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pH changes after manual or ultrasonic instrumentation and smear layer removal with EDTA or ultrasonic

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Abstract – The purpose of the present study was to evaluate the influence of instrumentation techniques associated with the smear layer removal, in the pH changes of the root surface. Thirty mandibular humans premolars were divided into three groups: Group I – instrumentation by Ohio technique and final cleaning with EDTA (3 minutes); Group II – instrumentation by Ohio technique and final cleaning with ultrasonic (1 minute); Group III – instrumentation by the ultrasonic technique and final cleaning with ultrasonic (1 minute). The pH was measured in the cavities prepared in the cervical, middle and apical thirds of the lateral wall of each root. The teeth were evaluated at the initiation of the experiment, and 3, 7, 14, 21, 30 days after of the intracanal dressing of the calcium hydroxide with camphorated *p*-monochlorophenol (Calen/PMCC). All the groups presented increasing pH values; group III presented the highest average pH, followed by groups II and I; the values for the apical third were lower than those of the middle and cervical thirds (ANOVA and Tukey test). The results showed that the biomechanical preparation by the ultrasonic technique and smear layer removal with ultrasonic showing the highest diffusion of the calcium and hydroxyl ions from the intracanal dressing ($P < 0.05$).

The endodontic treatment of pulpless teeth with chronic periapical lesion requires intense action to combat infection in the root canal. The biomechanical preparation of root canal reduces the number of bacteria; however micro-organisms may persist and increase in number between sessions, particularly anaerobic Gram-negative, if no antibacterial medication is applied (1–3).

Considering the complexity of dental root canal anatomy, which may cause certain areas not to be accessible to mechanical instrumentation, the use of an intracanal dressing between sessions plays a key role in the attempt to eliminate hidden bacteria from the root canal system (4).

Calcium hydroxide (CH) has been proposed as an intracanal dressing for pulpless necrotic teeth (5, 6) due to its antibacterial properties, including pH increase (5), lipopolysaccharide (LPS) detoxification (6), CO_2 absorption (7) as well as its anti-inflammatory action which may contribute to tissue repair (8, 9). When CH pastes are used as a dressing, the increase in pH in the surrounding tissues is expected. The augment in the external pH levels is driven by the release of hydroxyl ions from the CH pastes. The association of CH and camphorated *p*-monochlorophenol has demonstrated a controlled and progressive release of hydroxyl ions, reduced solubility and long-term contact with the dentine walls (10–13). Because of the bactericidal action against both anaerobic and aerobic populations, this association has been indicated as a root canal dressing for teeth with chronic periapical lesions (5).

In order to reach a high pH level at the outer surface of the root dentine, hydroxyl ions must be able to diffuse from the material throughout the root canal system (14). Therefore, the final cleaning of the root canal walls (smear layer removal) after instrumentation is considered essential (15) in order to allow the diffusion of ions released by the intracanal dressing into the dentinal tubules (16). The use of ultrasonic and EDTA improve smear layer removal (17), however it is still uncertain whether the use of either EDTA or ultrasonic for final cleaning may improve pH levels in teeth dressed with CH.

The objective of the present study was to evaluate pH changes in the radicular dentine after root canal dressing with CH in root canals prepared either manually or ultrasonically, associated with smear layer removal by either EDTA or ultrasonic.

Material and methods

This study was approved by the Ethics Committee for Research at São Paulo State University, Dental School of Araraquara-UNESP, CEP-FOAr, record number 70/03. Thirty mandibular human premolars recently extracted were used. All teeth were cleaned from debris and stored in 10% formaldehyde solution for 24 h and afterwards in saline solution until the moment of use.

Coronal access was performed using a #4 round bur in a high-speed hand-piece (KG Sorensen, São Paulo, SP, Brazil), under water cooling. The work length was determined at 1 mm from the apex, by inserting a #15 K-file (Dentsply Maillefer Indústria e Comércio Ltda, Petrópolis, RJ, Brazil) until the tip was just visible at the apical foramen.

Teeth were randomly divided into three groups ($n = 10$) and instrumented using the following techniques: groups I and II – biomechanical preparation was performed using Ohio technique (18). The preparation started with an apical enlargement using a #15 K-file (Dentsply-Maillefer, Ballaigues, Switzerland) up to #25. Gates Glidden #2 (ISO 60) was used to flare the coronal two thirds of the canal giving free access to #30 and #35 K-files to the working length. Then a #3 Gates Glidden drill (ISO 80) was used to enlarge the coronal segment to ensure a free access to a #40 K-file to the working length. This file was selected as the final instrument. Deionized water was used as irrigation solution throughout the preparation. The final cleaning of group I was performed by filling the root canal with EDTA solution (Odacan Dentsply Indústria e Comércio Ltda, Petrópolis, RJ, Brazil) and stirring the solution for 3 min using the final instrument. In group II, the final cleaning was performed using an ultrasonic device (Jet Sonic Four Plus; Gnatus Equipamentos Médico-Odontológicos Ltda, Ribeirão Preto, SP, Brazil) set at intensity level 3. An ultrasonic #15 K-file (Mani; Matsutani Seisakusho Co. Ltd, Utsunomiya, Tochigi, Japan) was let to oscillate in the root canal for 1 min with constant irrigation using deionized water.

In group III, the root canals were ultrasonically prepared using an ultrasonic #15 K-file for 3 min connected to an ultrasonic device (Jet Sonic Four Plus; Gnatus Equipamentos Médico-Odontológicos Ltda) set at an intensity level of 3. Constant irrigation with deionized water was carried out during the preparation. A final #25 K-file, modified to couple to the ultrasonic hand-piece, was used for 1 min. The final cleaning was carried out in the same conditions as described for group II. The root canals were thoughtfully dried with paper points (Dentsply-Maillefer).

Three cavities were drilled in the proximal outer surface of the root, positioned at the cervical, middle and apical thirds, using a round bur diamond #2 (KG Sorensen). The cavities were 0.75 mm in depth and 1.5 mm in diameter each. The root canals were filled with a CH paste (2.5 g of CH, 1 g of zinc oxide p.a., 0.05 g of colophony, 2 ml of polyethylene glycol 400, 0.04 g of paramonochlorophenol – Calen PMCC; S.S. White Artigos Dentários Ltda, Rio de Janeiro, RJ, Brazil), using a special syringe gauged with a G27 needle. The needle's tip was adjusted to the working length by gauging a rubber stop to the needle. After completely filling the root canal with CH paste, the coronal accesses were sealed with Z-250 composite resin (Filtek Z250; 3M-ESPE, St Paul, MN, USA).

A pHmeter (Orion model 240 A; Microeletrodes, Inc., Bedford, NH, USA) was used to record the pH levels in the proximal cavities (cervical, middle, and apical cavities) at 0 (baseline), 3, 7, 14, 21 and 30 days after the intracanal dressing. Each tooth was removed from its individual vial and rinsed with distilled water, then blotted dry. Two microlitres of distilled water was placed in the cavity. After 10 min, the pH was measured using a calibrated pHmeter. Teeth were kept during the experiment in individual bottles containing saline solution, and after measurements, each tooth was returned to its vial. The average pH of each cavity was determined for each test group at the different time periods.

ANOVA and Tukey test were used to detect differences in the pH levels among groups and cavities at each time periods. The level of significance was set at $P < 0.05$.

Results

ANOVA showed that pH levels among the groups were significantly different for all the time periods evaluated. Tukey test showed that the average pH in group I was significant lower than in group II ($P < 0.05$), which was significantly lower than in group III ($P < 0.05$) (Table 1).

Group I demonstrated a gradual increase in pH, peaking at 14 days, remaining stable until the end of the experiment. Group II showed slightly higher pH indexes than group I at all the periods, maintaining a gradual increase up to the 30th day. Group III showed the highest pH indices among the groups, reaching a peak at 14 days and remaining stable up to the 30th day (Fig. 1). The pH values for the apical third were significantly lower ($P < 0.05$) than for the middle and cervical thirds, for all the evaluated groups (Fig. 2).

Discussion

The aim of endodontic treatment of pulpless teeth with chronic periapical lesion is not only to eliminate micro-organisms, but also to inactivate the toxic effects of LPS (2, 19). The biomechanical preparation of the root canal only partially reduces the endodontic microbiota. However, it does not eliminate bacteria from the entire system, and does not act on LPS. Thus the use of an intracanal dressing is considered as necessary by many (2). Because micro-organisms can still be detected in the root canal after 72 and 96 h (20), an intracanal dressing is desired.

Table 1. Average pH accumulation at different groups during the evaluation time

	Group I	Group II	Group III
Average	8424[a]	8726[b]	9616[c]
SD	0.1499	0.5072	0.6569

Different letters show significant difference.

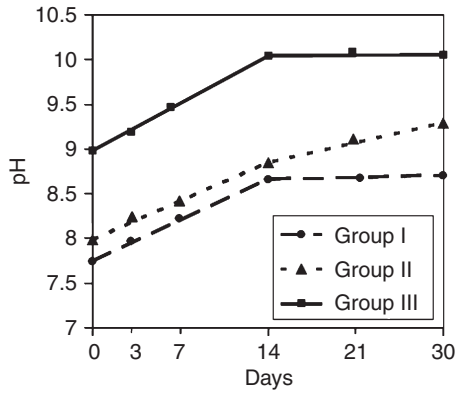

Fig. 1. Graph 1 – Average increase in pH levels among each interval time.

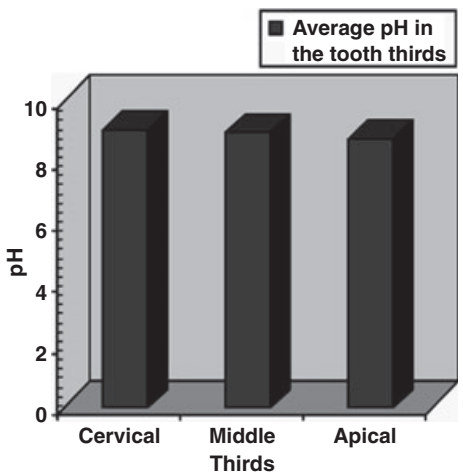

Fig. 2. Graph 2 – Average pH at each tooth third.

Considering that micro-organisms may hide at inaccessible areas, an antimicrobial intracanal dressing capable of diffusing throughout the root canal system play a key role in the attempt to reduce and inactive micro-organisms that escaped from the biomechanical preparation.

Two important enzymatic properties may be attributed to CH as an intracanal dressing: inhibition of bacterial enzymes, which takes place at the cytoplasmatic membrane (antimicrobial effect); and tissue enzymatic activation, by activation of alkaline phosphatase (mineralizing effect). Because those enzymatic reactions are consequences of a high pH environment (average 12.6), the release of hydroxyl ions is considered as an important characteristic of CH pastes. The more hydroxyl ions are released, the higher the pH. CH also inactivates the LPS, by hydrolysis of its toxic part (lipid A), changing its molecular arrangement into non-toxic molecules (6, 21). As the root canal system has plenty of areas inaccessible to the biomechanical preparation, the dissociation of hydroxyl ions from CH pastes may provide a desired clinical response by alkalizing the dentine (8), turning those zones into an inappropriate area for bacterial development and proliferation.

Several methods can be used to study the permeability and diffusion of calcium and hydroxyl ions throughout the dentine. The most common models are modulation of pH indicators, use of dyes associated with the pastes and use of electrodes (8, 11, 15, 22, 23).

In the present study, Calen PMCC paste was used as the intracanal dressing, and the changes in pH values along 30 days were recorded using the pHmeter electrode. Because the vehicle in the Calen PMCC paste is viscous (polyethylene glycol 400), the speed of dissociation of the calcium and hydroxyl ions is expected to be slow (24). This speed relies on the solubilization and resorption capacity of the pastes in the apical tissues, and therefore determines the antimicrobial and mineralizing action (9).

In the present study, standard cavities of 0.75 mm in depth and 1.5 mm in diameter were prepared in order to allow a correct positioning of the pHmeter needle during recordings at different time periods along the course of the experiment (22, 23).

In order to detect the effect of using EDTA or ultrasonic on the dentinal pH CH-induced, deionized water was used for root canal preparation to generate as much debris as possible. It was observed that using ultrasonic for final cleaning a higher dentinal pH level could be reached. Because ultrasonic can remove smear layer efficiently than EDTA, a final ultrasonic cleaning is indicated when use of CH paste is recommended. In group III as the root canals were both prepared and cleaned using ultrasound, less debris than manual preparation were expected. Therefore, as the smear layer was present in small amounts or not at all present in this group, the highest dentinal pH after root canal dressing with CH was observed. According to our data, ultrasonic preparation may improve the diffusion of CH paste throughout the dentine mass.

In the present study, lower levels of pH in the apical cavity were observed for all experimental groups, which is in line with a previous study (25). Considering that a complete root canal cleaning at the apical third of atresic root canals is usually difficult, the diffusion of the intracanal dressing at this level might be negatively affected.

On the basis of the results observed in the present study, it may be concluded that the three groups evaluated showed high pH values, with group III presenting the highest average pH, followed by groups II and I. The values for the apical third were lower than for the middle and cervical thirds. The biomechanical preparation and smear layer removal with ultrasound can increase the dentinal pH of root canals dressed with CH.

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