.
[2] Hargreaves KM, Berman LH, Rotstein I. Cohen's pathways of the pulp 11th Ed. St Louis: Elsevier; 2016. p. 256–257.
[3] Violich DR, Chandler NP. The smear layer in endodontics - a review. Int Endod J. 2010;43(1):2–15. doi:10.1111/j.1365-2591.2009.01627.x.
[4] Shahravan A, Haghdoost AA, Adl A, et al. Effect of smear layer on sealing ability of canal obturation: a systematic review and meta-analysis. J Endod. 2007;33(2):96–105. doi:10.1016/j.joen.2006.10.007.
[5] Torabinejad M, Khademi AA, Babagoli J, et al. A new solution for the removal of the smear layer. J Endod. 2003;29(3):170–175. doi:10.1097/00004770-200303000-00002.
[6] Mohammadi Z, Shalavi S, Jafarzadeh H. Ethylenediaminetetraacetic acid in endodontics. Eur J Dent. 2013;7(Suppl 1):S135–S142. doi:10.4103/1305-7456.119091.
[7] Mancini M, Armellin E, Casaglia A, et al. A comparative study of smear layer removal and erosion in apical intraradicular dentine with three irrigating solutions: a scanning electron microscopy evaluation. J Endod. 2009;35(6):900–903. doi:10.1016/j.joen.2009.03.052.
[8] Singla MG, Garg A, Gupta S. MTAD in endodontics: an update review. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2011;112(3):e70–6–e76. doi:10.1016/j.tripleo.2011.02.015.
[9] Jiang C, Schaudinn C, Jaramillo DE, et al. In vitro antimicrobial effect of a cold plasma jet against Enterococcus faecalis biofilms. ISRN Dent. 2012;2012:1–6. doi:10.5402/2012/295736.
[10] Ritts AC, Li H, Yu Q, et al. Dentin surface treatment using a non-thermal argon plasma brush for interfacial bonding improvement in composite restoration. Eur J Oral Sci. 2010;118(5):510–516. doi:10.1111/j.1600-0722.2010.00761.x.
[11] Jiang C, Chen MT, Gorur A, et al. Nanosecond pulsed plasma dental probe. Plasma Processes and Polymers. 2009;6(8):479–483. doi:10.1002/ppap.200800133.
[12] Lehmann A, Rueppell A, Schindler A, et al. Modification of enamel and dentin surfaces by non-thermal atmospheric plasma. Plasma Processes Polym. 2013;10(3):262–270. doi:10.1002/ppap.201200088.
[13] Malur MH, Chandra A. Curvature height and distance of MB canal of mandibular molar with schneider angle and its comparison with canal access angle. J Oral Biol Craniofac Res. 2018;8(3):212–216. doi:10.1016/j.jobcr.2017.07.002.
[14] Prado M, Gusman H, Gomes BPFA, et al. Scanning electron microscopic investigation of the effectiveness of phosphoric acid in smear layer removal when compared with EDTA and citric acid. J Endod. 2011;37(2):255–258. doi:10.1016/j.joen.2010.11.011.
[15] Torabinejad M, Handysides R, Khademi AA, et al. Clinical implications of the smear layer in endodontics: a review. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2002;94(6):658–666. doi:10.1067/moe.2002.128962.
[16] Zehnder M. Root canal irrigants. J Endod. 2006;32(5):389–398. doi:10.1016/j.joen.2005.09.014.
[17] Ciucchi B, Khettabi M, Holz J. The effectiveness of different endodontic irrigation procedures on the removal of the smear layer: a scanning electron microscopic study. Int Endod J. 1989;22(1):21–28. doi:10.1111/j.1365-2591.1989.tb00501.x.
[18] Dong X, Li H, Chen M, et al. Plasma treatment of dentin surfaces for improving self-etching adhesive/dentin interface bonding. Clin Plasma Med. 2015;3(1):10–16. doi:10.1016/j.cpme.2015.05.002.
[19] Dong X, Ritts AC, Staller C, et al. Evaluation of plasma treatment effects on improving adhesive-dentin bonding by using the same tooth controls and varying cross-sectional surface areas. Eur J Oral Sci. 2013;121(4):355–362. doi:10.1111/eos.12052.
[20] Torabinejad M, Cho Y, Khademi AA, et al. The effect of various concentrations of sodium hypochlorite on the ability of MTAD to remove the smear layer. J Endod. 2003;29(4):233–239. doi:10.1097/00004770-200304000-00001.
[21] Shabahang S, Torabinejad M. Effect of MTAD on Enterococcus faecalis–contaminated root canals of extracted human teeth. J Endod. 2003 Sep;29(9):576–579. doi:10.1097/00004770-200309000-00008.
[22] Mozayeni MA, Javaheri GH, Poorroosta P, et al. Effect of 17% EDTA and MTAD on intracanal smear layer removal: a scanning electron microscopic study. Aust Endod J. 2009;35(1):13–17. doi:10.1111/j.1747-4477.2007.00111.x.
[23] Dai L, Khechen K, Khan S, et al. The effect of QMix, an experimental antibacterial root canal irrigant, on removal of canal wall smear layer and debris. J Endod. 2011;37(1):80–84. doi:10.1016/j.joen.2010.10.004.

[24] Paul ML, Mazumdar D, Niyogi A, et al. Comparative evaluation of the efficacy of different irrigants including MTAD under SEM. J Conserv Dent. 2013;16(4):336–341. doi:10.4103/0972-0707.114367.

[25] Vemuri S, Kolanu S, Varri S, et al. Effect of different final irrigating solutions on smear layer removal in apical third of root canal: a scanning electron microscope study. J Conserv Dent. 2016;19(1):87–90. doi:10.4103/0972-0707.173207.

[26] Lotfi M, Vosoughhosseini S, Saghiri MA, et al. Effect of MTAD as a final rinse on removal of smear layer in ten-minute preparation time. J Endod. 2012;38(10):1391–1394. doi:10.1016/j.joen.2012.06.027.

[27] Wu L, Mu Y, Deng X, et al. Comparison of the effect of four decalcifying agents combined with 60 °C 3% sodium hypochlorite on smear layer removal. J Endod. 2012;38(3):381–384. doi:10.1016/j.joen.2011.11.013.