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Comparison of disinfection effectiveness through manual instrumentation, rotary system and photodynamic therapy in primary molars: An in vitro study

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Highlights

- This study would help the pediatric dentists in clinical decision-making regarding the type of instrumentation for treating the root canals of primary teeth.
- This study showed a bacterial reduction in all studied groups compared to the control group.
- The application of these protocols reveals the novelty of the research, reducing the knowledge gap on the subject.

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Abstract

Aim: Due to the particular anatomy of primary teeth, improvements in the materials and protocols used for disinfecting and modelling the root canal system are needed to develop a more objective and efficient approach. The aim of this in vitro experimental study was to compare the disinfection of the root canal system of primary teeth with manual, rotary and photodynamic therapy systems. **Methods:** Forty-eight canals of primary molar teeth were infected with a suspension of Enterococcus faecalis and divided into four groups (n = 12): group G1, composed of canals prepared with manual H-type files from the Angelus® system via the crown-down technique; group G2, treated with the Sequence of Baby Files™ rotary system by the pecking motion technique; group G3, using the G2 treatment + photodynamic therapy, with methylene blue as photosensitizer at a concentration of 0.005%, irradiated with a 660 nm diode laser for 100 seconds, 100 mW power, 10 J energy and 357 J/cm² fluence; and group G4 (negative control), which received no treatment. Samples of the canal contents were collected with sterile paper cones before and after instrumentation, diluted and seeded on BHI agar plates. The bacterial colonies formed were counted, and the results were log transformed and subjected to analysis of variance (ANOVA) and Dunnett's test. **Results:** Showed a bacterial reduction in all studied groups compared to the control group. **Conclusions:** We concluded that the manual, rotary and photodynamic therapy systems all significantly reduced the E. faecalis count.

Keywords: Deciduous Teeth; Enterococcus faecalis; Root Canal Therapy

INTRODUCTION

The deciduous dentition exerts a significant influence on the growth and development of the child, affecting feeding, phonation, and functional aesthetics, and on the principles of an adequate occlusion, particularly affecting the maintenance of space for future permanent dentition.[1] This biological condition imposes a basic objective on pediatric dentistry: the preservation of primary teeth until the genetically determined period for their physiological exfoliation.[2] However, a large number of deciduous teeth are affected by caries or traumatic injuries. These two factors contribute substantially to the need for endodontic interventions in pediatric dentistry.[3]

Consequently, the root canal system of primary teeth is an aggravating factor that may compromise the success of pulp therapy. This is because in addition to the originally complex formation of the main, secondary and accessory canals, they also develop topographic changes as the rhizolysis process begins.[4] Thus, when pulp is compromised, particularly in deciduous molars, microorganisms and their toxins, residues of pulp decomposition, infiltrate this system of canals, greatly hindering their removal.[5] Classically, disinfection of the root canals of primary teeth is recommended using manual instruments, which are able to provide satisfactory cleaning, although it requires a relatively substantial amount of time in the clinic.[6]

On the other hand, continuous technical and scientific developments in endodontics have led to alternative mechanical instruments, such as rotary, ultrasonic and reciprocating instruments. Among the advantages of mechanized instrumentation is the reduction of operative time and a greater capacity for cleaning and shaping.[7-9] In addition, the implementation of photodynamic therapies (PDT) has been suggested, as it can potentially achieve an even greater reduction in the residual bacteria contained in root canal systems.[10]

The present study aimed to compare the "in vitro" disinfection capacity of the root canals of deciduous molars previously contaminated with *Enterococcus faecalis* and treated with the Angelus® manual system, the Sequence Baby Files™ rotary system, and PDT after instrumentation.

METHODS

This study was approved by the Research Ethics Committee of the São Leopoldo Mandic Dental Research Center, Campinas, São Paulo, Brazil, under protocol number 4,805,717 and was conducted according to the Declaration of Helsinki. The primary teeth used were obtained from the Biobank of the School of Dentistry of the University of São Paulo USP (BDH-FOUSP), with informed consent from a donation term.

Sample selection

A total of 24 deciduous molars were selected, of which 48 root canals were used for the experiment. This sampling is supported by a study conducted by Fonseca et al.[11] Prior to implementation of the research procedures, the teeth were cleaned using toothbrushes and washed in running water, removing all residual organic content.

The following inclusion criteria were considered: presence of coronary remnant, rhizolysis up to half of the root length, integrity of the furcation region, visible absence of areas of pathological root resorption, no previous endodontic manipulation and diameter of canals compatible with a K-file#10. Teeth that did not meet these requirements were excluded.

Sample preparation

Initially, endodontic access surgery was performed for all teeth in the sample. Access to the pulp chamber was performed with a file with

a high-speed spherical diamond drill (1012 FG, Angelus, Londrina, PR, Brazil) and finished with a file with an inactive, truncated diamond conical drill (3081 FG, Angelus, Londrina, PR, Brazil) to remove excess dentin and refine the opening. The crowns were flattened with a double-sided diamond disc (KG Sorensen, Barueri, SP, Brazil) to standardize the root length. Working length (WL) was determined visually by introducing a manual endodontic instrument (K-file#10, Angelus, Londrina, PR, Brazil) into the canal until its active tip could be visualized beyond the apical foramen. The instrument was removed, and the working length was determined with the aid of an endodontic ruler after subtracting 1 mm. To facilitate contamination of the root canal system, initial instrumentation was performed up to the manual K-file#15 (Angelus, Londrina, PR, Brazil) at the established working length.

The roots were waterproofed on their external surfaces with two layers of epoxy adhesive (Brascola Ltda., Taboão da Serra, SP, Brazil) and inserted vertically into individual wells of polystyrene plates for microtitering (Kasvi Imp. And Dist. Laboratórios Ltda., Paraná, Brazil). A thin layer of cyanoacrylate (Loctite Super Bonder, Henkel Ltda., São Paulo, Brazil) was also applied around the root and epoxy mass (Loctite Durepoxi, Henkel Ltda, São Paulo, Brazil) to seal the root foramen.

The specimens were numbered and autoclaved at 121 °C for 15 minutes for subsequent laboratory contamination with *E. faecalis* ATCC (ATTC 29212, Labcenter, São Paulo, Brazil). Two roots were randomly selected and submerged in a container with BHI broth (Difco, Michigan, USA) for 24 hours to serve as a control for sterilization effectiveness

The canals were contaminated with 1 mL of a suspension of a standard strain of *E. faecalis*, and a sterile K-file #10 (Angelus, Londrina, PR, Brazil) was used to uniformly disperse the bacterial suspension throughout the working length of the canal. The inoculated samples were incubated under microaerophilic conditions at 35-37 °C for 21 days, during which the culture media were replaced to maintain viability as a growth medium for the selected bacterial species. After the incubation period, the experiment began with irrigation of the root canals with 1 mL of 0.9% sterile saline solution (Arboretum, Minas Gerais, Brazil). Then, the material were initially collected from the interior of the canals to analyze the degree of contamination prior to endodontic instrumentation by inserting a sterile #15 absorbent paper cone (Tanari, Manacapuru, AM, Brazil) up to the working length for 1 minute. The paper cones were immediately dispensed in polypropylene flasks (Eppendorf, Hamburg, Germany) containing 900 μL of sterile saline solution. These samples were homogenized for 30 seconds in a vortex (AP56 Phoenix Luferco, São Paulo, Brazil) and then diluted (dilutions in 1:10), inoculated on BHI agar plates and incubated at 35-37 °C for 24 h. All manipulation of the specimens and instrumentation of the canals were performed in a laminar flow chamber by a single operator.

Group formation

The specimens were randomly divided by lottery (www.random.org) into four groups, each consisting of 12 root canals, according to the chosen instrumentation system: G1 - manual instrumentation; G2 - rotary instrumentation; G3 - rotary instrumentation with photodynamic therapy; G4 - negative control.

Laboratory phase

The specimens in group G1 (n = 12) were instrumented with an H-type stainless steel file (Angelus, Londrina, Paraná, Brazil) by the crown-down technique, starting with file size #40, then size #35 and so on until reaching the working length and then stopping at the apex with file size #30. The G2 (n = 12) specimens were treated

with the Sequence Baby Files continuous rotary system (MKLife, Porto Alegre, RS, Brazil), formed from NiTi M-wire alloys generated through thermomechanical processing. The system was used in its activated form with the E-Connect Pro endodontic motor (MKLife, Porto Alegre, RS, Brazil) according to the following protocol: sequential use of files 17/0.08, 20/0.04, and 25/0.04, then finalized with a 30/0.04 file depending on the initial apical diameter of the canal at 350 rpm with a torque of 1.5 N/cm², a 16:1 reduction handpiece, with the pecking motion and in-and-out techniques until passively reaching the working length.

In group G3 (n = 12), the specimens were instrumented in the same way as those of group G2 and then irrigated with 1 M H2O2 for 1 minute and inoculated with the photosensitizer methylene blue at a 0.005% concentration through a syringe with an irrigation needle (DMC, São Carlos, SP, Brazil) and the preirradiation time was 1 minute per channel. After this time, the canals were irradiated at the working distance with a red diode laser (Therapy EC, São Carlos, SP, Brazil) with a wavelength of 660 Nm, 10 J energy and fluence of 357 J/cm² for 100 seconds with a fiber length of 150 mm (DMC, São Carlos, SP, Brazil) in a helical pattern within the channel.

The canals of the G4 group constituted the negative control and did not receive any endodontic preparation, only irrigation with sterile saline solution.

For the instrumentation groups, the canals were individually irrigated with a standard total volume of 9 mL of sterile physiological saline solution (ASFER, São Paulo, Brazil) during preparation so that only the mechanical disinfection effect provided by the systems tested was considered. The samples were irrigated at each file change or at each removal-insertion for every third instrumentation, performed with a 10 mL disposable plastic syringe (Injex Ind. Cirurgicas Ltda., São Paulo, Brazil) coupled to a 26-G Safetips needle (Angelus, Londrina, PR, Brazil), introduced within the canal 2 mm below the WL for standardization of the procedure, followed by a patency K-file #10 (Angelus, Londrina, PR, Brazil).

After endodontic preparation, photodynamic therapy and irrigation, the final collection of bacterial samples was performed to verify the degree of decontamination achieved with insertion of #30 standard sterile paper cones (Tanari, Manacapuru, AM, Brazil) in groups G1, G2 and G3 until the working length, left in contact with the root walls for 1 minute. The collected samples were processed following the same protocol used for the initial collection. In G4, the final collection was performed following the same protocol as the initial phase.

Statistical analysis

After the incubation period, the bacterial colonies formed were counted, and the results were analyzed in statistical software R for Windows version 4.1.3. The results, obtained in CFU/mL, were $\log_{10}$ transformed and subjected to analysis of variance (ANOVA) and Dunnett's test.

RESULTS

The results of the selection and calculation of the counts are shown in the following tables (Table 1,2 and 3). The numbers were rounded to whole numbers because they were count data and the magnitudes of the standard deviations were in the tens of thousands.

Similar to the counts before the interventions, we performed bacterial counts after the interventions in the four experimental groups, as shown in Table 2.

Table 1. CFU/mL counts before the interventions

	G1	G2	G3	G4
	443.333	80.333	20.333	40.000
	386.667	333.333	14.333	5.667
	190.000	42.000	56.000	73.333
	146.667	68.667	90.000	27.000
	576.667	96.667	186.667	280.000
	296.667	173.333	166.667	16.333
	450.000	1.516.667	15.667	20.000
	290.000	520.000	44.333	16.667
	263.333	66.000	7.000	11.667
	58.000	17.000	6.967	12.667
	24.667	576.667	23.333	20.333
	27.000	690.000	20.333	14.000
Mean	262.750	348.389	54.303	44.806
Standard deviation	180.004	435.984	62.054	76.217
Minimum	24.667	17.000	6.967	5.667
1st quartile	124.500	68.000	15.333	13.667
Median	276.667	135.000	21.833	18.333
3rd quartile	400.833	534.167	64.500	30.250
Maximum	576.667	1.516.667	186.667	280.000
Interval	552.000	1.499.667	179.700	274.333

Table 2. CFU/mL counts after the interventions

	G1	G2	G3	G4
	4.500	4.167	740	6.233
	20.000	1.933	650	14.333
	10.667	2.567	503	14.667
	39.000	7.367	1.367	6.567
	12.667	3.633	653	16.667
	2.200	2.200	560	5.133
	7.333	24.000	1.200	5.700
	7.433	3.633	507	3.433
	15.333	2.433	220	2.733
	7.600	1.900	140	4.833
	6.300	16.667	41	3.733
	1.100	5.900	25	2.700
Mean	11.178	6.367	551	7.228
Standard deviation	10.286	6.908	422	5.010
Minimum	1.100	1.900	25	2.700
1st quartile	5.850	2.375	200	3.658
Median	7.517	3.633	533	5.417
3rd quartile	13.333	6.267	675	8.508
Maximum	39.000	24.000	1.367	16.667
Interval	37.900	22.100	1.341	13.967

Table 3. Differences between the log reductions of the treatment and control group

		%95 confidence interval	
Groups	Difference	Lower limit	Upper limit
G1-G4	0.7664	0.2881	1.2448
G2-G4	0.9832	0.5049	1.4616
G3-G4	1.3595	0.8812	1.8400

Data transformation

Except for two extreme points, group G4 also showed the smallest overall distribution in the log reduction in bacterial count, while the three groups of interventions show a greater variance in the results (Figure 1).

Difference based on gross means

Dunnett's test shows the difference of the log reduction among all the groups (Figure 2).

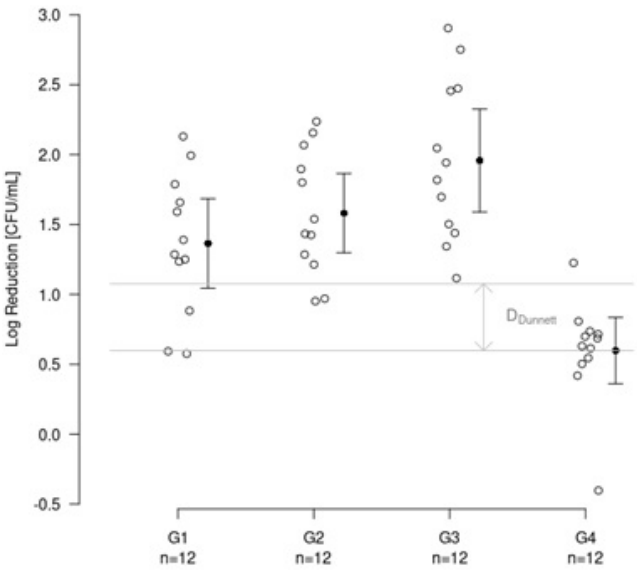

Figure 1. Log reduction in the number of colony-forming units for all treatment groups

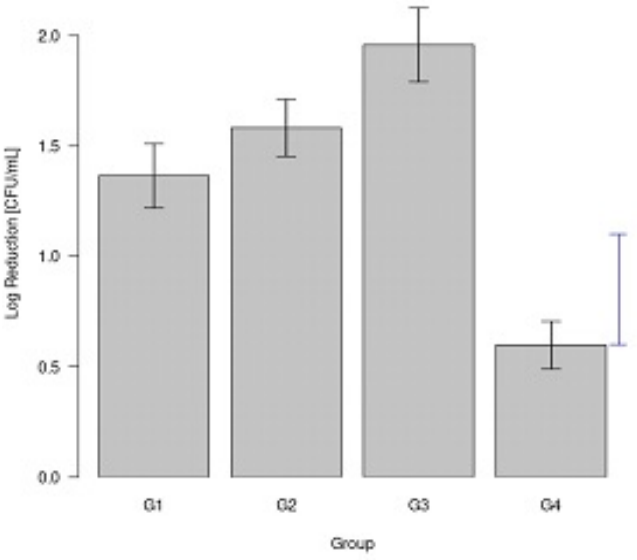

Figure 2. Comparisons between the groups. G1 (M = 1.36; sd = 0.50; p value = $9.69 \cdot 10^{-04}$); G2 (M = 1.58; sd = 0.45; p value = $3.07 \cdot 10^{-05}$); and G3 (M = 1.96; sd = 0.58; p value = $9.66 \cdot 10^{-08}$)

DISCUSSION

The dental literature has exhaustively explored treatment techniques and philosophies regarding pulp therapy in primary teeth, proposing and reviewing references on this subject.[12] However, there is no consensus among the different treatment modalities, either in the instrumentation techniques for the root canal system or in the drugs used in the disinfection and obturation of the root canals.[13-15] These considerations motivated the execution of the present study in order to support the pediatric dentist in his or her clinical decision-making regarding the type of instrumentation for the root canals of endodontically compromised primary teeth.

The results of the experiment in our study, obtained using the plate method, revealed a logarithmic reduction in bacterial load for each of the four groups with twelve replicates per leg of the study. The experiment can thus be described as single-factor and completely randomized, comparing the disinfection systems chosen in the methodology—manual, rotary, and photodynamic therapy—to nonintervention. In addition, because one of the groups is a control, the experiment conducted planned comparisons, that is, it compared each intervention with the control. Similar to this study, other experimental studies have allowed the creation of controlled situations for the observation of isolated factors, quantifying mechanical disinfection in the root canals.[3,11,16,17]

Considering the established methodology, our results showed a bacterial reduction in all studied groups when compared to the control group (Fig 2). It is plausible to credit the postinstrumentation bacterial reduction observed in our results exclusively to the mechanical effect associated with irrigation of the canals with saline solution, given that no antimicrobial solution was used; these results are consistent with those of Fonseca et al.[11] The reduction in *E. faecalis* was more significant with the use of PDT than with the techniques used in the other groups (Table 3), similar to the findings in the study by Mohan et al.[18] This condition can be explained by the physical, chemical and biological action provided by the combined reaction of photosensitizing agents, light and oxygen, resulting in cell death by oxidation.[19,20] Despite all the benefits of PDT, there is still no consensus or protocol for its use

in primary teeth in clinical practice, and it is considered to play an adjuvant role in the disinfection of the canals.[21,16]

In addition to the disinfection of the root canals obtained by the reduction in bacterial density, it is important to discuss the modeling of instrumented canals. Although it has not been the object of research, modeling is fundamental in the disinfection process, reducing interference and contributing significantly to root canal filling. Laboratory research and clinical trials have shown that the mechanical and chemical preparation of the root system of primary teeth impacts the quality of root canal filling.[22,23] Thus, scientific evidence indicates that mechanized instrumentation, compared to manual instrumentation, has better performance, although manual instrumentation allows greater tactile control and can help prevent iatrogenesis.[23-25] Additionally, Manchanda et al.[2] and Babu & Kavyashree[26] reported differences between instrumentations and found that rotary techniques require significantly shorter instrumentation and obturation times. In addition, Chauhan et al.[27] demonstrated that mechanized instrumentation requires greater skill and imposes a significant cost on the clinician.

Considering the thin topography of the root canals of the deciduous teeth, the apical preparations of the samples of this study were intentionally performed with a #30 tip and 2% and 4% taper in both the manual instrumentation and the rotary technique. Fonseca et al.[11], in a similar study, exclusively used rotary and reciprocating files with 6%, 7% and 8% tapers, observing similar levels of bacterial reduction, implying that the difference in the conicity of the files does not influence the disinfection capacity.

We emphasize that the instrumentation system H and the rotatory "Sequence Baby Files" were recently introduced exclusively for use in deciduous teeth, and there wasn't localized papers to make a comparative analysis. Therefore, this can be considered a study limitation. Nonetheless, this research is unprecedented and have the basement that can be used by future works to conduce approaches for this instrumentation system. Assuming clinical applicability of mechanized instruments in both the private and public sectors, the cost–benefit ratio must be considered. While manual instruments are more economically accessible, mechanized instruments provide a more effective bacterial reduction and modeling that facilitates subsequent obturation. In addition, the clinical duration of the intervention is reduced, a relevant factor in the practice of pediatric dentistry. We also emphasize that postinstrumentation PDT should be encouraged due to its synergistic ability in disinfecting the root system of primary teeth.

CONCLUSIONS

According to the results of the study, we can conclude that both the manual and rotary instrumentation systems showed a bacterial reduction in relation to the nonintervention group, potentiated by the implementation of PDT after instrumentation.

REFERENCES

1. Pawar B, Pawar AM, Bhardwaj A, Wahjuningrum DA, Rahardjo AK, Luke AM, et al. Effect of Adaptive, Rotary, and Manual Root Canal Instrumentation in Primary Molars: A Triple-Armed, Randomized Controlled Clinical Trial Biology 2021;42:1-11

2. Manchanda S, Sardana D, Yiu CKY. A systematic review and meta-analysis of randomized clinical trials comparing rotary canal instrumentation techniques with manual instrumentation techniques in primary teeth. Int Endod J 2020;53:333–353

3. Shah HS, Patil VM, Kamath AP, Mathur AA. Comparative Evaluation of Instrumentation Time, Obturation Time, and Radiographic Quality

of Obturation Using Two Rotary Systems and Manual Technique for Primary Molar Pulpectomies: In vivo Study. Contemp Clin Dent 2021;12: 55-62

4. Boonchoo K, Leelataweewud P, Yanpiset K, Jirarattanasopha V. Simplify pulpectomy in primary molars with a single-file reciprocating system: a randomized controlled clinical trial. Clin Oral Investig 2020;24:2683-2689

5. Barasuol JC, Massignan C, Bortoluzzi EA, Cardoso M, Bolan M. Influence of hand and rotary files for endodontic treatment of primary teeth on immediate outcomes: Secondary analysis of a randomized controlled trial. Int J Paediatr Dent 2021;31:143-151

6. Natchiyar N, Asokan S, Priya PRG, Kumar TDY. Comparison of Clinical and Radiographic Success of Rotary with Manual Instrumentation Techniques in Primary Teeth: A Systematic Review. Int J Clin Pediatr Dent 2021; 14:8-13

7. Rathi N, Jain S, Thosar N, Baliga S, Ahmed F, Metha J. Comparative Evaluation of Cleaning Efficiency and Apical Extrusion of Debris Using Two Pediatric Rotary Endodontic Files: An In Vitro Study. Int J Clin Pediatr Dent 2021;14:196-200

8. Seema T, Ahammed H, Parul S, Cheranjeevi J. Comparative Evaluation of Dentin Removal and Taper of Root Canal Preparation of Hand K File, ProTaper Rotary File, and Kedo S Rotary File in Primary Molars Using Cone-beam Computed Tomography. Int J Clin Pediatr Dent 2020;13:332-336

9. Tyagi R, Khatri A, Kalra N, Sabherwal P. Comparative Evaluation of Hand K-flex Files, Pediatric Rotary Files, and Reciprocating Files on Instrumentation Time, Postoperative Pain, and Child's Behavior in 4–8-year old Children. Int J Clin Pediatr Dent 2021;14:201-206

10. Maciel Martins CR, de Andrade MV, Carvalho AP, Afonso Pereira RM, Bresolin CR, Mello-Moura ACV, et al. Photodynamic therapy associated final irrigation in root canals of the primary teeth. Photodiagnosis Photodyn Ther 2021;33:102182

11. Fonseca LA, Cangussu RA, Oliveira AS, Pinheiro SL, Shitsuka C, Duarte AD. Comparison of endodontic disinfection of primary teeth root canals using rotary and reciprocating system: An in vitro study. Res Soc Dev 2020;9:e457985882

12. Panchal V, Jeevanandan G, Erulappan SM. Comparison between the Effectiveness of Rotary and Manual Instrumentation in Primary Teeth: A Systematic Review. Int J Clin Pediatr Dent 2019;12:340–346

13. Moraes R, Perez R, Silva A, Machado AS, Lopes RT, Pintor A, et al. Micro-CT evaluation of root canal preparation with rotary instrumentation on prototyped primary incisors. Braz Oral Res 2021;35:e132

14. Öter B, Topçuog Lu N, Tank MK, Çehreli SB. Evaluation of Antibacterial Efficiency of Different Root Canal Disinfection Techniques in Primary Teeth. Photomed Laser Surg 2018;36:179–184

15. Walia V, Goswami M, Mishra S, Walia N, Sahay D. Comparative Evaluation of the Efficacy of Chlorhexidine, Sodium Hypochlorite, the Diode Laser and Saline in Reducing the Microbial Count in Primary Teeth Root Canals - An In Vivo Study. J Lasers Med Sci 2019; 10: 268–274

16. Pinheiro SL, da Silva JN, Gonçalves RO, Villalpando KT. Manual and Rotary Instrumentation Ability to Reduce Enterococcus faecalis Associated with Photodynamic Therapy in Deciduous Molars. Braz Dent J 2014;25:502-507

17. Tewari RK, Ali S, Mishra SK, Kumar A, Andrabi SMUN, Zoya A, et al. Mechanical reduction of the intracanal Enterococcus faecalis population by Hyflex CM, K3XF, ProTaper Next, and two manual instrument systems: an in vitro comparative study. J Investig Clin Dent 2016;7:168–173

18. Mohan D, Maruthingal S, Indira R, Divakar DD, Al Kheraif AA, Ramakrishnaiah R et al. Photoactivated disinfection (PAD) of dental root canal system - An ex-vivo study. Saudi journal of biological sciences 2016;23:122–127

19. Plotino G, Grande NM, Mercade M. Photodynamic therapy in endodontics. Int End J 2019;52:760–774

20. Garcez AS, Hamblin MR. Methylene blue and Hydrogen peroxide for photodynamic inactivation in root canal - a new protocol for use in Endodontics. Eur Endod J 2017;2:29-35

21. Okamoto CB, Bussadori SK, Prates RA, da Mota ACC, Tempestini Horliana ACR, Fernandes KPS, et al. Photodynamic therapy for endodontic treatment of primary teeth: A randomized

controlled clinical trial. Photodiagnosis Photodyn Ther 2020;30:101732

22. Musale PK, Mujawar SA. Evaluation of the efficacy of rotary vs. hand files in root canal preparation of primary teeth in vitro using CBCT. European archives of paediatric dentistry: official journal of the Eur Acad Paediatr Dent 2014;15:113–120

23. Jafarzadeh M, Saatchi M, Jafarnejadi P, Gooran M. Digital Radiographic Evaluation of the Quality of Different Root Canal Obturation Techniques in Deciduous Mandibular Molars after Preparation with Rotary Technique. J Dent 2019;20:152–158

24. Subramaniam P, Raveendran N, Madhusudhan KS. Comparative evaluation of two pediatric rotary file systems in primary teeth: A scanning electron microscope study. Contemp Pediatr Dent 2022;3:8-14

25. Dalzell O, Mohd Ariffin S, Patrick CJ. Hardiman R, Manton DJ, Parashos P *et al.* Root canal instrumentation efficacy of non-fused and fused primary molar roots: a micro-computed tomography study. Eur Arch Paediatr Dent 2021;22:911–927

26. Babu KLG, Kavyashree GH. Evaluation of the clinical efficiency of rotary and manual files for root canal instrumentation in primary teeth pulpectomies: A comparative randomized clinical trial. Contemp Pediatr Dent 2021;2:21-34

27. Chauhan A, Saini S, Dua P, Mangla R. Rotary Endodontics in Pediatric Dentistry: Embracing the New Alternative. Int J Clin Pediatr Dent 2019;12:460-463

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