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The Effectiveness of Gargling with 25% Rosella Tea and 25% Green Tea Infusum to the Mechanism of In Vitro Microbial Culture of the Mouth and Teeth

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ABSTRACT: Gargling is one of the ways to get germs or other substances out from our mouth. When gargling, the remnants of food and germs inside the mouth will come out so as to prevent the occurrence of dental infections and transmission of disease. Gargling in the right way will produce better benefits. According to the results of the Ministry of Health's Basic Health Research Indonesia in 2013 stated that the national prevalence of oral and dental problems is 25.9% [1]. The Indonesian DMF-T index is 4.6, which means Indonesian tooth decay is 460 teeth per 100 people. According to [2] The Role of Antimicrobials is a biological or chemical compound that can interfere with microbial growth and activity, especially destructive microbes and food decay. Antimicrobial substances can be bactericidal (kill bacteria), bactericidal (inhibit bacterial growth, catechins contained in green tea, believed to be able to reduce dental plaque formation with two mechanisms, namely killing causative bacteria such as *Streptococcus mutans* and glycosyltransferase from bacteria [3] Rosella flowers can inhibit the development of *Streptococcus sanguis* bacteria which triggers the formation of dental plaque [4]. The purpose of this study was to determine the effectiveness of use between 25% rosella tea infusum rinse and 25% green tea. The method in this study is a true experiment, which is a real experiment. With the design of the Pretest-Posttest with Control Group, which can be expanded by involving more than one independent variable, the treatment of more than one group with different forms of treatment? Data analysis was performed using the statistical analysis method SPSS program version 23. To find out whether there are differences or not before the normality and homogeneity test of the data was carried out, if the data were normally distributed and homogeneous, the ANOVA version was tested followed by the Tukey HSD test, if it was not normal, use mann-widney-test, with a 95% confidence level H_1 is accepted, if the value of p value is <0.05 . The results showed that rosella tea infusion was 25% more effective than 25% green tea infusum for use rinsing to inhibit the mechanism of invitro microbial culture of the mouth and teeth. This can be seen from the results of the analysis where the value of p value <0.05 . While for the data normality test where the treatment of infusion of rosella tea has a mean \pm SD of 34.90 ± 9.07 , whereas infusum of green tea has a mean \pm SD of 14.56 ± 5.37 , while mineral water has Mean \pm SD 4.81 ± 0.82 .

KEYWORDS: Bacterial Culture, Gargling, Health, Infusum of Rosella Tea and Green Tea, Mouth and Teeth

1. INTRODUCTION

Gargling is one way to get rid of germs or other substances in our mouth. When gargling, the remnants of food and germs in the mouth will come out so as to prevent the occurrence of dental infections and transmission of disease. According to the results of the Basic Health Research (RISKESDAS) Ministry of Health in 2013 stated that the national prevalence of oral and dental problems was 25.9%. The Indonesian DMF-T index is 4.6, which means Indonesian tooth decay is 460 teeth per 100 people. Antimicrobial substances can be bactericidal (kill bacteria), bactericidal (inhibit bacterial growth), fungicidal (kill



mold), fungistatic (inhibit mold growth), or germicidal inhibit germination of bacterial spores. According to [5], Antibacterial compounds are needed to help eliminate inflammation by inhibiting bacterial growth and reducing bacterial concentration in dental plaque. To inhibit the growth of bacteria in plaques Today there are many mouthwashes based on chemicals, this is quite dangerous for the stomach if swallowed. Green tea has an active component, namely catechins and contains 30-40% polyphenols which can inhibit the growth of *Streptococcus mutans* as one component of dental plaque formation[3].

Rosella petals contain active ingredients such as flavonoid, phenol or polyphenols, citric acid, saponins, tannins. Flavonoids function to inhibit the growth of microorganisms, because they are able to form complex compounds with proteins through hydrogen bonds[6]. Rosella flowers can inhibit the development of *Streptococcus sanguis* bacteria which triggers the formation of dental plaques[4]. While the content of green tea, catechins, also has the ability to reduce the formation of *Streptococcus mutans* as a component of dental plaque[7].

Microorganisms need all the elements in their organic matter and all the complementary ions needed for work processes and catalysts. In addition, there must be an energy source to produce proton motions and enable macromolecular synthesis. The need for food substances and metabolic energy sources in microorganisms is very diverse. According to [2]the concentration of minimum inhibition of rosella flower extract of 0.20 g / ml against *Escherichia coli*, *Salmonella typhi* and *Staphylococcus aureus*, rinsing with white tea and green tea are equally effective in reducing plaque accumulation. This is caused by the content of white tea which is a tannin compound which can inhibit the growth of *Streptococcus mutans*. While the content of green tea, catechins, also has the ability to reduce the formation of *Streptococcus mutans* as a component of dental plaque[7].

II. MATERIAL AND METHODS

The research used the true experiment research method, which is a real experiment. That is, in this design randomization or grouping of experimental group members is done randomly, with the pre-test (O1) method then after the pre-test results from the total respondents are obtained, then the calculation of the results of each respondent is then grouped based on the results of the most colonies a few to the most will be sorted and divided into 3 (three) groups in each treatment group then followed (intervention) (X) after some time post-test (O2).

Table 1: Research design

	Pre-test	Treatment	Post-test experimental
Experimental Group A	01	X _(a)	02
Experimental Group B	01	X _(b)	02
Control Group	01	X _(c)	02

Description:

- 01 : Observation of bacterial colonies before rosella tea and green tea infusion gargle as well as mineral water as control
- X_(a) : Gargling with rosella tea infusum
- X_(b) : Gargling with green tea infusum
- X_(c) : Gargling with mineral water as control
- 02 : Observing the number of bacterial colonies after gargling with rosella tea and green tea infusum as well as mineral water as control

The subject of this study were 30 people from the Department of Health Polytechnic Department of Dental Nursing, in this study called sample research, intending to generalize the results of research and raise research conclusions as a validity for the population. Sampling is a stratified sample or stratified sample, that is, sampling is done when the researcher believes that the population is divided into levels or strata. According to [8], the research was carried out by the researchers themselves and the team as the supervisors of microbiology courses, and carried out in the Microbiology laboratory of the Dental Nursing Department and Laboratory of the Medical Laboratory Engineering Department of the Ministry of Health Semarang from September 27th to October 6th 2018.

Data was analyzed by statistical tests using SPSS version 23 with the One Way Anova test method, to determine the different test, a normality and homogeneity test will be conducted first. Furthermore, for further testing (*Post Hoc Test*) using the Tukey HSD test to determine the effectiveness of each variable with a confidence level of 95% ($\alpha = 0.05$). H_1 is accepted, if the value of p Value < 0.05 . Whereas to see the effectiveness will be seen the distribution of the mean value between the treatment groups compared to the control group.

III. RESULTS

A. VARIABLE DESCRIPTION

The distribution of average percentage by the reduction of microorganism colonies before and after gargling is shown in Table 2.

Table 2: Distribution of colonies microorganism average percentage by the reduction of colonies microorganism before and after gargling using rosella tea, green tea infusum as well as mineral water with 30 respondents

NO.	ROSELLA TEA	GREEN TEA	MINERAL WATER
Respondent	Average per respondent	Average per respondent	Average per respondent
1	42.9	25	3.5
2	20.1	6.5	5.4
3	40.5	10.9	4.7
4	35.6	18.7	5.6
5	46.9	15.9	4.2
6	29	8.5	4.9
7	36.9	18.2	5.5
8	40.2	13.4	5.7
9	36.6	15.2	3.5
10	20.3	13.3	5.1
Total	349	145.6	48.1
Total average	34.9	14.56	4.81

Source: Primary data in 2018

Table 2 shows that the percentage of reduction in microorganism colonies was greater in respondents who gargled with rosella tea infusum, which was in average of 34.9%, while those who gargled with green tea infusum amounted to 14.56, while those who gargled with mineral water as a control has 4.81%.

B. NORMALITY TEST VARIABLE DESCRIPTION

The following is the test of normality by the reduction of microorganism colonies with the treatment using rosella tea and mineral water.

Table 3: Normality test by the microorganism colonies before and after gargling using rosella tea and mineral water

N	Variable group	Kolmogorov-Smirnov ^a			Shapiro-Wilk		
		Statistic	df	Sig.	Statistic	df	Sig.
Average distribution	Rosella tea	.231	10	.140	.901	10	.224
	Green tea	.120	10	.200*	.977	10	.947
	Mineral water	.162	10	.200*	.880	10	.129

*. This is a lower bound of the true significance.
Lilliefors Significance Correction

While below is the description of data each group in variable with the detail of its mean, standard deviation, minimum and maximum value.

Table 4: Description of data per group of variables

Variable Group	Mean	Std. Deviation	Minimum	Maximum
Rosella tea infusum	34.9	9.08	20.1	46.9
Green tea infusum	14.7	5.4	6.5	25
Mineral water (control)	4.8	0.82	3.5	5.7

Table 5: Distribution of gargling average between rosella tea, green tea and mineral water with 30 respondents

Independent Variable	Number of Bacteria		Result Difference	Percentage of Reduction (%)
	Before	After		
Rosella tea infusum	28.92	18.56	10.28	35.5
Green tea infusum	69.8	59.52	10.26	14.8
Mineral water (control)	93.68	89.24	4.42	4.8

Source: Primary Data in 2018

- I. The normal distribution test is a test to measure whether the data obtained has normal distribution so that it can be used in
- II. parametric statistics (inferential statistics), in other words, the normality test is a test to find out whether the empirical data
- III. obtained from the field is normally distributed, the data is normal if the significance column is > 0.05. This study used
- IV. normality test Kolmogorov-Smirnov³, as for the results including normal data with a very good score.

Table 6: Distribution of Mean ± SD Average Reduction of Microorganism Colonies Before and After Gargling Using Rosella Tea Infusum and Mineral Water

Treatment	Rosella Tea	Green Tea	Mineral Water
Mean±SD	34.90 ± 9.07	14.56 ± 5.37	4.81 ± 0.82

C. HOMOGENEITY TEST OF VARIABLE DESCRIPTION

The homogeneity distribution test is a test to measure whether the data obtained has a homogeneous distribution so that it can be used in parametric statistics (inferential statistics), in other words homogeneous tests are tests to find out whether the empirical data obtained from the field is distributed homogeneous (having the same variance), the data is said to be homogeneous if the significance column is less than 0.05, while the results include homogeneous data with a very good score, which is $0.002 < 0.05$.

Table 7: Distribution of homogeneity reduction of microorganism colonies before and after gargling using rosella tea infusum and mineral water

Levene Statistic	df1	df2	Sig.
8.084	2	27	.002

D. EFFECTIVITY TEST OF VARIABLE DESCRIPTION

Test data distribution is a test to determine whether the data found to have significance based on the research hypothesis, the data is said to be effective if the ANOVA test table the average distribution in the significance column is smaller than 0.05. This study used ANOVA test for the data, as for the results of 0.000 and in the advanced test table using the Tukey HSD test the distribution of data in the table shows the results of the average score distribution for each variable in different columns and rows which can be seen in table 8.

Table 8: Difference test of 3 groups with colonies microorganism reduction before and after gargling using rosella tea infusum and mineral water

ANOVA					
Average distribution					
	Sum of Square	df	Mean Square	F	Sig.
Between groups	4713.954	2	2356.977	63.159	.000
Within groups	1007.593	27	37.318	-	-
Total	5721.547	29	-	-	-

Table 9: Further test of microorganism colonies reduction before and after gargling using rosella tea infusum and mineral water

Average Distribution					
	Variable Group	N	Subset for alpha = 0.05		
			1	2	3
Tukey HSD ^a	Mineral water	10	4.8100	-	-
	Green tea	10	-	14.5600	-
	Rosella tea	10	-	-	34.9000
	Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed

a. Uses Harmonic Mean Sample Size = 10.000

IV. DISCUSSIONS

The results show manually processed data based on tables 1 and 2, and in tables 3 through table 7 which are processed and analysed using software One Way Anova where respondents received treatment in the form of mouth rinse interventions with 25% rosella tea infusum, 25% green tea infusum and mineral water in the form of control have a reduction in the number of colonies in the calculation through invitro microorganism culture using isolation method pour plate with saliva samples before and after 25% rosella tea infusion, 25% green tea infusion and control water.



Manually processed results showed that rosella tea infusum with a concentration of 25% had the ability to be more effective in avoiding microbial culture mechanisms by looking at the processed data, namely 25% rosella tea infusum was able to inhibit / reduce the number of colonies before and after intervention with an average of 34.9 %, while green tea infusum averages 14.56%, while mineral water averages 4.81%. While the One Way Anova statistical test and the *Tukey HSD post hoc test* showed very meaningful and varied data as can be seen in table 9., namely the normality test, the average percentage reduction before and after intervention was very clear the effectiveness of rosella tea infusion 25% compared to 25% infusum of green tea and mineral water, although in terms of taste and aroma of infusion of rosella tea has an acidic taste that is quite pronounced, and the aroma is rather stinging, it is caused by a considerable amount of citric acid. According to [2], minimum rosella flower extract of 0.20g / ml against *Escherichia coli*, *Salmonella typhi* and *Staphylococcus aureus*, this is due to the content of alkaloids, flavonoid, saponins and tannins, of which the active substances have activities as antibacterial.

The results above relate to gargling activities in both the intervention group and the control group, each with a reduction / decrease in the number of cultures of microorganism colonies in saliva from each sample, although the degree of reduction varies greatly, it is related to the function or rinsing function that is one way to get germs or other substances in our mouth when gargling.

Regarding the reduction of significant microorganism colonies in respondents with rosella tea mouth rinses 25% is strongly influenced by the content of active substances which have the ability of bacterised and bacteriostatic which can interfere with the growth of microbial activity, on roselle petals containing flavonoid and polyphenols, citric acid, saponins, tannins that function to inhibit the growth of microorganisms, because they are able to form complex compounds with proteins through hydrogen bonds[6].

Gargling with green tea infusion 25% can also inhibit the growth of microorganisms, although the results are not as effective as 25% rosella tea rinse, from the results of data studies from respondents who gargled with green tea infusum experienced a decrease / reduction in the average number of colonies before and after gargling which amounted to 14.56%, the ability of green tea infusum 25% to reduce the number of microorganism colonies to saliva by gargling. Green tea is believed to be able to reduce the formation of dental plaque with two mechanisms, namely making bacteria that cause the formation of plaques such as *Streptococcus mutans* and glycotransferase of bacteria (Kelwandi 2012). It was how about the opinion of [9] that green tea contains 30-40% polyphenols, most of which are known as polyphenol, which are strong antioxidants, stronger than vitamins E, C and beta-carotene, besides green tea also contains alkaloids and minerals that are beneficial for health.

V. CONCLUSION

The number of microorganism colonies before rosella tea infusion gargle is 25%, which is an average of 29.92 colonies. After gargling a total of 18.56 colonies, there was a difference of 4.42 colonies resulting in a reduction of 35.5% before and after.

The colonies of microorganisms before 25% of green tea infusion i.e. an average of 69.8 colonies, after gargling a number of 59.52 colonies, there is a difference of 10.26 colonies so that the results of reduction of colonies of microorganisms before and after amounted to 14.8%

The number of colonies of microorganisms before rinsing mineral water as control is an average number of 93.68 colonies, after gargling a total of 89.24 colonies, there is a difference of 4.42 colonies resulting in a reduction in colonies of microorganisms before and after as large as 4.8%.

Using 25% infusion of rosella tea is more effective for gargling mouth and teeth microbial culture compared to 25% green tea infusion and mineral water.

VI. THANKS

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Yodong is the sixth daughter of seven brothers from the late Mustafa and Hj. Tanawali. Born in Malimpung Village, South Sulawesi Province on December 9, 1965. Current address is Villa Durian Kav 34 Jalan Durian Raya, Banyumanik District, Semarang City, Central Java Province. Graduated from SDN 12 Pinrang in 1977. Continuing to SMU 1 Pinrang, graduated in 1981. Continuing to the Ujung Pandang Department of Dental Care (SPRG) School of Health, graduated in 1984. Continuing Diploma III to the Dental Nursing Academy (AKG) of the Department of Health Makassar, graduated in 2003. Continuing Diploma IV to Dental Polytechnic Department of Nursing in Semarang, graduated in 2009. Continuing with Master of Health Law in Unika Soegjapranata

Semarang, graduated in 2012. In 2010, she received the award of "Bakti Husada Tri Windu Work" by the Minister of Health. conducted a study titled Irene's Donut's Method of Improving Knowledge, Attitudes and Practices of Parents Against Hygiene Teeth and Mouth Levels in 2011, conducted a study entitled Evaluation of Service Implementation of School Dental Health Program (UKGS) in Semarang District in 2011, conducting research entitled Turmeric Concentration (Curcuma Domestica val) In Mouthwash To Cure Gingivitis in 2012, Conducted a Research Entitled Old Use of Mouthwash Preparation from Betel Leaves (Piper Betle L.) Extract with Turmeric (Curcuma Domestica Val.) On Changing Plaque Index and Gingivitis Healing in 2012, conducted a study entitled The Effect of Red Betel Leaf Decoction (Piper Erotum) on Staphylococcus Aureus Bacterial Growth in 2014, and conducted a study entitled Effectiveness of Drinking Sweet Starfruit Juice and Guava Juice with and without Sugar on Ph Saliva in Sema First Semester Dental Nursing Students rang in 2017.



Surati was born in Semarang Regency, Indonesia, in 1975. She received her bachelor degree of chemistry in Diponegoro University, Indonesia in 2000 and her Master of Bio medic in the same university in 2012. Her current research interests include engineering in healthscience. She has done research lately about Effects Extract Skin Kaffir Lime (Citrus hystrix DC) on the morphology and histology Larva Aedes Aegypti. She also has done a research involving electrical engineering in health entitled Android based mosquito monitoring model for Dengue Hemorrhagic Fever Control in 2018. Not to mention, she managed to put her dedication to a research of Receptivity of vetiver (vetiveriazanoides) Antiseptic tooth paste in various concentration as well as Levels of Infused

Beverage Safety Water with Different Storage Diversification in 2017. Besides, it was also done a research about Relationship between Sanitation Hygiene and Occurrence of Soil Transmitted Helminths Infection in Scavengers at Jatibarang Polling Station.



ErniMardiati was born in Semarang on March 28, 1967. Born and raised at Jl. Puspowarno Tengah IX no. 38 Rt. 2 RW 6, Gisik Drono Village, West Semarang District, Semarang city. The sixth child of seven siblings from Mr. Djoemadi and Mrs. Sri Sulastri, Graduated from Kindergarten in 1976. Continued Elementary School at the Semarang IKIP Development School (PPSP), Graduated in 1981. Then continued her junior high school education at SMP N 165 East Jakarta, graduated in 1983/84, then continued in the dental care school in 1983/84 in Semarang and graduated in 1988. In 2003 she went on to study at the Semarang Dental Health Academy (AKG) graduating in 2006, then in 2007 she continued her undergraduate study at FKM (Public Health Faculty) of Diponegoro



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Tri Anonim was born in Batang, Central Java on July 3, 1965. Born and raised at Jln RE. Martadinata No. 288 Karangasem Selatan Sub-District, Batang District, Batang Regency. The ninth child of nine siblings, from Mr. Casmudri and Ms. Kasturi. Education that had ever been taken, SDN Karangasem II Batang graduated in 1977, SMP 1 Batang, graduated in 1981, SMA NIBatang, graduated in 1984, then continued to tertiary education at the Department of Health in Semarang, graduating in 1988, D IV Nurse Educator Diponegoro University graduated in 1999 and continued in the MIKM Diponegoro University postgraduate program graduating in 2012. Conducted research entitled Effect of LDL and Diabetes Mellitus on Preeclampsia Pregnancy in 2017 as chairman and Effect of Psychoeducation Therapy on Anxiety Levels of Breast Cancer Chemotherapy Patients in

2018 as member.