Insisiva Dental Journal: Majalah Kedokteran Gigi Insisiva
Website: http://journal.umy.ac.id/index.php/di/index

Research Article

The Effect of *Curcuma Longa* Rhizome Extract on the Root Canal Dentin Discoloration

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Received date: October 1st, 2022; revised date: October 23rd, 2022; accepted: November 12nd, 2022
DOI: 10.18196/di.v11i2.16370

Abstract

Root canal treatment aims to remove vital and necrotic tissue, microorganisms, and their products from the root canal. In this case, chemical endodontic irrigants such as NaOCl and Chlorhexidine (CHX) are required. Unfortunately, both have side effects. Twenty percent of *C. longa* rhizome extract may be a potential alternative due to its antibacterial activity and the ability to dissolve the smear layer. However, it contains curcumin, a colorant that can stain dentin. This study aims to evaluate the influence of 20% *C. longa* rhizome extract on root canal dentin discoloration. Thirty decoronated incisor root segments were prepared using the crown-down technique and divided into three groups (n=10). Each group was irrigated with a different solution. Twenty percent of *C. longa* rhizome extract was the treatment group, the combination of 5.25% NaOCl and 2% CHX was the positive control group, and distilled water was the negative control group. UV Vis 2401 PC spectrophotometer was utilized based on the dE*ab value to assess the discoloration on the root canal. The One Way ANOVA and Post Hoc Games Howell tests were employed to analyze the data. The highest discoloration value was revealed in the treatment group, and the least was in the negative control group, with a significant difference between the two. Furthermore, One-way ANOVA showed no significant differences between the treatment and positive control group ($p<0.05$). *C. Longa* 20% extract had an effect on the discoloration of root canal dentin.

Keywords: *Curcuma longa; dentin discoloration; root canal treatment*

INTRODUCTION

Successful treatment of the infection in teeth depends on the removal of vital and necrotic tissue and microorganisms from the root canal. Irrigation plays a significant role in this stage.[1] It serves several functions mechanically and chemically, such as cleaning debris, being a lubricant to facilitate the instrumentation, and dissolving organic and inorganic tissues during instrumentation. Meanwhile, biological purposes are related to the antibacterial effect of the solutions.[2]

Besides the ability as a broad-spectrum antimicrobial agent, an ideal irrigation solution must have several characteristics, such as preventing the formation of a smear layer, being able to dissolve tissue, being a non-toxic agent, having low surface tension, and not staining tooth structure.[2,3] The top three types of irrigation solutions most frequently used were sodium hypochlorite (NaOCl), followed by EDTA, and chlorhexidine (CHX).[4]

Chemical irrigating solutions can cause unexpected side effects in patients.

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NaOCl and CHX can cause toxic effects on human cells and discoloration of teeth.[5,6] The discoloration of enamel and dentin caused by irrigation solutions. It was revealed that the combination of 5.25% NaOCl and 2% CHX (gel or solution) could cause discoloration of the enamel and dentin even after being rinsed with 10 ml of distilled water.[7] It is also in line with a study by Krishnamurthy et al. (2010) that examined the deposits formed due to the interaction between NaOCl and CHX. It demonstrated that the combination of 2.5% NaOCl and 2% CHX caused the formation of brownish-orange deposits along the walls of the root canals.[8] These side effects encourage researchers to look for alternative root canal irrigation materials from plant extracts as they have the advantages of antibacterial activity, lower side effects, and are easily obtained.[9]

Turmeric (*Curcuma longa*) is one of the plant extracts that have the potential to be used as an alternative irrigation solution. Kumar et al, reported that 20% of *C. longa* rhizome extract showed antibacterial activity against *Enterococcus faecalis* bacteria with an inhibition zone diameter of 22 m.[10] Dhariwal et al. also revealed that *C. longa* rhizome extract had a similar antibacterial sensitivity as 3% NaOCl in bacteria found in the root canals of infected teeth.[11]

Besides having antibacterial properties, *C. longa* rhizome extract can dissolve the smear layer. Sulgante et al, compared the effectiveness of 0.25% *C. longa* rhizome extract and 17% EDTA and 3% NaOCl dissolving the smear layer in tooth root canals. The results showed no significant difference between the extract and the combination of 17% EDTA and 3% NaOCl in dissolving the smear layer in the middle and apical third of the root canals.[12]

The rhizome of *C. longa* is known to have an active component in the form of curcuminoids, ranging from 3–5%. Curcuminoids are yellow dyes that can be used as a coloring agent with curcumin as the main component. It consist of curcumin (77%), desmethoxycurcumin (18%), and bisdesmethoxycurcumin (5%).[13] Previous studies investigated the effect of colored solutions, including C. longa rhizome extract, on the color stability of bleached teeth. They reported that a 5gr/500ml concentration of *C. longa* rhizome extract could cause discoloration of the enamel.[14] Antibacterial properties and the ability to dissolve the smear layer enabled the rhizome of C. longa to be developed as an alternative irrigating solution. However, it is necessary to consider the presence of curcuminoid components that has the potential to cause discoloration of root canal dentin. Therefore, this study aims to determine the effect of 20% *C. longa* rhizome extract on root canal dentin discoloration.

MATERIALS AND METHODS

This experimental research was conducted at the Biochemistry Laboratory of the Faculty of Medicine, Sriwijaya University, and the Textile Evaluation Laboratory of the Textile Engineering Study Program, Faculty of Industrial Technology, Islamic University of Indonesia.

The sample size was determined using Lemeshow two-side test. The calculation obtained a minimum sample size of 8 teeth with an anticipated dropout of 20% resulting in the number of samples of 10 teeth per group. The total sample size required for this study was 30 tooth roots for three groups. Group A was the negative control group using the specimens immersed in distilled water. The specimens used 20% extract of *C.longa* for Group B, while Group C, a positive control group, used 5,25% NaOCl and 2% CHX as an irrigation solution.

The teeth were carefully selected based on the inclusion criteria: the maxillary central incisors extracted teeth with intact roots and one root canal, and the apical foramen completely closed. The exclusion criteria were maxillary central incisor with root caries, root fracture, root

canal treatment, and discolored central incisors.

Preparation of *C.longa* extract

This study utilized Curcuma longa rhizome material aged 11-12 months from the plantation of the Indonesian Spice and Medicinal Research Institute (Balittro), Bogor, West Java. The rhizomes of *C. longa* were collected, peeled, washed, then sliced longitudinally as thin as possible with thickness ranging from 1-2 mm. The pieces of rhizome were dried using an oven at 50^{o} C for 8 hours, ground to form a fine powder, then added with 96% ethanol solvent with a ratio of powder: solvent 1:6 and macerated for three days. After 3x24 hours, Whatman filter paper no. 1 was used to filter the residues with the filtrate. The resulting liquid was then evaporated until it was ethanol-free using an oven at 40^{o} C to obtain a concentrated 100% extract.[15] Distilled water was used to dilute the extract to a concentration of 20%.

Root specimens preparation

Thirty extracted maxillary central incisors were cleaned from debris and calculus using an ultrasonic scaler. The remaining debris was removed with a rubber cup and pumice and washed under running water. Each crown was separated from the tooth root and cut with a diamond separating disc 1 mm above the cement-enamel junction (CEJ). The root segment was placed in a wax block measuring 15x15x17 mm, leaving ±1 mm above the surface of the block (Fig. 1).

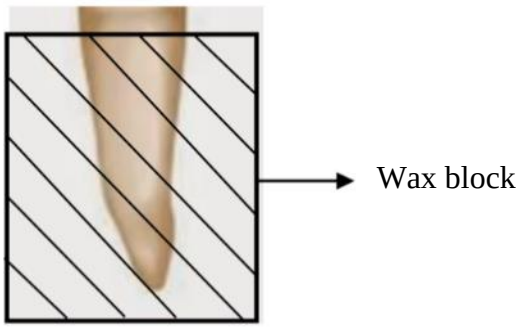

Figure 1. Illustration of a root segment embedded in a wax block.

The root canals of each sample were prepared using a crown-down pressure-less technique, then 3 ml of saline solution was applied with light and passive pressure using a 5 ml syringe with a 27G side-vented irrigation needle. The irrigation needle was inserted into the root canal until it reached the apical third, then withdrawn 2-3 mm to prevent the blockage of the needle and perforation of the irrigation solution into the apical.[16]

After preparation, each root fragment was cut longitudinally in the buccal and lingual directions using a diamond separating disc separating the mesial and distal sides. The tooth roots were then immersed in sterile distilled water in a plastic tube and stored in the refrigerator until used.

Specimen treatment procedure

Prior to the immersion, the entire surface of the root was coated with clear nail varnish three times and then covered with black duct tape, except for the dentin surface of the root canal. Furthermore, the initial color of the root canal dentin surface was measured (measured values of L_0, a_0, b_0) using a UV-Vis 2401 PC spectrophotometer. The dentin surface of the root canal was directed to the optical fiber spectrophotometer and measured.

Three 100 ml measuring cups were prepared and marked according to their respective groups (groups A, B, C), then filled with 50 ml of solution from each group. Root specimens were immersed for 30 seconds then the solution was discarded and replaced. It was repeated every 30 seconds for 5 minutes with the solution renewed. Specimens in Group A and B were immersed in 50 ml of distilled water and 20% C. longa rhizome extract, respectively. In group C, the root specimens were immersed in 5.25% NaOCl solution for 4 minutes 30 seconds, then the final immersion in 2% CHX for 30 seconds.

Afterward, the root specimens were removed from the solution and dried, and the final color of the dentin surface of the

root canals (values L_1, a_1, b_1) was measured using a UV-Vis 2401 PC spectrophotometer using the same procedure as the previous one.

Tooth Discoloration Measurement

The discoloration of the dentin in the root canals was measured using a UV-Vis 2401 PC spectrophotometer. The Commission International de I'Eclairage's (CIE) L* a* b* color system scale was utilized to report the data. Changes in the color of the dentin of the root can be seen from the dE*ab value, obtained from comparing the values of L*, a*, and b* dentin of the root canals pre- and post-immersion. The initial data analysis was conducted using the Shapiro-Wilk normality test ($p>0.05$) and the Levene homogeneity test ($p>0.05$), followed by the One-way ANOVA test. Furthermore, the Post hoc Games Howell test was employed to determine the significant difference between the study groups with a significance level of 0.05 and a 95% confidence level ($\alpha=0.05$).

RESULT

The mean value of dE*ab in the distilled water group, 20% *C. longa* rhizome extract, and 5.25% NaOCl followed by 2% CHX are shown in Table 1. Discoloration of root canal dentin after immersion is shown in Figure 2.

Table 1. The mean value of dE*ab

Group	Mean dE*ab ± SD	P-value
Distilled water	0.6611 ± 0.25360	0.000*
20% *C. longa* rhizome extract	13.052 ± 1.62854	
5.25% NaOCl and 2% CHX	12.287 ± 1.06311	

*p-values are determined by one-way ANOVA

(a) (b) (c)

Figure 2. Root fragment specimen after immersion in (a) distilled water, (b) *C. longa* rhizome extract, and (c) NaOCl followed by CHX

Table 1 demonstrates that the average dE*ab values from the highest to the lowest are C. longa rhizome extract 20%, 5.25% NaOCl followed by 2% CHX, and distilled water, respectively. The Shapiro-Wilk and Levene test results showed that the data were normally distributed (p-value> 0.05) and were not homogeneous (p-value <0.05), respectively.

The result of the One-way ANOVA test revealed a p-value = 0.000 (Table 1), indicating a significant difference in the effect of discoloration in the three groups. Furthermore, a Post hoc test was carried out with the Games Howell test to determine differences between research groups.

Table 2. The results of the Post hoc Games Test Howell

Groups	Distilled water	20% *C. longa* rhizome extract	5.25% NaOCl with 2%CHX
Distilled water		0.000*	0.000*
20% *C. longa* rhizome extract	0.000*		0.446
5.25% NaOCl with 2%CHX	0.000*	0.446	

(*) = indicates a significant difference ($p<0.05$).

Table 2 displays no significant difference between the group of specimens immersed in the *C. Longa* extract and the positive control.

DISCUSSION

Many factors influence the perception of tooth color, including the microstructure of the underlying substances. Enamel is the outer layer and acts as a scattering medium when light enters, and the inner dentin reflects it and is perceived as a color. As the enamel is translucent, it does not entirely conceal the color of the underlying dentin. Thus, dentin plays a significant role in determining tooth color.[17]

Dentin is the thickest in the cervical third of the tooth. The dentin layer is the most chromatic histological layer, causing the color saturation of the teeth to be seen more clearly in the cervical third of the crown of the tooth. Simultaneously, the thinnest layer of enamel is revealed in the cervical area. Therefore, if staining occurs in the pulp chamber and root canal area, and the dye continues to penetrate the dentinal tubules until it reaches the dentin-enamel junction area, it can affect the perception of color in the teeth, causing tooth discoloration. [18]

In this study, the root fragments were immersed in an irrigation solution, and color changes were observed. The light reflected from the dentin surface of the root canals in this study was recorded using a spectrophotometer as a spectral reflectance curve value. The value obtained was converted into L*, a*, and b*. It was then used to determine the value of dE*ab. A dE*ab value < 1 was interpreted as a color change that was not clinically visible. The dE*ab value between 1 and 3.7 indicated that the color change was identified by the observer, but it was still clinically acceptable. The color change could be easily identified by observers when the value of dE*ab was > 3.7.

The 20% *C. longa* rhizome extract group and the positive control group (NaOCl 5.25% and CHX 2%) had a dE*ab value of more than 3.7, indicating a lot of color pigment attached to the dentin surface of the root canal. The Games Howell statistical analysis (Table 2) demonstrated no significant difference in the discoloration effect between the 20% C. longa rhizome extract group and the positive control group.

The C. longa rhizome extract could cause discoloration of tooth enamel that had been bleached previously due to the active component in C. longa in the form of curcumin.[14] The C. longa rhizome extract curcumin could easily stain the collagen network in the oral mucosa.[19] The C. longa rhizome extract strongly colored the collagen network with a yellowish-orange color due to the presence of curcumin.[20]

In relation to these statements, the sample in this study included root dentin containing collagen. Extracted teeth retain large amounts of organic content as solid apatite from the external teeth can preserve internal organic components for a long time.[21] The basic principle of staining is due to the presence of ionic bonds between tissue components and C. longa coloring constituent.[20]

In addition, another factor that can cause discoloration of root canal dentin by C. longa rhizome extract is the molecular weight of curcumin which is relatively low, 368.38 g/mol.[22] A dye with low molecular

weight, such as Rhodamine B (479 g/mol), can quickly penetrate the dentinal tubules and cause staining of the dentin.[23] Furthermore, Methylene blue and toluidine blue with low molecular weights (373.91 g/mol and 107.17 g/mol, respectively) are also capable of causing staining of teeth after application in root canals.[18]

The staining effect in the positive control group specimens could be related to the presence of a brownish-orange precipitate formed due to the interaction between NaOCl and CHX. It also becomes a limitation in this current study due to the lack of rinsing procedures between the two solutions.

CONCLUSION

Based on the study's results, it can be concluded that there was an effect of 20% turmeric (Curcuma longa) rhizome extract on the discoloration of the dentin of the root canals. *C. longa* rhizome extract 20% had the potential to be used as an alternative irrigation solution, although it can cause discoloration. Therefore, further research needs to be conducted to minimize discoloration.

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