



The Difference of Color Change After Using the Red Watermelon Extract (*Citrullus Lanatus*) and Commercial Toothpaste Towards Tooth Discoloration (in Vitro)

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Abstract. Background: Aesthetics has a value that is not absolute and very subjective. Discoloration teeth that can interfere with aesthetics require treatment, the instance is whitening toothpaste. The amount of chemical content contained in toothpaste is considered to have an effect on health. 99% malic acid in red watermelon has been shown to whiten teeth. This research aimed to determine the difference between the use of red watermelon extract (*Citrullus lanatus*) toothpaste and commercial toothpaste on color changes in vitro.

Methods: This research is a true experimental laboratory used 12 samples of post-extraction incisors and canines. All samples were discolored with black tea. The treatment group was divided into 3 groups, there are the group that was brushed with extract red watermelon, the group that was brushed with commercial toothpaste, and the group that was brushed with aquadest. Each group was brushed for 56 min. Measurement of color changes used shade guide and spectrophotometer was carried out before and after brushed. The results of the color changes were analyzed used One Way Annova Test and Paired T-Test.

Results: The result of this research used Paired T-Test obtained that the three groups has significance value of $p > 0.05$ and the result used One Way Annova Test has a significance value of 0.307 ($p > 0.05$).

Conclusion: This study showed that there was no significant difference in tooth color between using extract red watermelon, commercial toothpaste, and aquadest.

Keywords: Watermelon extract · Whitening toothpaste · Aquades

1 Background

Aesthetics is something that can affect appearance. Several factors such as shape and proportion, color and position, and tooth size are things that affect dental esthetics [1]. The most significant factor affecting dental esthetics is tooth color [2].

The color of a tooth is influenced by its constituent layers, such as the translucency and thickness of the enamel and the yellowish color of the dentin. Color changes that occur in both layers or in the coronal pulp structure can result in changes in light transmission that affect the color of teeth [3–5]. Tooth discoloration varies according to etiology, appearance, localization, severity, and persistence of tooth structure. It can be classified into [3], namely extrinsic or intrinsic discoloration or a combination of both [6].

External factors staining teeth such as drinking, food, smoking habits and the use of certain drugs can leave stains attached to the tooth surface. Internal factors for tooth staining include fluorosis, the aging process, and the use of tetracycline antibiotics in pregnant women [7, 8].

There are several things that cause tooth discoloration on a daily basis, including plaque, food and drink, tobacco use, poor oral hygiene, calculus, and gingival bleeding [6]. The staining of teeth needs to be overcome by doing teeth whitening treatments. It is also known that people's desire to have whiter teeth has increased over the last decade and many teeth whitening products are claimed to be able to whiten teeth either by bleaching method or by removing extrinsic stains [9].

There are three main approaches to teeth whitening, namely whitening at home under the supervision of a dentist, whitening in a clinic, and whitening using over-the-counter whitening products such as strips, mouthwash, and toothpaste [10]. The use of toothpaste is one option that can be done every day to clean extrinsic staining on teeth [11].

Generally, toothpastes have the same functional basic ingredients and each ingredient has a specific role in the formulation. These materials include abrasives, humectants, thickening agents, surfactants, active substances such as fluoride, sweeteners, flavors, opacity agents, buffering agents, and preservatives [12].

The return of the back to nature trend has caused people to become increasingly aware of the importance of using natural ingredients for health [13]. Based on research conducted by Nur and Setyawati (2020) showed that the malic acid in red watermelon can whiten the color of teeth [14]. The malic acid contained in red watermelon is 99%, higher than that of apples (95%) and cherries (94%). The malic acid content in watermelon belongs to the carboxylic acid group which can whiten teeth through the oxidation process of the enamel surface so that it becomes neutral and produces a whitening effect [15].

One of the commercial whitening toothpastes on the market is Pepsodent Plus Whitening. This toothpaste has 2 layers, namely a white paste layer containing fluoride and calcium and a blue paste layer containing perlite. Pepsodent has claimed that the toothpaste can whiten teeth if used for 2 weeks because of the perlite content. Perlite is an abrasive that serves to remove debris and extrinsic stains on teeth. Perlite is a strong abrasive with a higher hardness than hydroxyapatite. Therefore, this material can damage exposed enamel and dentin and have a risk of damaging the gingiva if excessive pressure is used when brushing the teeth [16].

Based on the description above, the researcher was interested in knowing the difference between the use of red watermelon extract (*Citrullus lanatus*) and commercial toothpaste on tooth discoloration *in vitro*.

2 Method

In this research, pure in vitro laboratory experimental research is used. The research subjects used were post-extraction permanent incisors and canines as many as 12 teeth which were divided into 3 groups. The inclusion criteria included upper and lower permanent incisors and canines with intact roots. The red watermelon used is fresh and ripe. Meanwhile, the exclusion criteria were primary teeth, anomalous teeth, teeth with caries, and teeth with restorations.

The tools used in this study were UV-2401 PC Spectrophotometer, VITA Classical shade guide, plastic containers, stationery, black duct tape, ziplock plastic, digital scales, mortar and pestle, spatel, and toothbrush. In addition, the ingredients used are red watermelon (*Citrullus lanatus*) fruit extract, toothpaste formulation ingredients, "Pepsodent Plus Whitening" commercial toothpaste, incisors and canines, black tea, hot water, and distilled water.

This research consists of a preparation stage and an implementation stage. The preparation stage carried out was collecting and dividing the research subjects who used 4 teeth for each group, making red watermelon extract using the 96% ethanol maceration method, isolating the teeth from the cervical part to the root of the tooth and marking each sample, making black tea solution. And soaking the sample for 6 days using black tea solution, and then measuring the color of the research subject after soaking or staining the teeth using a shade guide and a spectrophotometer.

Furthermore, at the implementation stage, the teeth were given treatment according to their group for 56 min, namely; (1). The first group: the samples were rubbed using red watermelon extract (EBSM); (2). The second group: the samples were treated by rubbing using a commercial whitening toothpaste "Pepsodent Plus Whitening"; and (3). The third group: the samples were rubbed using distilled water. The three groups that had been treated were measured using a shade guide and a spectrophotometer.

The analytical test method used is the Paired T-Test test to determine the difference before and after, and the One Way Anova test to determine the average comparison of the three groups. In this analysis test used a significance level of 0.05 and a 95% confidence level ($\alpha = 0.05$).

3 Result

The results of organoleptic tests carried out on the application of EBSM and commercial toothpaste in the form of observations of shape, smell and color. The results of the evaluation of the two toothpastes can be seen in Table 1.

The results of measurements using a Shade guide and a spectrophotometer before and after scrubbing can be seen in Table 2. In this case, sample number 1 shows the color A1 with an E value of 6.60 before applying EBSM toothpaste and changes color to B1 with an E value of 15.10 after applying it. This shows that the application of EBSM toothpaste can affect the discoloration of the sample. Meanwhile, sample number 5 from the commercial toothpaste smearing group had a D2 color with an E value of 1.74 before applying and changed to A2 with an E value of 10.66 after application. In subjective measurements using a shade guide, the color score of sample number 5 has decreased

Table 1. Evaluasi Pasta Gigi

No.	Evaluation	Commercial	EBSM
1	Organoleptik		
	Shape	Mild solid	Mild solid
	Color	White-blue	Cream
	Odor	Mint	Sugar
2	Homogeneity	Homogen	Homogen
3	pH	7	6

Table 2. Shadeguide and Spektrofotometer

No.	Sample	Shadeguide		ΔE	
		Before	After	Before	After
1	EBSM toothpaste	A1	B1	6.60	15.10
2		A35	A2	53.45	36.01
3		A3	A1	17.34	13.65
4		A2	A2	19.37	22.38
5	Commercial tooth paste	D2	A2	1.74	10.66
6		D3	A2	12.25	12.52
7		A35	A2	19.09	20.59
8		D2	A1	10.19	25.19
9	Aquadres	A35	A35	13.05	14.55
10		D3	D3	10.47	16.77
11		A2	A2	12.35	31.93
12		A1	A1	5.72	6.54

by 1 level, but the value of E in objective measurements using a spectrophotometer has increased. This may be due to poor lighting when measuring the second color using a shade guide.

Sample number 9, which is included in the aquades application group, did not change color when measured using a shade guide, namely A35 during the first and second measurements. On the other hand, sample number 9 in the measurement using a spectrophotometer experienced an increase in the value of E by 1.50. This can be caused by the absence of a chemical reaction that occurs when applying aquades which only contains H₂O molecules.

To find out the difference between before and after smearing on the three groups, the Paired T Test was carried out. Statistical analysis to perform the Paired T Test begins with normality test using the Shapiro-Wilk.

Table 3. ΔE normality value test before and after application

No	Activity	Significance	
		Before	After
1	EBSM	0.248	0.324
2	Commercial tooth paste	0.912	0.518
3	Aquades	0.316	0.658

Table 4. Paired T-test

No.	Activity	Significance
1	EBSM	0.696
2	Commercial toothpaste	0.159
3	Aquades	0.204

Table 5. Difference Nilai ΔE

EBSM tooth paste		Commercial tooth paste		Aquades	
No.	Difference	No.	Difference	No.	Difference
1	-8.5	5	-8.92	9	-1.5
2	17.44	6	-0.27	10	-6.3
3	3.69	7	-1.5	11	-19.58
4	-3.01	8	-15	12	-0.82

Based on Table 4, it can be seen that the EBSM toothpaste application group has a significance of 0.696 with an average value of 2.405 ($p > 0.05$), the commercial toothpaste application group has a significance value of 0.159 with an average value of -6.4225 ($p > 0.05$), and in the third group with aquades, the average value was -7.05 and the significance value was 0.204 ($p > 0.05$). From the three groups, it can be seen that all groups have $p > 0.05$ which indicates that there is no significant difference between before and after smearing in the three groups.

Table 5 shows the difference in E values before and after smearing in the three groups. The EBSM smear group had the largest average E value of 2,405, the highest difference was 17.44 in sample number 2 and the lowest value was -8.5 in sample number 1. In the group of smearing with commercial toothpaste with an average of -6.4225, there was a difference the highest value is -0.27 in sample number 6 and the lowest value is -8.92 in sample number 5. Then in the aquades smearing group, the highest difference value is in sample number 12 with a value of -0.82 and the lowest value in sample number 11 is -1958, and an average of -7.05.

Table 6. Shapiro-Wilk *normality test*

No.	Treatment	Significance
1	EBSM	0.744
2	Commercial toothpaste	0.478
3	Aquades	0.156

Table 7. One way Annova

ΔE	Significance
223.621	0.307

Based on the difference value data in Table 5, an analysis of the Shapiro-Wilk normality test was carried out. In Table 6, it can be seen that the significance values in the three groups, namely the EBSM toothpaste application group, the commercial toothpaste application group and the aquadest application group, each of which has a significance value of 0.744, 0.478 and 0.156. The three groups have a significance value greater than 0.05 which indicates the data is normally distributed. Furthermore, to test the hypothesis with a normal distribution, the One Way Annova parametric test was carried out.

In the One Way Annova test using the difference in E data, a significance value of $p > 0.05$ was obtained. From this, it can be concluded that there is no significant difference between the three smearing groups. The analysis data of the One Way Annova test can be seen in Table 7.

4 Discussion

This study discusses external whitening techniques on extracted teeth. The tooth samples were divided into 3 treatment groups, namely the group that was scrubbed using EBSM, commercial toothpaste and distilled water.

In the compounding of EBSM toothpaste, the active substance used as a whitening agent is 10% of the 200g preparation of EBSM toothpaste. This is because the consideration that when using a higher concentration of red watermelon extract causes the viscosity of EBSM toothpaste to be lower. In addition to the use of these active substances, whitening using EBSM toothpaste is also supported by the use of glycerin. Glycerin which is the best humectant can be used to make teeth shiny after use such as the use of optical agents [17]. According to Değer & Müjdecı (2020), optical agents are an alternative approach to whitening teeth by lowering the b^* value which is characterized by a temporary decrease in yellowish color.

The commercial toothpaste used in this study was Pepsodent Plus Whitening. The main content is perlite which is a natural material with relatively large particles of about 20–25 μm and has sharp edges that can produce a mechanical effect [19]. Commercially,

perlite can be found in toothpaste with a concentration of 0.7%. Because of these relatively large particles, in the same period of time, perlite is more effective in whitening teeth when compared to Speedy Whitening Agent (SWA) and micro-cleansing crystal [20]. On the other hand, the effect of using perlite can cause exposed enamel and dentin to be damaged and possibly damage the gingiva also when brushing teeth using excessive pressure [16].

The formulated EBSM toothpaste was evaluated for homogeneity test, organoleptic test and pH test together with commercial toothpaste. The two toothpastes have homogeneous characteristics and different colors due to their content, namely the presence of coloring agents in commercial toothpastes. In addition, both of these toothpastes have a pH that resembles the pH of oral saliva. Seralurin et al. [21] in his study said that the pH of saliva is normal, which ranges from 5.6 to 7.0 with an average pH of 6.7. On the other hand, according to Abbas et al. [22], the normal pH of saliva is between 6.8–7.5. However, EBSM toothpaste still has a safe and not too low pH. This is supported by Gratia et al. [23] which states that in an acidic environment ranging from 4.5 to 5.5 can cause irritation and support the growth of acidogenic bacteria.

Before scrubbing, each sample was soaked in black tea for 6 days. Then the tooth color was measured twice, namely before and after brushing the sample. In the first measurement, there were 2 A1 scores (red–brown level 1), 2 A2 scores (red–brown level 2), 1 A3 score (red–brown level 3), 3 A35 scores (red–brown level 4), 2 D2 scores (red–gray level 1), and 2 D3 scores (red–gray level 3). Meanwhile, the second measurement resulted in 1 score B1 (red–yellow level 1), 3 scores A1 (red–brown level 1), 6 scores A2 (red–brown level 2), 1 score A35 (red–brown level 4), and 1 D3 score (red–gray level 3).

From these subjective measurements, it can be seen that there is a change in the color score using the VITAPAN classical shade guide before and after brushing the tooth samples in the group that was rubbed with EBSM toothpaste and commercial toothpaste. The color change in EBSM toothpaste occurs due to the presence of malic acid which can cause an oxidation process because it has an OH group. Malic acid can bind calcium and cause the occurrence of porosity of enamel crystals. This causes a break in the conjugate bonds with dye molecules and erodes the enamel layer so that it gives a whitening effect and makes the tooth color more neutral [24]. When measuring using a shade guide, lighter colors are indicated by lower scores. On the other hand, darker colors are indicated by high scores [25]. Based on research conducted by Diansari et al. (2019), which examined tooth discoloration after the application of green pears and carbamide peroxide, showed a decrease in shade guide scores followed by a change in tooth color to whiter in samples soaked with 16% carbamide peroxide. Research conducted by Yauri et al. (2021), also supports this, where teeth soaked with virgin coconut oil can whiten teeth which is indicated by a decrease in shade guide scores.

On the other hand, the subjective measurement method using shade guides is inadequate to identify varying tooth colors. In addition, in determining tooth color there are other factors such as lighting, experience and operator understanding of color science. Therefore, additional measurements are needed using digital techniques such as spectrophotometers which can provide better and adequate color compared to using conventional techniques which are subjective [28].

A spectrophotometer is an absorbance measuring device that works by passing light with a certain wavelength on tooth enamel. Some of the light rays will be reflected and some will be absorbed by the color pigments of the teeth. The reflected light will appear as a value of E which is quantitative and indicates a color change. The value consists of 3 color coordinates, namely L* which represents the brightness of the object from black to white, a* represents the variation of red–green and b* represents the variation of the yellow–blue color.

The results of this study can be seen from the E value of the sample after and before rubbing, as well as the difference in the values of the two E data. The value of E of tooth samples after and before brushing mostly increased. When viewed from the color coordinates, the average L* value increases after rubbing. On the other hand, the average values of a* and b* decreased. This shows that there is an increase in the brightness of the color of the teeth and a decrease in the level of redness and yellowness of the teeth. This is similar to the research of Terra et al. (2021) who investigated the effects of using 4% hydrogen peroxide on a daily basis. In this study, there was an increase in the color of the teeth to become whiter as indicated by an increased average L* value and a decreased a* and b* mean value. This shows an increase in the brightness of the teeth and a decrease in the level of chroma which makes the yellowness and saturation of the teeth decrease.

In this study, it can be seen that there was an increase in the value of E after scrubbing, from 15.135 to 18.824. On the other hand, teeth that have changed color to become whiter are indicated by a decrease in the value of E, where the tooth sample will be whiter if the value of E sample is smaller [31]. Setyawati and Nur (2020) in their research on teeth soaked with 100% red watermelon extract, explained that teeth can turn whiter with a decrease in the value of E. On the other hand, Diansari et al. (2019) in his research on changes in tooth color after the application of green pears and 16% carbamide peroxide, had a sample of teeth that experienced a darker discoloration. This is probably due to the contact of pear juice with air. Teeth will turn darker in color if pear juice is left for a long time. In this study, the red watermelon fruit extraction process takes a long time, resulting in the extraction solution being in contact with air.

From the data on the E value that has been obtained, a Paired T-Test was carried out and it can be seen that the data before and after brushing using EBSM toothpaste, commercial toothpaste, and distilled water did not have a significant value with a significance value of each group of 0.696, 0.159 and 0.204. This indicates that the brushing of the tooth samples in the EBSM toothpaste group and the commercial toothpaste group was similar when compared to the distilled water group, i.e. there was no significant change in tooth color after brushing. In addition, the difference data from the value of E before and after treatment were used in the One Way Anova Test to determine the difference in values between the three groups. The results showed a significance of 0.307 ($p > 0.05$), so it could be interpreted that there was no significant difference between the three groups of dental samples.

In this study, the treatment group that was scrubbed using EBSM toothpaste and commercial toothpaste did not have a significant difference with the distilled water group, although subjective measurements showed color changes. In addition, there are several difficulties that are the authors' weaknesses in this study. These include differences in enamel thickness and age of the teeth, as well as the different positions of the teeth on

irradiation in the spectrophotometer before and after. According to Rosidah et al. (2019) in his research, stated that enamel thickness and tooth age can affect discoloration. In a thick layer of enamel and dentin, which thickens with age, it can make the bleaching process take longer and the color changes to vary.

The insignificant difference from the spectro-photometer measurement data before and after rubbing is in line with the study conducted by Sofiani and Yudasmara [29]. In this study, tooth samples soaked in 3% hydrogen peroxide did not change after immersion. This could be due to the non-uniform thickness of the tooth samples, the different tooth positions and the small number of samples so that the statistical calculations did not show too large an effect.

5 Conclusion

Based on the results of the study, a significance value of 0.307 ($p > 0.05$) was obtained which means that there is no significant difference in tooth color between treatments using red watermelon extract, commercial toothpaste and aquades.

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