The Resistance of Bandotan \textit{(Ageratum Conyzoides)} Leaf Extract and Siwak Stem Extract on the Growth of Buttermilk Streptococcus Mutans

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Abstract. Streptococcus mutans is a known bacterium that can cause dental caries and is a serious problem in the oral cavity. The purpose of this study was to determine the inhibition of Bandotan leaf extract and siwak stem powder extract on the growth of the S mutans bacteria. This research used the descriptive method, with the design of this study was experimental. The research subject used was the bacteria S mutans, the design in this study was by making a solution of Bandotan \textit{(Ageratum Conyzoides)} leaf extract and siwak stem powder with a concentration of 100%, 75%, 50%, 25% and 10% and 4 times repetition, then made suspense bacteria S mutans i.e. bacteria grown on PCA media were given a paper disk that had previously been dipped in a Bandotan \textit{(Ageratum Conyzoides)} leaf extract solution and siwak stem powder, then incubated for 1x24 hours in the incubator, the results were observed and measured (posttest) in the oligodynamic region of the inhibitory area diameter growth of bacterial mutans by using calipers (mm). The data obtained were analyzed using quantitative descriptive analysis methods and then performed iji annova. The results showed that both extracts were able to inhibit the growth of the bacteria S mutans, although it has a very weak inhibitory power that is an average of 4x experiments is 9.32 mm and it is hoped that this research can be developed again with other methods so that the results obtained are optimal.

1. Introduction
Dental and oral health is part of general health. Dental and oral health need to be maintained properly because a person's activities can be disrupted if dental and mouth health is disrupted. One of the bacteria known as mutans is a bacterium that is considered to have a role in the disruption of dental health. Siwak group can be an alternative to be a bacterial inhibitor because physiological sodium there is a salt content that can be used as an anti-microbial at low salt water concentrations will increase bacterial growth. Conversely, salt in pure form with high concentrations can reverse because there is no sodium chloride in it which is a halogen group. Halogens have strong oxidizing properties and are able to fight bacteria [1]. Other ingredients in the siwak stem are also very good.

The purpose of this study was to determine the inhibition of Bandotan \textit{(Ageratum Conyzoides)} leaf extract and siwak stem powder extract on the growth of the S mutans bacteria. The siwak group is known and recognized by the world as a good antiseptic in the oral cavity. [2]
2. Research Method

The research method used was experimental. This research was conducted to obtain an objective picture of a situation where the cause or risk and consequence variables or cases occurring on the research object would be measured or collected at the same time. In this study, the condition of the bacteria S. mutans was investigated after being given Bandotan (Ageratum Conyzoides) leaf extract. The research plan carried out was an experiment, an observation without a control group and could be described in the model design as follows:

Experiment | Posttest
--- | ---
Group of Bandotan (X1) | O-1
Group of siwak (X2) | O-2

Notes:

(X1) : Provision of Bandotan (Ageratum Conyzoides) leaf extract
O-1 : Observation and measurement of the inhibitory area
(X2) : Provision of Siwak Stem extract
O-2 : Observation and measurement of the inhibitory area

The sample in this study was 1 ml of S. mutans bacteria which was inserted into a Petri dish to obtain bacterial suspense media. The suspension was grown on PCA media on a petri dish which was then placed filter paper on the media. In this study, the data obtained from the results of measurements and calculations are made in tabular form in the form of tables then made in the form of percentages. In analyzing the data, this study uses a quantitative analysis method that is to get a picture of the inhibitory properties of the extract on the growth of the bacterial mutants. Anova test was then performed to test the difference in mean (average) data of more than two groups, namely the analysis of variance of one factor (one-way anova). Some assumptions that must be met in the Anova test are:

1. Samples come from independent groups
2. Variants between groups must be homogeneous
3. Data for each group is normally distributed

The first assumption must be met when random sampling of several (> 2) independent groups, where the value in one group does not depend on the value in the other group. While the fulfillment of the second and third assumptions can be checked if the data has been entered into the computer, if this assumption is not met, transformation can be made on the data. If the transformation process cannot meet this assumption then the Anova test is invalid, so it must use a non-parametric test such as Kruskal Wallis.

3. Result and Discussion

An illustration of the inhibition of Bandotan (Ageratum Conyzoides) leaf extract on the growth of the bacteria S. mutans has been carried out in the Tasikmalaya BTH Analyst laboratory. The research results can be seen in the following table:

| Concentration | UL I | UL II | UL III | UL IV | UL I | UL II | UL III | UL IV |
|---------------|------|-------|--------|-------|------|-------|--------|-------|
| 100           | 13,00| 11,00 | 13,00  | 11,00 | 20,20| 23,00 | 23,00  | 20,00 |
| 75            | 10,40| 9,60  | 13,00  | 9,80  | 20,00| 19,40 | 19,50  | 19,20 |
| 50            | 9,00 | 9,00  | 8,50   | 8,00  | 16,00| 18,00 | 18,00  | 18,20 |
| 25            | 9,00 | 8,00  | 9,4    | 9,50  | 14,00| 14,00 | 15,00  | 13,70 |
| 10            | 9,00 | 9,00  | 8,30   | 8,00  | 9,60 | 9,50  | 8,50   | 9,40  |
| Average       | 10,08| 9,32  | 10,44  | 9,26  | 15,96| 16,78 | 16,8   | 16,1  |

Table 1. The Total Average inhibition of Bandotan (Ageratum Conyzoides) leaf extract and Siwak stem extract against S. Mutans

| Concentration | UL I | UL II | UL III | UL IV | UL I | UL II | UL III | UL IV |
|---------------|------|-------|--------|-------|------|-------|--------|-------|
| 100           | 13,00| 11,00 | 13,00  | 11,00 | 20,20| 23,00 | 23,00  | 20,00 |
| 75            | 10,40| 9,60  | 13,00  | 9,80  | 20,00| 19,40 | 19,50  | 19,20 |
| 50            | 9,00 | 9,00  | 8,50   | 8,00  | 16,00| 18,00 | 18,00  | 18,20 |
| 25            | 9,00 | 8,00  | 9,4    | 9,50  | 14,00| 14,00 | 15,00  | 13,70 |
| 10            | 9,00 | 9,00  | 8,30   | 8,00  | 9,60 | 9,50  | 8,50   | 9,40  |
| Average       | 10,08| 9,32  | 10,44  | 9,26  | 15,96| 16,78 | 16,8   | 16,1  |

Total r 9,96 16,41
Based on Table 1 above it can be explained that the average total inhibition of Bandotan (*Ageratum Conyzoides*) leaf extracts against bacterial growth *s. mutans* mounted to 9.96 mm weak weak category, whereas the average total inhibition of Siwak stems extracts against bacterial growth *s. mutans* is 16.41 mm in the medium category.

It can be seen that the average of siwak extract is stronger than Bandotan (*Ageratum Conyzoides*), but to test the difference statistically, anova test is used, such as the results below.

**Table 2.** Bandotan and siwak test against *s. mutan* (Oneway-Anova)

|                   | Sum Of Squares | df | Mean Square | F     | Sig.  |
|-------------------|----------------|----|-------------|-------|-------|
| Between Groups    | 299.331        | 4  | 74.833      | 4.512 | .005  |
| Within Groups     | 580.476        | 35 | 16.585      |       |       |
| Total             | 879.808        | 39 |             |       |       |

In the different test, the two groups showed the results of p = <0.05, meaning that there were significant differences between the Bandotan (*Ageratum Conyzoides*) leaves and the siwak stem groups.

**Table 3.** From the results of further analysis

| Konsentrasi Persen | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval |
|---------------------|-----------------------|------------|------|-------------------------|
| 10.00               | -2.6625               | .688       | -8.5168 | 3,1918                 |
| 50.00               | -4.1750               | .264       | -10.0293 | 1,6793                |
| 75.00               | -6.2090*              | .033       | -12.0543 | -3,457                 |
| 100.00              | -7.8625*              | .004       | -13,7168 | -2,0082                |
| 25.00               | 2.6625                | .688       | -3,1918 | 8,5168                 |
| 50.00               | -1.5125               | .945       | -7.3668 | 4,3418                 |
| 75.00               | -3.5375               | .425       | -9.3918 | 2,3168                 |
| 100.00              | 5.0000*               | .102       | -11,0543 | 6,5434                |
| 50.00               | 4.1750                | .264       | -1.6793 | 10,0293                |
| 25.00               | 1.5125                | .945       | -4,3418 | 7,3668                 |
| 75.00               | -2.0250               | .856       | -7.8793 | 3,8293                 |
| 100.00              | 3.6875                | .384       | -9.5418 | 2,1668                 |
| 75.00               | 6.2090*               | .033       | 3,457   | 12,0543                |
| 25.00               | 3.5375                | .425       | -2,3168 | 9,3918                 |
| 50.00               | 2.0250                | .856       | -3,8293 | 7,8793                 |
| 100.00              | 1.6625                | .724       | -3,6745 | 4,9198                 |

The best results are in the levels of 100% (0.004), 75% (0.033) while at other concentrations the results of the analysis show the two groups on average do not show any homogeneous differences. Shown in the results below

**Table 4.** From the results of any homogeneous differences

| Konsentrasi Persen | Subset for Alpha=.05 |
|--------------------|-----------------------|
|                    | N  | 1  | 2  |
| Tukey HSD          |    |    |    |
| 10.00              | 8  | 8,9125 |
| 25.00              | 8  | 11,5750 | 11,5750 |
| 50.00              | 8  | 13,0875 | 13,0875 |
Both groups did not show any significantly different concentration distribution, Bandotan (Ageratum Conyzoides) leaves 0.264 and siwak stems 0.102. Based on the results of research conducted at the Microbiology Laboratory to determine the effect of both extracts on bacterial growth s. mutans indicates that there are or are their effects of both types of extract material on bacterial growth. mutans. Streptococcus mutans is usually found in the cavities of injured human teeth and become the most conducive bacteria to cause caries in tooth enamel [1]. This study shows the chemical content of the Bandotan (Ageratum Conyzoides) leaves, namely linoleic acid and essential fatty acids, alkaloids, saponins, and tannins. But which can inhibit bacteria are tannins and saponins. Tannins have antibacterial activity, an outline of the mechanism that is estimated is the toxicity of tannins that can damage the bacterial cell membrane, astringent tannin compounds can induce the formation of tannin bonding complexes to metal ions which can increase the toxicity of the tannin itself [3]. Saponins have good antibacterial and antifungal effects, antifungal and antibacterial effects are disturbed by the presence of monosaccharide groups and their derivatives [4]. Maceration extract using 70% ethanol solvent then soaked for 24 hours to dissolve the substances in the Bandotan (Ageratum Conyzoides) leaves. According to Greenwood in Pratama [5]. Classification of bacterial growth inhibition responses that have a bright zone diameter of more than 20 mm, the growth inhibition response is categorized as strong, the bright zone diameter of 16-20 mm is categorized as medium, the bright zone diameter of 10-15 mm is considered weak, the diameter of the bright zone is less than 10 mm weak, 0 mm bright zone diameter categorized as no inhibitory response to bacterial growth. In the Bandotan (Ageratum Conyzoides) leaf extract has an average bacterial inhibition zone of 9.96 mm, then for the Bandotan leaf has a bacterial growth inhibition response in the weak category and for the Siwak stem, group is 16.41 mm still in the medium category. But as a local content as a plant that grows around us the inhibition of bacteria is better to use Bandotan (Ageratum Conyzoides) leaf extract even with a difference of 40% antibacterial. The low value of the average inhibition of Bandotan (Ageratum Conyzoides) leaves against bacteria S mutans shows that bacteria are less sensitive to the test material.

The small inhibitory zone is likely influenced by the quality of the Bandotan (Ageratum Conyzoides) leaf extract. In this study the levels contained in the Bandotan (Ageratum Conyzoides) leaf extract could not be determined yet, so the small inhibitory value could be due to the active substance saponin and tannin contained in the Bandotan leaf extract was not insufficient levels to cause an antibacterial effect. Extract quality is influenced by 2 factors: biological and chemical factors. Biological factors include plant species, time of harvest, and parts of plants used. The second factor is chemical factors including external and internal factors. External factors include heavy metal content and pesticides in plants. Internal factors that affect extract quality include the type, quantitative composition and average levels of active compounds contained in the Bandotan (Ageratum Conyzoides) leaves.

The technical research on the implementation cannot be taken into account which includes the cleanliness of the equipment, the unsterile room, the room temperature, the incubation time, the PH of the media and the lack of knowledge of the researchers when conducting research, can also affect the results of this study. The results obtained indicate that the average results of bacterial inhibition zone measurement changes or are not constant, this is due to technical factors that occur at the time of the study, namely the amount of Bandotan (Ageratum Conyzoides) leaf extract that is absorbed in the paper disk experience differences and affects the inhibitory power. Based on research that has been done shows that s.mutans is sensitive to the test material. This can be observed from the inhibition zone seen in each treatment having different inhibitory even though the difference is only slightly.
4. Conclusion

- Bandotan (*Ageratum Conyzoides*) leaf extract can inhibit the growth of the bacteria s.mutans with a weak category.
- Bandotan (*Ageratum Conyzoides*) leaf extract has an average bacterial area free of mutants that is equal to 9.96 mm.
- As for the siwak stem, the group is 16.41 mm, the medium category.

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