Original Article

Anti-Microbial Efficacy of Root Canal Preparation in Deciduous Teeth with Manual and Rotary Files: A Randomized Clinical Trial

L Lakshmanan, G Jeevanandan, S Vishwanathaiah[1], Prabhadevi C Maganur[1], KJ Alzahrani[2], A Alkahtani[3], N Boreak[4], L Testarelli[5], HA Baeshen[6], S Patil[7]

Department of Pediatric and Preventive Dentistry; Saveetha Dental College, Chennai, Tamil Nadu, India, [1]Department of Preventive Dental Sciences, Division of Pediatric Dentistry, College of Dentistry, Jazan University, [4]Department of Restorative Dental Sciences, College of Dentistry, Jazan University, [7]College of Dental Medicine, Roseman University of Health Science, South Jordan, UTAH-84095, USA, College of Dentistry, Jazan University, Jazan, [2]Department of Clinical Laboratories Sciences, College of Applied Medical Sciences, Taif University, Taif, [3]Department of Restorative Dental Sciences, College of Dentistry, King Saud University, Riyadh, Saudi Arabia, [5]Department of Oral and Maxillofacial Sciences, Sapienza University, University of Rome, Rome, Italy, [6]Department of Orthodontics, College of Dentistry, King Abdulaziz University, Jeddah, Saudi Arabia

Received: 28-Jan-2022;
Revision: 25-Mar-2022;
Accepted: 19-Apr-2022;
Published: 26-Oct-2022

ABSTRACT

Background: In a pulpectomy, the eradication of microbes from the primary root canal is accomplished through biomechanical preparation, which could be carried out with either manual or rotary instruments. **Aims:** The objective of this clinical trial was to evaluate the efficiency of manual K-files, H-files, and Kedo-S Square rotary files in reducing microbial flora after canal preparation in primary molars. **Materials and Methods:** This randomized clinical trial consisted of 45 primary molars requiring pulpectomy. The teeth were randomly allocated to one of the three groups: Group I: Manual K-files, Group II: Manual H-files, and Group III: Kedo-S Square rotary files, based on the type of instrumentation. Pre-and Post-instrumentation sampling was performed using clean absorbent paper points and kept in a clean Eppendorf tube having thioglycolate broth as the transport medium. Culturing was performed on agar media from which both aerobic and anaerobic microbial counts were estimated. Collected data were statistically analyzed using one-way analysis of variance (ANOVA) and Wilcoxon signed-rank test. Following root canal preparation, 87–89% reduction of the aerobic and anaerobic microbial load was noted in group I, whereas it was an 89–92% reduction in group II and a 93–95% reduction in group III. **Results:** Biomechanical preparation with Kedo-S Square rotary file showed higher efficacy in microbial reduction compared to manual instrumentation. **Conclusion:** Manual and rotary files were equally effective in removing root canal microbes. Biomechanical preparation with a KedoS Square rotary file resulted in greater microbial efficacy. Hence In children, effective root canal cleaning in a short period of time is a major consideration.

KEYWORDS: *Hand files, microbial, primary teeth, root canal preparation, rotary files*

INTRODUCTION

Root canal therapy of deciduous teeth is a routine procedure in pediatric dentistry that is carried out

Access this article online

Quick Response Code:

Website: www.njcponline.com

DOI: 10.4103/njcp.njcp_71_22

Address for correspondence: Dr. S Patil, College of Dental Medicine, Roseman University of Health Science, South Jordan, UTAH-84095, USA. E-mail: dr.ravipatil@gmail.com

How to cite this article: Lakshmanan L, Jeevanandan G, Vishwanathaiah S, Maganur PC, Alzahrani KJ, Alkahtani A, *et al.* Anti-microbial efficacy of root canal preparation in deciduous teeth with manual and rotary files: A randomized clinical trial. Niger J Clin Pract 2022;25:1681-6.

to eliminate microorganisms and prevent infection subsequently. It is the only treatment to preserve the teeth that exhibit pulp necrosis.[1] A pulpectomy is a process that entails mechanically removing all infected and dead pulp tissue from the root canals of primary teeth, then cleaning, shaping, and disinfecting the root canals with irrigants, and finally obturating the canals with a resorbable material.[2]

The success of pulpectomy is defined by the lack of clinical and radiographic signs and symptoms. The determining factors begin from access opening, biomechanical preparation, and root canal obturation in three dimensions.[3] In many cases, procedural errors do not imperil the result of endodontic therapy, except if a concomitant infection is present. Residual infection remains to be a significant factor for pulpectomy failure. Henceforth, complete elimination of infection is essential for the positive outcome of pulp therapy.[4]

Incomplete root canal disinfection can result from a morphological difference that persists in the apical portion of the primary roots.[5] This necessitates the use of appropriate instrumentation to effectively remove bacteria, provide sterile canals, and improve obturation quality. Deficient removal of microbes, ledge formation, and increased instrumentation time are some of the many limitations of the traditional manual instrumentation systems.[6] However, rotary instruments with a uniform taper and the capacity to turn around the center point by maintaining the standard taper of primary root canals, lead to their establishment in endodontic therapy of deciduous teeth.[2,6]

The effectiveness of rotary instruments in terms of the quality of root filling, preparation time, and postoperative pain following pulpectomy in primary teeth has been studied in the literature. Only limited studies have assessed the efficiency of rotary files in eradicating the pathogenic microbes in the primary root canals. A recently introduced pediatric single rotary file system (Kedo-S Square) with variable taper preserving the root dentin has been widely utilized.[7] Due to the paucity of studies concerning the cleaning efficiency of the Kedo-S Square single pediatric rotary system in primary teeth regarding microbial assessment, this clinical trial was undertaken to assess the antimicrobial efficiency of root canal preparation with manual K-files, H-files, and Kedo-S Square rotary files in primary molars.[8]

Materials and Methods

The ethical approval for this clinical trial was attained from the Institutional Review Board of Saveetha Dental College (SRB/SDC/PEDO-1803/20/02). The trial was registered at clinical trials.gov (CTRI/2021/02/031400) and after explaining the purpose and process of the study, all parents/caregivers on behalf of the children gave their written consent to participate in the trial.

Sample size and selection of participants

Based on the data from a previous study,[9] a sample size of 45 was obtained with 95% power using G-power. Inclusion criteria were 1) cooperative children aged between 4 and 9 years necessitating pulpectomy in any of the primary molars diagnosed as having irreversible pulpitis; 2) with no significant medical condition; 3) without any antibiotic exposure in the previous two weeks; 4) primary teeth with adequate coronal structure and minimum of two-thirds root structure; and 5) without sinus tract and pathological root resorption.

Between February 2021 and March 2021, 45 children who necessitated single-visit pulpectomy in deciduous molars were chosen from Outpatients, Department of Pedodontics. Following an extensive clinical and radiographic investigation, the participants were randomly allocated to one of three groups.

Clinical procedure

After administering local anesthesia containing 2% lignocaine in 1:200,000 adrenaline (LOX*2% ADRENALINE, Neon Laboratories Limited, India), the selected tooth was isolated with a rubber dam (GDC marketing, India). Five percent iodine solution was used to disinfect the rubber dam and tooth to phase out the microbial contamination from the operative field. The access was opened with sterile no. 6 round bur (Mani, Inc., Japan) in the high-speed handpiece, and a non-end cutting bur was used for removing the roof of the pulp chamber. Pulp extirpation was done using a barbed broach, and the working length of the canals was measured using no. 15 size K-file (Mani, Inc., Tochigi Japan) in all three groups using Ingle's method.

The distal canal of the mandibular primary molar and the palatal canal of the maxillary primary molar were selected for sampling. Preceding sample collection, the orifices of other canals were sealed with Cavit (3M ESPE), a temporary restorative material, to preclude any contamination. The pre-instrumentation sample was obtained by placing a 15-size clean absorbent paper point (Maillefer, OK, USA) into the root canal for 1 min until it reached the specified working length. The collected sample was stored in a clean Eppendorf tube having 1 mL of thioglycolate broth as the transport media.

Based on the groups assigned, biomechanical preparation was performed using either manual or rotary instrumentation until the working length after the pre-instrumentation sampling.

Group I (15 teeth)- Manual K- file (Mani, Inc., Tochigi, Japan) group:

The canals were instrumented using a quarter-turn pull (Step-back technique) with Hand K-files. The canals were instrumented to 35 sizes until the working length.

Group II (15 teeth)-Manual H-file (Mani, Tochigi, Japan) group:

The canals were instrumented using conventional H-files up to no. 35 size file using retraction motion (Step-back technique).

Group III (15 teeth)-Kedo-S Square rotary file (KEDO Dental, India) group:

Post-working length measurement using hand k-files, the canals in the rotary file group were prepared using the single file system Kedo-S Square rotary file in brushing motion (Crown-down technique) using X-Smart endodontic motor (Dentsply Maillefer, OK, USA) at 300 rpm and 2.2 N cm torque.

The lubricant used during the use of rotary or manual instrumentation was Endo prep RC (Anabond Stedman, Pharma Research Ltd., India). After instrumentation, 5 mL of sterile saline solution (Fresenius Kabi India, Pvt. Ltd) was used for irrigation of the canal. After that, the post-instrumentation sampling was done using a no. 25 size clean absorbent paper point, as portrayed previously. The leftover canals were then prepared as described earlier and dried with paper points of suitable size. Obturation was done with Metapex (Meta Biomed Co. Ltd., Korea) and the tooth was restored with type II glass ionomer cement (GC, India) and stainless steel crown (3M ESPE, USA).

Processing of sample

Both pre- and post-instrumentation samples were then transferred to the microbiology laboratory for culturing. The samples were gradually diluted with physiologic saline (Fresenius Kabi India, Pvt. Ltd.) to attain 10^{-4} dilution. One mL each from the dilution was inoculated onto thioglycolate agar plates and blood agar plates. Subsequently, in an anaerobic jar (Dextrose Technology Pvt. Ltd., Bengaluru, India) containing 5% hydrogen, 85% carbon dioxide, and 10% nitrogen, the thioglycolate agar plates were incubated at 37°C for a week. Aerobic incubation of blood agar plates was performed at 37°C for 36 h. Figure 1 shows the pre- and post-instrumentation samples collected from a tooth included in the study. An investigator who was blinded to the groups studied and documented the aerobic and anaerobic microbial counts using a digital colony counter (Labline, Mumbai, India) that was expressed as CFU/mL [Figure 2].

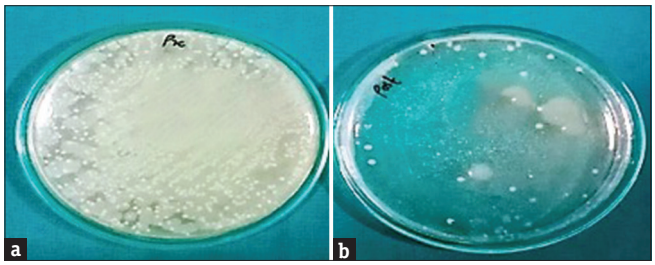

Figure 1: (a) Culture media before instrumentation; (b) Culture media after instrumentation

Statistical analysis

The results were tabulated and analyzed in SPSS software version 23.0 (SPSS Inc., USA) by a statistician who was blinded to the study framework. Wilcoxon test was used for comparing the mean values between pre- and post-instrumentation samples. One-way analysis of variance (ANOVA) was performed to find the statistical difference among the three groups. The level of significance was set at $P < 0.05$.

Results

Post-instrumentation, there was a significant reduction in the microflora (CFU/mL) of the canals in all three groups ($P < 0.05$) [Table 1]. There was an 89% reduction in the aerobic microbial count and an 87% reduction in the anaerobic microbial count in group I (preparation with conventional K-files). In group II (preparation with conventional H-files), the percentage of reduction was 92% for aerobic and 89% for an anaerobic microbial load. In group III (rotary files), there was a reduction of 93% and 95% in the aerobic and anaerobic bacterial count, respectively. In comparison, group III followed by group II and group I were efficient in reducing the microbial load. However, the difference between the three groups was not statistically significant ($P > 0.05$).

Figure 2: Digital colony counter utilized for estimating microbial count

Table 1: Mean microbial count in the three groups

Groups	Microflora	Sample	Mean (CFU/mL)	Std. deviation	Mean difference (CFU/mL)	Microbial reduction (%)	*P*
K-files	Aerobic	Pre-instrumentation	0.513×10^5	0.167×10^5	0.460×10^5	89	0.001*
		Post-instrumentation	0.053×10^5	0.020×10^5			
	Anaerobic	Pre-instrumentation	1.560×10^5	0.674×10^5	1.362×10^5	87	0.001*
		Post-instrumentation	0.197×10^5	0.411×10^5			
H-files	Aerobic	Pre-instrumentation	1.270×10^5	0.642×10^5	1.164×10^5	92	0.001*
		Post-instrumentation	0.106×10^5	0.080×10^5			
	Anaerobic	Pre-instrumentation	1.742×10^5	0.762×10^5	0.155×10^5	89	0.001*
		Post-instrumentation	1.183×10^5	0.122×10^5			
Kedo-S2 files	Aerobic	Pre-instrumentation	0.418×10^5	0.207×10^5	0.395×10^5	93	0.001*
		Post-instrumentation	0.022×10^5	0.015×10^5			
	Anaerobic	Pre-instrumentation	0.921×10^5	0.566×10^5	0.854×10^5	95	0.001*
		Post-instrumentation	0.067×10^5	0.025×10^5			

*$P<0.05$ (statistically significant)

Discussion

The purpose of carrying out pulpectomy in a primary tooth is to eradicate microbes and inhibit the recurrence of infection, thereby favoring the healing of the peri-radicular structure and decreasing the child's pain and displeasure.[5,10] Eradication of microbes from primary tooth roots is accomplished through biomechanical preparation, which could be carried out with either manual or rotary instruments.[9]

In the current study, the recently introduced pediatric rotary file system (Kedo-S Square) was compared with the widely used conventional K files and H files. Instruments with small diameters and predefined conic forms are widely preferred to perform pulpectomy in primary teeth.[11] One such system that combines this design with great flexibility and progressive taper is the Kedo-S Square rotary files. The brushing motion and clockwise rotation of files remove the pulp and debris off the root canals. As it is a single file system with a smaller diameter, it results in less root dentin preparation, offering less smear layer formation and a reduced rate of primary root resorption, unlike other instruments resulting in excessive dentin preparation.[12]

It is well established in the literature that for almost any microorganism to infect, it must achieve a proportional amount that favors harm, whether induced by the microorganism or by impaired host immunity reaction to pathogens.[13] Endodontic procedures should ideally eradicate entire microbes present in the root canals. However, considering the complicated configuration of the primary root canals, it is accepted that, with available instrumentations and methods, attaining this objective is not possible in the majority of conditions.[13,14] In the current study, reduction of overall aerobic and anaerobic microorganisms was taken into consideration for evaluating the efficacy of the files as the attainable aim is to only lower the microbial load below the threshold that is essential to initiate or endure the disease.

Culturing permits all viable microorganisms in a sample to grow and it is estimated as colony-forming units (CFU).[15] As the endodontic microflora is intricate and diverse, it is essential to utilize a variety of culture media containing different nutrients and incubation under varied conditions (aerobic and anaerobic) for the isolation of vast microorganisms from the root canal.[16] Culturing is preferred to molecular procedures as the contamination during the laboratory process may be more readily identified.[15]

Because this trial was aimed at assessing the efficacy of instruments on microbial flora, only saline irrigation protocol was followed to prevent any prejudice caused by using antimicrobial agents. Byström A and Sundqvist G. reported more reduction in the mean bacterial count by utilizing saline as an irrigating agent.[17] Both pre- and post-instrumentation samples were collected from the same single canal in every tooth after covering the orifice of other canals using temporary restoration to prevent cross-contamination. Paper points were utilized for collecting samples as they can absorb the substances and have the ability to reach the apex of the root canal, which is considered the least instrumented zone during biomechanical preparation.[18]

In the present study, significantly higher counts of anaerobic microbial flora were observed in all samples obtained, which was consistent with the studies conducted by Ledezma-Rasillo *et al*. and Subramaniam *et al*. on primary molar teeth, where 68% of the samples showed poly-microbial nature.[9,19] A significant microbial

reduction was observed in post-instrumentation samples of all file groups compared to pre-instrumentation samples, but neither instrumentation were able to render the root canals free of microorganisms. This was analogous to the findings of Dalton *et al*.,[20] who showed a significant microbial reduction with each type of instrument technique followed. Manual K and H files are reported to suppress microbial colonies by a factor of 100 to 1000 folds.[21] In the current study, rotary files were more efficient in decreasing the microbial load when compared to manual K and H files. The same was agreed upon by Subramanyam *et al*.[22] on an aerobic bacterial count in primary root canals.

In this study, investigation with only one sort of rotary file and estimation of colony-forming units through conventional methods instead of utilizing various recent technologies can be considered as limitations. Further studies identifying the reduction of specific microbes in primary root canals with different file systems and irrigation protocols are warranted.

Conclusions

Under the conditions of this clinical trial, both manual and rotary files were significantly effective in eradicating root canal microbes. Biomechanical preparation with Kedo-S Square pediatric rotary file showed higher efficacy in microbial reduction compared to manual instrumentation. Effective cleaning of root canals in decreased duration is a significant consideration in children.

Acknowledgments

All authors have read and agreed to the published version of the manuscript.

Informed consent statement

Informed consent was obtained from all subjects involved in the study

Institutional review board statement

Not applicable.

Data availability statement

No new data were created or analyzed in this study. Data sharing does not apply to this article.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1. Figdor D, Sundqvist G. A big role for the very small--understanding the endodontic microbial flora. Aust Dent J 2007;52 (1 Suppl):S38-51.
2. Canoglu H, Tekcicek MU, Cehreli ZC. Comparison of conventional, rotary, and ultrasonic preparation, different final irrigation regimens, and 2 sealers in primary molar root canal therapy. Pediatr Dent 2006;28:518-23.
3. Figini L, Lodi G, Gorni F, Gagliani M. Single versus multiple visits for endodontic treatment of permanent teeth: A Cochrane systematic review. J Endod 2008;34:1041-7.
4. Punathil S, Bhat SS, Bhat SV, Hegde SK. Original research article microbiolgical analysis of root canal flora of failed pulpectomy in primary teeth. IntJCurrMicrobiolAppSci 2014;3:241-6.
5. Tay FR, Gu L-S, Schoeffel GJ, Wimmer C, Susin L, Zhang K, *et al*. Effect of vapor lock on root canal debridement by using a side-vented needle for positive-pressure irrigant delivery. J Endod 2010;36:745-50.
6. Barr ES, Kleier DJ, Barr NV. Use of nickel-titanium rotary files for root canal preparation in primary teeth. Pediatr Dent 2000;22:77-8.
7. Lakshmi Lakshmanan GJ. Evaluation of quality of obturation and instrumentation time using kedo-S square file, H file and K file in primary molars– A randomized controlled trial. Int J Pharm Res 2020;12:3507-14.
8. Ellatif SA, Abed NN, Mahfouz AY, Desouky EM, Abd El Hamid DK. Production, purification, and identification of Glycine, N-(m-anisoyl)-methyl ester from Pseudomonas aeruginosa with antimicrobial and anticancer activities. Kuwait J Sci 2021;48.doi: 10.48129/kjs.v48i3.9150.
9. Subramaniam P, Tabrez TA, Babu KLG. Microbiological assessment of root canals following use of rotary and manual instruments in primary molars. J Clin Pediatr Dent 2013;38:123-7.
10. Faria G, Nelson-Filho P, Freitas AC de, Assed S, Ito IY. Antibacterial effect of root canal preparation and calcium hydroxide paste (Calen) intracanal dressing in primary teeth with apical periodontitis. J Appl Oral Sci 2005;13:351-5.
11. Silva LAB, Leonardo MR, Nelson-Filho P, Tanomaru JMG. Comparison of rotary and manual instrumentation techniques on cleaning capacity and instrumentation time in deciduous molars. J Dent Child (Chic) 2004;71:45-7.
12. Dhanyakumar NM, Vasundhara S, Garg S. SEM evaluation of smear layer formation after using three different nickel titanium rotary instru-ments- Endowave, K3 and Protaper-an invitro study. J Endodontol 2010;22:28-38.
13. Siqueira JF, Rôças IN. Clinical implications and microbiology of bacterial persistence after treatment procedures. J Endod 2008;34:1291-301.e3.
14. Sjogren U, Hagglund B, Sundqvist G, Wing K. Factors affecting the long-term results of endodontic treatment. J Endod 1990;16:498-504.
15. Sundqvist G, Figdor D. Life as an endodontic pathogen. Ecological differences between the untreated and root-filled root canals. Endod Top 2003;6:3-28.
16. da Silva LAB, Nelson-Filho P, Faria G, de Souza-Gugelmin MCM, Ito IY. Bacterial profile in primary teeth with necrotic pulp and periapical lesions. Braz Dent J 2006;17:144-8.
17. Byström A, Sundqvist G. Bacteriologic evaluation of the efficacy of mechanical root canal instrumentation in endodontic therapy. Scand J Dent Res 1981;89:321-8.
18. Siqueira JF, Alves FRF, Almeida BM, de Oliveira JCM, Rôças IN. Ability of chemomechanical preparation with either rotary instruments or self-adjusting file to disinfect oval-shaped root canals. J Endod 2010;36:1860-5.
19. Ledezma-Rasillo G, Flores-Reyes H, Gonzalez-Amaro AM, Garrocho-Rangel A, Ruiz-Rodriguez M del S,

Pozos-Guillen AJ. Identification of cultivable microorganisms from primary teeth with necrotic pulps. J Clin Pediatr Dent 2010;34:329-33.

20. Dalton BC, Orstavik D, Phillips C, Pettiette M, Trope M. Bacterial reduction with nickel-titanium rotary instrumentation. J Endod 1998;24:763-7.
21. Bystrom A, Sundqvist G. The antibacterial action of sodium hypochlorite and EDTA in 60 cases of endodontic therapy. Int Endod J 1985;18:35-40.
22. Subramanyam, Divya GJ. Microbiological evaluation of root canals after biomechanical preparation with manual and rotary file system-An *in vivo* study. Biomed J 2019;39:147-52.