The Influence of Cleansers on the Permeability Index of Primary Tooth Root Dentin

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The aim of this study was to evaluate the action of cleansers on the permeability index (PI) in the root canal of primary dentin, by dye penetration into the length of the root canal and deep dentin. The data were submitted to ANOVA, F, and Student t tests ($p<0.05$). Lengthwise, chlorhexidine 1% and 2% presented significantly higher mean values than Dakin´s liquid. Considering depth, in the cervical third, chlorhexidine 1% and 2% mean values were higher than Dakin´s liquid+urea peroxide and Dakin´s liquid mean values; distilled water mean values were higher than Dakin´s liquid. In the middle and apical thirds, chlorhexidine 1% and 2% mean values were significantly the highest.

Key words: *Root Cleanser; Primary Tooth; Irrigation.*
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INTRODUCTION

Root canal treatment can be summarized as a series of procedures for cleaning, shaping and filling the root canal system. One of the most important procedures during treatment is chemo-mechanical preparation of the root canal, based on the correct use of instruments and irrigation solutions.[1] In this context, the use of cleansers in the irrigation process is very important for the success of the endodontic treatment, acting as an auxiliary in the removal of the smear layer produced during instrumentation. Irrigation is currently the best method for removing tissue remnants and dentin debris. In addition, the cleansers provide gross debridement, lubrication, microorganism destruction and tissue dissolution.[2]

The cleansing of the root canal may make dentin tubules more permeable, thus the irrigating fluids may more effectively flush out debris and remaining microorganisms permiting greater penetration of medicaments into dentinal tubules.[3] Alterations in the permeability of primary human radicular dentin is very important to both physiopathology and endodontic therapy.[4] The root canal system must be cleaned, decontaminated, shaped, and enlarged, since the filling has to be made with non-setting pastes. These pastes have to penetrate the dentinal tubules in order to minimize bacterial contamination and not allow re-infection of the root canal system.

The roots dentinal permeability may increase, because of the chemical-mechanical preparation of the root canals. The technique of dye penetration into the dentin is frequently used for evaluating the increase of dentinal permeability.[3] In addition, human root dentin permeability can be measured as the hydraulic conductance before and after treatment with K files. Before and after subsequent treatment of the endodontic smear layer with sodium hypochlorite (NaOCl), 50% citric acid or 3% monopotassium-monohydrogen oxalate.[4] Fogel *et al.*[4] observed that filing reduced dentin permeability by 25% and 49% respectively, depending upon whether the outer or inner root dentin was filed. These smear layers were unaffected by 5% NaOCl, but increased many times after treatment with 50% citric acid for two minutes.

The combination of NaOCl with acidic or base substances, and even NaOCl+H2O2 (Hydrogen Peroxide) have been widely used as an irrigant in pulp therapy.[5] At present, NaOCl solutions in concentrations ranging from 1% to 5.25% and commercial solutions of 5.25% (Clorox®) are often used in endodontic therapy.[5] This concentration is effective as a tissue solvent with antimicrobial effects. However, it would appear that used as a flush in instrumented root canals, does not remove the smear layer. The main organic dentin matrix is a compound of molecules of type I collagen and proteoglycans. Those molecules are highly hydrophilic and polyanionic due to carboxyl and sulfate groups present in glycosaminoglycan chains. This allows them to regulate water content and intratubular permeability.[5] Nakurama *et al.*[6] observed that NaOCl has a proteolytic and dissolving collagen. As a consequence, the NaOCl induces alterations in dentin collagen and glycosaminoglycans and shows the protective role of hydroxyapatite on the organic matrix stability.[5]

The use of chlorhexidine digluconate as an endodontic irrigant has spread, due to its substantial antibacterial effect and cleaning ability. Ari and Erdemir[7] observed a significant decrease in the calcium and phosphorus levels after treatment of 15 min irrigation with 0.2% chlorhexidine gluconate, 3% H2O2, 17% EDTA, 5.25% and 2.5% NaOCl and distilled water. They concluded that endodontic irrigation solutions have an effect on the mineral contents of root dentin.

It was observed that the reactivity of dentin tissue varies according to the tooth type (primary or permanent) and to the studied area.[8]

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The root surface of primary teeth has not yet been studied either for reactivity to cleansers or even to the acids during endodontic procedures.

Thus, the choice of a cleanser in the pulpal therapy of primary teeth should take into account the differences among the dentinal substrata, particularly with regard to the following factors: They must not irritate the periapical tissues in order to avoid harming the germ of the permanent successor tooth; they should facilitate instrumentation; they may possibly lead to variations in the root dentin permeability of primary teeth; and also their bactericidal and potential irritating action

The purpose of this study was to evaluate the effect of cleanser action on the permeability index in the root canal in the primary dentin. The null hypothesis evaluated was that there were no differences among irrigant solutions (0.5% NaOCl, 0.5% NaOCl associated with urea peroxide and distilled water) and gels (1% and 2% chlorhexidine digluconate) at this depth in dentin and length in the root canal in primary teeth.

MATERIALS AND METHODS

Thirty infected human maxillary and mandibular posterior primary teeth were extracted for clinical reasons and were stored in 10% formaldehyde for 24 hours. Next, they were washed and stored in saline solution, under refrigeration until further use.

Only teeth with at least two-thirds of intact root were selected. The roots were sectioned transversely at the cement-enamel junction (approximately 0.5mm above the root bifurcation) and the crowns were discarded. The teeth were randomly divided into 5 groups (n= 6) depending upon the type of irrigant used. The groups were treated with irrigation solutions as follows:

a. Group 1 (D): 1 ml of 0.5% sodium hypochlorite (Dakin's liquid), between each instrumentation (a total of 5 ml of cleanser);
b. Group 2 (DE): 1 ml of 0.5% sodium hypochlorite (Dakin's liq uid) associated with urea peroxide (Endo-PTC - 10% Urea Peroxide, 15% Tween 80 and 75% Carbowax – Polidental Industry and Commercial, São Paulo, Brazil, Batch 6220) between each instrumentation (a total of 5 ml of cleanser);
c. Group 3 (C1): filling root canal with 1% chlorhexidine digluconate gel followed by 1 ml of distillated water between each instrumentation (a total of 5 ml of cleanser);
d. Group 4 (C2): filling root canal with 2% chlorhexidine gluconate gel followed by 1 ml of distillated water between each instrumentation (a total of 5 ml of cleanser);
e. Group 5 (DW): 1 ml of distillated water used between each instrumentation (a total of 5 ml of cleanser).

The same manufacturer (Proderma, Laboratory of Manipulation, Piracicaba, Brazil) prepared Dakin's liquid and chlorhexidine gels. After determining the working length (1.0 mm shorter than root canal length) by inserting the thinnest file (#15, Dentsply/Maillefer, Ballaigues, Switzerland) the root canals were manually instrumented with a K-file (#15-#35–Dentsply/Maillefer, Ballaigues, Switzerland). The solutions and gels were inserted in the root canals using a 1 ml insulin syringe with suitable needles (12.7 x 0.33mm) with round edges (Becton Dickinson and Co., New Jersey, USA)

The root canals were dried with absorbent paper tips (Tanari FDA, Manaus, Brazil, Batch 005001P), and externally made impermeable with nail varnish and apically with wax. Next, they were immersed in 2% methylene blue solution for four hours (Figure 1). After that, they were washed to remove dye excess, and were longitudinally sectioned to evaluate the permeability index (PI). This was verified by the extent of dye penetration (cervical through apical direction) and depth into the root dentin. Pictures were taken of the halves, and a diagram was obtained. The dyed areas were divided into thirds (cervical, middle and apical). These parts were vertically and horizontally marked in three parts to allow dye penetration to be observed: length (canal access/apical foramen) and depth (canal light/root cement) consisting of 9 parts/third. The PI was expressed in percentages: dye penetration parts/9 in agreement with Marshall *et al.*[3] (Figure 2).

Figure 1: Illustration of the root canal of primary dentin by dye penetration into the length of the root canal and deep dentin.

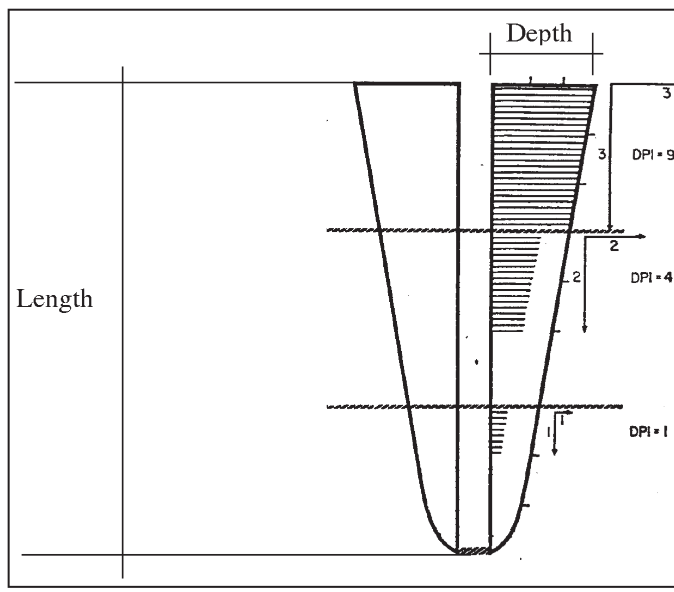

Figure 2: Illustration of the diagram of the root canals for permeability analysis.

The permeability index was calculated by the following procedure:

$$Permeability\ Index^{*} = \frac{Parts\ penetrated\ by\ dye}{9} \times 100$$

The data were submitted to the ANOVA, F, and t Student tests. The tests were performed at the 95% level of confidence ($p < 0.05$).

RESULTS

Regarding the full extent of the canal (length), 1% and 2% chlorhexidine digluconate gels produced the higher PI compared to Dakin's solution. The Dakin's solution and urea peroxide association and distilled water showed no differences between them and intermediate PI (Figure 3). For PI in the dentin depth, 1% and 2% chlorhexidine digluconate produced the highest PI in all thirds of root canals. Dakin's solution showed the lowest percentage of dye penetration (Figure 4).

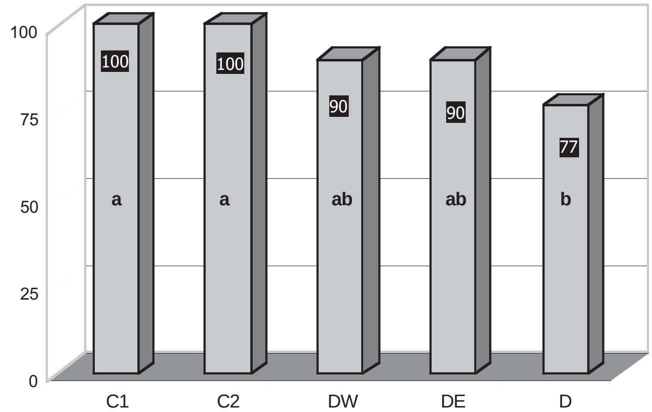

Figure 3: The permeability index of the dye penetration in length (canal entrance/apical foramen).

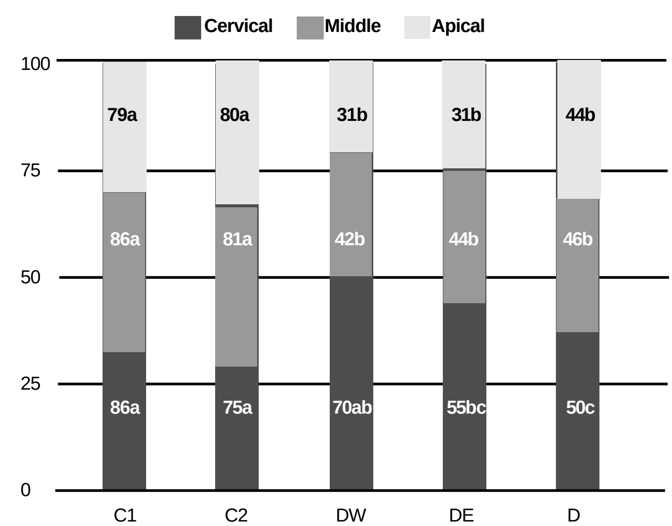

Figure 4: The permeability index of the dye penetration in depth (canal light/root cement).

It was observed that DW, DE and D, in spite of being highly capable of penetrating the length of the canal, did not reach a high PI in the depth of dentin in all thirds evaluated.

DISCUSSION

Teeth with infected root canals, particularly those in which the infection reached the periradicular tissues, are a common problem in the primary dentition. Early loss of these teeth can cause a number of problems, including space loss for successor permanent teeth. Thus, every effort should be made to maintain the primary teeth in the dental arch, disease-free, and provide their functional rehabilitation.[9]

The rapid progression of dental caries in primary teeth produces pulp damage, due to pulpar tissue contamination by bacteria and the toxins derived from them. Pulp damage requires us to perform treatment to remove the injured tissue, eliminating the microorganisms present in the canals and dentinal tubules, and preventing recontamination after treatment.[10] In this context, cleansers are very important to the success of the endodontic treatment because they act as lubricants during instrumentation, flush debris and bacteria out of the canal, and react with pulpal necrotic tissues, microorganisms and their by-products.[11]

Endodontic treatment involves the mechanical instrumentation of the root canals and the use of an auxiliary cleanser. Irrigating substances, such as cleansers, have been used with the objective of eliminating pulpal remains and residues, increasing dentinal permeability (removing the smear layer), facilitating instrumentation and for promoting cleaning and disinfection of the root canals.[3,12-18] In addition, they are soluble in water and biocompatible with the periapical tissues.[19]

This study showed that 0.5% NaOCl solution alone produced the lowest ability to increase the root dentin permeability by dye, in comparison with 1% and 2% chlorhexidine digluconate gels, both in length and in depth of dentin. In spite of primary dentin having a lower amount of mineral and higher organic matrix, it would appear that the Dakin's solution alone was not able to reach the permeability index showed by the other solutions and gels. The lower effect of Dakin's solution can be attributed to the low concentration of NaOCl (0.5% NaOCl) and to it being neutralized by boric acid. Marshall *et al.*[3] using 5.25% NaOCl solution observed a small increase in dentinal permeability of the root dentin. The NaOCl concentration used in this study was very low, but was the choice for primary pulp therapy, because of its highly biocompatibility, as well as its low organic dissolution action when used alone.

Changes in the protein components, which lead to alterations in dentin permeability, have also been reported. The structure of the interface between dentin and localized formations of irregular secondary dentin varies considerably. If there is no tubular communication between primary and secondary dentin, the interface will act as an impermeable barrier. Changes in dentin, including irregular secondary dentin formation, affect the permeability of the tissue.[20]

Dentin consists of both organic and inorganic components. It is a highly mineralized tissue that is penetrated by tubules across the root canal and pulp chamber to the external surface of the root. Calcium (Ca) and phosphorous (P) present in hydroxyapatite crystals are the main inorganic elements of dentin. Dogan and Çalt[21] showed that 2.5% NaOCl alone changed the Ca/P ratio of root dentin significantly. Any change in this ratio can alter the original proportion of organic and inorganic components, which could increase dentin permeability and solubility. It is possible that the use of high concentration of sodium hypochlorite as an irrigant may affect the inorganic material preventing further dissolution of the dentin or could dissolve the organic components and leave a smear layer of the mineralized tissue. Therefore, this mineralized layer could be one of the reasons why 5.25% NaOCl irrigation does not decrease de Ca and P levels of dentin surface significantly. In addition, changes in the phosphorous (P) and magnesium (Mg) levels may cause damage to the organic component of the matrix.[7]

NaOCl is a weak base that acts on the albumin (remains of pulpal tissue, foods and microorganisms), denaturing them and making them soluble in water. Like soap, it facilitates the removal of debris from the root canals and in spite of being a necrotizing agent (acting on the organic matter), it is hardly irritating to live tissues.[22] The NaOCl alkali in contact with organic products in decomposition liberates chlorine and nascent oxygen promoting its bactericidal action.[22] Sodium hypochlorite has long been the irrigant of choice because of its antimicrobial activity and tissue dissolving ability, but it has been found to cause severe inflammatory reactions when placed in contact with vital tissues[23,24] aside from its unpleasant odor and taste.[10]

Moreover, the very low concentration of NaOCl used in this study, when it was associated with Endo PTC (10% urea peroxide, 15% Tween 80 and 75% Carbowax) allowed significantly increased root dentin permeability in comparison with Dakin's solution alone. The peroxides are oxidizing agents that react chemically, liberating great amounts of nascent oxygen, which explains their bactericidal action. The effervescence due to oxygen liberation contributes to the removal of pulp tissue remains and dentinal particles during the chemical-mechanic preparation to suspend the particles. In Brazil, 10% urea peroxide is found under the trade name of Endo-PTC and is made up of: 10% Urea Peroxide, 15% Tween 80 and 75% Carbowax. International literature shows that urea peroxide used in an anhydrous glycerol base, but without any added detergent, is sold under the name of Gly-Oxide.

The urea peroxide in the Carbowax base possesses several desirable characteristics for root canal irrigation in primary teeth. It presents detergent and homeostatic properties, in addition to being non-irritating to the periapical tissues and non-allergenic. Rome *et al.*[14] and Stewart et al.[25] observed that the bactericidal activity of the urea peroxide product (Gly-Oxide) was shown to be superior to 3% hydrogen peroxide in the preparation of infected root canals. The association of urea peroxide/sodium hypochlorite liberates great amounts of nascent oxygen, which produces effervescence in addition to its bactericidal action, facilitateing the removal of solid and semi-solid particles from the root canals.[12] As also shown by Stewart *et al.*[25], the use of urea peroxide associated with sodium hypochlorite promotes significant increase in dentinal permeability to dye and drugs.[25] However, when used separately it only promotes a small increase in the dentinal permeability index, according to Marshall *et al.*[3] Nevertheless, when 5.25% NaOCl solution was used in association with 3% hydrogen peroxide solution, a significant increase in permeability was observed. The results obtained in this study corroborate those obtained by Marshall *et al.*[3]

Very few studies in literature target primary teeth. A study conducted by Bengston *et al.*[26] and Bengston *et al.*[27] evaluated the capacity of medicines *in vitro* to improve dentin permeability during the endodontic treatment of primary teeth. They found a higher permeability index for Dakin's solution associated with Endo PTC's when compared with the other groups (Tricresol-Formaline, Endo PTC neutralized to Dakin's Liquid and Distilled Water). They concluded that Endo PTC's associated with Dakin's Liquid was the best technique for increasing the dentin permeability index.

Two main factors that would appear to directly affect root dentin permeability are the formation of a smear layer and a reduction in dentin thickness after root canal instrumentation. This debris layer contains both mineralized and non-mineralized components and is created after instrumentation of the canal. It is interesting to note that the 2.5% NaOCl, its association with Endo PTC, and distilled water did not produce a significantly high permeability index in the apical end of the canal. These results are similar to those found by Abbott *et al.*[28], who carried out a SEM study and verified that NaOCl either associated with EDTA or not, showed a significant decrease in cleaning efficiency at the apical end of the canal.

This study showed the highest permeability index for 1% and 2% chlorhexidine as an endodontic irrigant in primary dentin root canals. This medicine presents the lowest surface tension when compared with other irrigants, such as 2.5% and 5% NaOCl, as demonstrated by Tasman *et al.*[29]. The low surface tension they present could allow the high penetration in the length of the canal and probably in the dentin depth. Although the chlorhexidine alone has a broad spectrum of antimicrobial action and a relative absence of toxicity, it is not known to possess a tissue-dissolving property, which led to its use as an endodontic irrigant being recommended in combination with NaOCl.[25,26]

According to Arends and Bosch[30] microhardness determination can provide indirect evidence of mineral loss or gain in dental hard tissues. Pashley *et al.*[31] reported an inverse correlation between dentin microhardness and tubular density. The degree of mineralization and amount of hydroxyapatite in the intertubular substance are considerable factors in determining the intrinsic hardness profile of dentin structure. In addition, the Arends and Bosch study[30] showed that chlorhexidine did not affect root canal dentin microhardness. Chlorhexidine absorption by dentin might have produced a positive effect.[32,33] This positive effect could probably be an explanation for this result. It is Interesting that both chlorhexidine concentrations demonstrated great ability to increase the permeability, both in canal length and in depth in all its thirds.

Chlorhexidine has been studied for its various properties: antimicrobial activity, residual antimicrobial activity, biocompatibility and action on bacterial lipopolysaccharide with the objective of being an alternative to sodium hypochlorite.[11] However, further studies concerning chlorhexidine action and its effects on dentin surface, dentin permeability, and dentin structure have to be conducted with regard to both primary as permanent teeth.

CONCLUSION

In accordance with the data of this study it was concluded that 1% and 2% chlorhexidine provided the highest permeability index, length in root canal and in depth in dentin, in primary root canals *in vitro.*

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