**Comparison of Immersion Time between Strawberry (Fragaria x ananassa) Juice and 35% Carbamide Peroxide on Tooth Discoloration**

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**Abstract:**
Tooth color is one of the aesthetic problems in society. This condition is influenced by extrinsic and intrinsic factors. 35% Carbamide peroxide is an ingredient approved by the American Dental Association (ADA) as safe and effective tooth whitening. Strawberry (Fragaria x ananassa) is a natural substance containing ellagic acid that can be used for tooth whitening. The purpose of this study is to determine the tooth discoloration after the immersion of F. x ananassa juice 100% and 35% carbamide peroxide. The method of this research was a pure laboratory experiment. The sample used was 24 premolar postpartum teeth which had been discolored using tea. The specimens were divided into 2 groups, the first group was immersed with F. x ananassa juice 100% and the second group was immersed with 35% carbamide peroxide, respectively for 30, 60, and 90 minutes. The color of the immersed teeth was then measured by a spectrophotometer and a shade guide to determine the color change before and after the treatment. Data were analyzed using Two Way ANOVA and Kruskal-Wallis test. The results of this study indicated that the immersion of F. x ananassa juice 100% was able to make the teeth become brighter, with the same level of
brightness using 35% carbamide peroxide. The conclusion of this research was F. x ananassa juice can be used as an alternative material for tooth whitening.

**Key words:** Teeth Whitening; Strawberry Fruit (Fragaria x ananassa); 35% Carbamide Peroxide; Shade Guide; Spectrophotometer

**INTRODUCTION**

The aesthetic is an important thing for the society today. A research shows that 95% of people having tooth discoloration into yellow and even brown feel dissatisfied with their physical appearance. It can be said that tooth discoloration is an important aesthetic problem. Tooth discoloration into yellow and even brown can make people feel uncomfortable and lack of confidence.

The color of the tooth is influenced by the combination of internal discoloration i.e. dentine and enamel and the external discoloration i.e. the stain on the tooth surface. External discoloration can be caused by substances entering the oral cavity, such as coffee, tea, chocolate, tobacco, mouthwash, or dental plaque. Causes of internal discoloration are due to systemic, metabolic, genetic, and local causes.

Bleaching is a teeth whitening technique that changes tooth color with chemical improvement process. Bleaching material commonly used in dentistry is carbamide peroxide because it is safer and causes fewer side effects. Carbamide peroxide with a concentration of 35% is commonly used in in-office bleaching procedures, i.e. bleaching done in clinics or hospitals by dentists. The mechanism is (O-) react with Ca\(^{2+}\) in hydroxyapatite (Ca\(_10\)(PO\(_4\))\(_6\)(OH)\(_2\)) to form CaO, causing the teeth to look whiter.

One of the natural ingredients having tooth whitening effect is strawberry fruit (Fragaria x ananassa) because it has ellagic acid content. This study was conducted to examine the most effective time for F. x ananassa juice 100% to be able to change tooth discoloration compared with 35% carbamide peroxide.

**MATERIALS AND METHOD**

The design of this research is a pure laboratory experiment conducted in Pharmacy Laboratory of Muhammadiyah University of Yogyakarta (UMY). The tested sample consisted of 24 premolar postpartum teeth. Tooth color measurements before and after the immersion of F. x ananassa juice 100% were done using UV-2401 PC spectrophotometer (Shimadzu, Japan) at Textile Engineering Laboratory of Islamic University of Indonesia and using shade guide (Vita Classical, United States) at Dental lab RSGM UMY.

The experiment began by soaking the postpartum teeth into black tea for 12 days until discoloration occurred on the enamel surface. Then, tooth color measurement using a spectrophotometer was done by performing irradiation to the teeth to determine the color value (\(dE^{*ab}\)) of the teeth. In addition, tooth color measurement using a shade guide (Vita Classical) was performed as well by matching the color of the specimen teeth with the color code on the shade guide. Then, the dental specimen is divided into two groups at random. The first group was immersed with 35% carbamide peroxide per 30 minutes, 60 minutes, and 90 minutes, and the second group was soaked with F. x ananassa juice 100% with the same duration. Immersion duration was adjusted with the application of in-office bleaching materials. After being treated, re-measurements were completed using the spectrophotometer and the shade guide.

The data were analyzed using Two Way ANOVA on spectrophotometer measurement data and using the nonparametric test of Kruskal-Wallis and Mann Whitney U Test on shade guide measurement data.

**RESULT**

The measurement data using spectrophotometer were analyzed using Two Way ANOVA test to compare 1) the type of immersion material (F. x ananassa juice 100% and 35% carbamide peroxide); 2) immersion time (30, 60, and 90 min), and 3) type of material and immersion time to see the significant differences.

The result of the test using Two Way ANOVA in Table 1 showed the significance of \(p>0.05\) (0.707) on the comparison of the immersion material type. Thus, there was no significant difference between F. x ananassa juice 100% and 35% carbamide peroxide. Comparison between immersion time at 30, 60, and 90 minutes showed the significance of \(p>0.05\) (0.523), so it can be concluded that there was no significant difference in immersion time. Comparison between material type and immersion time showed the significance of \(p>0.05\) (0.267). Hence, there was no difference between material types compared with immersion time.

The measurement data using shade guide was ordinal scale, so it was analyzed using Kruskal-Wallis statistic test to compare: 1) immersion time of F. x ananassa juice 100%; 2) immersion time of 35%
carbamide peroxide; and 3) the type of immersion material to see the significant differences, as seen in Table 2.

Based on Kruskal-Wallis test, a significant difference of \((p<0.05)\) on the type of immersion material to tooth discoloration was found. Therefore, Mann Whitney U Test was performed to determine the most significant materials for tooth discoloration.

The value of tooth discoloration before and after immersion in Table 2 showed the significance of 0.135 \((p>0.05)\) for F. x ananassa juice 100% and the significance of 0.099 \((p>0.05)\) for carbamide peroxide which means time/duration of immersion had no effect on tooth discoloration. The result of statistic test comparing tooth discoloration before and after immersion between F. x ananassa juice 100% and 35% carbamide peroxide obtained the significance of 0.001 \((p>0.05)\) which means there was a significant difference between the immersion.

### Table 1. Summary of Two Way ANOVA Statistics on Tooth Discoloration Compared to Material Type, Immersion Time, and Material Type and Immersion Time Using Spectrophotometer

| Dependent Variables | Sig. |
|---------------------|------|
| Type                | 0.707* |
| Time                | 0.523* |
| Type of Time *      | 0.267* |

Description: (*) there was no significant difference \((p>0.05)\)

Table 2. Summary of Kruskal-Wallis Statistic Test on Tooth Discoloration after the Immersion of F. x ananassa Juices and 35% Carbamide Peroxide using Shade Guide for 30, 60, and 90 Minutes.

| Tooth discoloration | Sig. |
|---------------------|------|
| Immersion time of F. x ananassa juice 100% | 0,135 |
| Immersion time of 35% carbamide peroxide | 0,099 |
| Type of immersion material | 0,001* |

Description: (*) there was significant differences \((p<0.05)\).

Table 3. Summary of Mann-Whitney U Statistic Test on Tooth Discoloration after the Immersion of F. x ananassa Juices and 35% Carbamide Peroxide using Shade Guide for 30, 60, and 90 minutes.

| Type                | Average | Sig. |
|---------------------|---------|------|
| F. x ananassa Juice 100% | 11.17   | 0.353* |
| 35% Carbamide peroxide | 13.83   |       |

Description: * There was no significant difference \((p>0.05)\)

Therefore, the researchers continued to perform Mann Whitney Test to know the most effective material in tooth whitening.

Based on the Mann Whitney Test table (Table 3), the average rank of F. x ananassa 100% was 11.17, while the average rank of carbamide peroxide was 35% higher that is 13.87 with the significance of 0.353 \((p>0.05)\). Based on these data, it can be concluded that the tooth whitening process was more effective at 35% carbamide peroxide immersion, but there was no significant difference with F. x ananassa juice 100%.

### DISCUSSION

This study used an external bleaching technique performed by immersing dental specimens into F. x ananassa juice 100% and 35% carbamide peroxide for 30 minutes, 60 minutes, and 90 minutes. Tooth color measurements were performed using a spectrophotometer and a shade guide.

Tooth color measurement using the spectrophotometer was done by irradiating the tooth surface which was divided into three coordinate axes i.e. \(L^*, a^*,\) and \(b^*\). The sum of the values of \(L^*, a^*,\) and \(b^*\) is the total value of the color intensity absorbed by the spectrophotometer called \(dE*ab\).

Tooth color measurements using the shade guide was done visually by comparing the color of the dental specimen crown with the available color on a shade guide consisting of 16 different tooth colors.

The result of the research by comparing the change value of \(dE*ab\) data between F. x ananassa juice 100% and 35% carbamide peroxide using spectrophotometer showed no significant difference. Tooth color measurement using shade guide with Kruskal-Wallis followed by continued test using Mann Whitney Test showed the average value of carbamide peroxide was higher than the average value of F. x ananassa juice 100%, but the value difference showed no significant difference. Therefore, it can be concluded that F. x ananassa juice 100% has the bleaching ability which is almost the same as 35% carbamide peroxide because both contain tooth whitening ingredients. Carbamide peroxide contains peroxide material, while F. x ananassa has ellagic acid content.

The peroxide as an oxidizing agent with free radicals having unpaired electron will be released and then received by the enamel, resulting in the oxidation process. These electrons are oxidized by organic matter which causes tooth discoloration. Free radicals from peroxide are perhidroksil and oxygenase. After the formation of perhidroksil in...
large quantities, it will react with unsaturated bonds and cause energy absorption in the organic molecule of enamel. The result is smaller organic molecules with lighter colors.\(^8\)

The researchers then compared the tooth discoloration after the immersion of \(F. x\) ananassa juice 100% and 35% carbamide peroxide at 30, 60, and 90 minutes. There was no significant difference between the color of the teeth after being measured using the spectrophotometer and the shade guide. The absence of tooth discoloration after the 30-minute duration is possible because the experimental teeth have passed the endpoint, a condition in which the teeth in the bleaching process cannot be whiter again. Teeth with endpoints may be characterized by more translucent color.\(^9\)

Tooth discoloration between specimens and the time of immersion of \(F. x\) ananassa juice 100% and 35% carbamide peroxide at 30, 60, and 90 minutes showed no significant difference. Therefore, it can be concluded that the color of the teeth produced by the immersion of \(F. x\) ananassa juice 100% and 35% carbamide peroxide was not different, making \(F. x\) ananassa juice as a natural ingredient that can be used as an alternative tooth whitening agent. This is in accordance with previous research conducted by Juwita, 2008 cit. Hartanto et al. (2012),\(^10\) which stated that the application of \(F. x\) ananassa paste within 2 weeks clinically had the same tooth-whitening ability as factory-made tooth whitening. The content of ellagic acid in \(F. x\) ananassa can produce free radicals in chromophore chain breakdown to produce an email bleaching comparable to carbamide peroxide.\(^10\)

The mechanism of \(F. x\) ananassa in tooth whitening has been reported by some researchers, namely by the presence of oxidation reactions. The discoloration of enamel and dentine undergoes an oxidation reaction by a strong oxidizing agent of ellagic acid in \(F. x\) ananassa. The ellagic acid destroys the dye molecule with free oxygen released, so the color becomes neutral and causes a bleaching effect.\(^10\) However, this reaction results in unpaired electrons bind to free radicals from both organic and inorganic dental matrices to achieve stability, affecting enamel demineralization and resulting in reduced enamel hardness.\(^10\) Hartanto et al. (2012),\(^10\) in their study stated that the enamel color change according to the original color occurred after 3 weeks of \(F. x\) ananassa paste application, whereas the decrease of enamel hardness occurred after \(F. x\) ananassa paste application for 2 weeks. Therefore, the effective time in using the paste \(F. x\) ananassa as bleaching material was 2 weeks.

A lot of research has been done related to natural materials as bleaching agents. Nurbauety et al. (2014)\(^5\) stated that citric acid in lime fruit flesh had the same OH group as the ellagic acid in \(F. x\) ananassa. The results of the research concluded that lime with a concentration of 2.5% had a pH of ±3 which was almost the same as pH of \(F. x\) ananassa and in-office teeth whitening. Tooth discoloration because of Robusta coffee can be removed within 30 minutes based on this study.

In addition, Sugianti (2014)\(^7\) used Rosella (Hibiscus sabdariffa) containing bioactive saponins and vitamin C (ascorbic acid) to whiten teeth for 3-day immersion. Saponins which bound the dyes due to the resulting foamable was able to whiten teeth, while vitamin C function was to damage the dye that stuck to the teeth. The results were the tooth color became neutral and white.

Based on Nuzulya et al.’s (2012) study\(^9\), the malic acid contained in apples with juice concentration of 75% was able to whiten the tooth color after experiencing discoloration by coffee. Furthermore, Syahland and Setyawati (2013)\(^12\) examined wine due to its peroxidase enzymes which form hydrogen peroxide from water and oxygen to increase the speed of reducing tooth color. However, grape juice is unable to whiten teeth; both observed using a shade guide and a spectrophotometer.

**CONCLUSION**

Based on the research, it can be concluded that strawberry (\(Fragaria x ananassa\)) can be used as an alternative agent for tooth whitening with no significant difference in 30-minute, 60-minute, and 90-minute immersion of \(F. x\) ananassa juice. Therefore, it can be concluded that in the 30-minute immersion, \(F. x\) ananassa juice has been effective in whitening the teeth. The absence of tooth discoloration after being immersed for 30 minutes in \(F. x\) ananassa juice 100% or 35% carbamide peroxide is probably due to the endpoint of the tooth color.

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