

## The effects of rinsing red beet root (*Beta vulgaris* L.) juice on streptococcus sp. dental plaque



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### Abstract

**Objective:** The aim of this study was examine the effectiveness of rinsing red beetroot juice to streptococcus sp. on dental plaque.

**Material and Methods:** Antiplaque have side effects, so it is necessary to do research of alternative antiplaque materials. One of them is the use of red beet root because it contains some antibacterial substances.

**Results:** Analysis of One-Way ANOVA have significance 0.000, which means it decreased the number of streptococcus sp.

**Conclusion:** Rinsing red beet root juice is effective to decrease the number of streptococcus sp. on dental plaque.

**Keywords:** Red beet root, Streptococcus sp, Dental plaque

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### Introduction

Dental plaque is a biofilm contains of bacteria and firmly attach to tooth surface, restoration and prosthesis.<sup>1</sup> The early process of plaque formation is preceded by pellicle formed 1 minute after dental cleaning. The formation of pellicle is started by bacteria attachment dominated by streptococcus sp colony, continued by the attachment of the other microorganisms.<sup>2</sup> One of the many ways to eliminate plaque and obstruct the forming of plaque is doing plaque control. Mechanical plaque control can be done by brushing teeth and using dental floss.<sup>3</sup> Chemical plaque control can be done with gargling antimicrobial solution. One of the antimicrobial materials is mouthwash.<sup>4</sup> Plaque control only with mechanical way cannot give the best result, that is why another way is needed to control plaque, that is by the chemically way.<sup>5</sup>

Mouthwash is one of the solutions or liquid used for giving a freshness to oral cavity and cleaning it from plaque and organisms which can cause an illness in oral cavity. A mouthwash that is usually used and studied is chlorhexidine gluconate. Chlorhexidine gluconate is a large spectrum antibacterial.<sup>6</sup> Chlorhexidine helps plaque clearance optimally but chlorhexidine has negative effects such as can cause stain in teeth, tongue and dentist materials, changing the sensation of taste, burn feeling in oral mucosal and a epithelial desquamation reaction.<sup>7</sup> The negative effects of mechanical contained in mouthwash encourage the researcher to do a research about alternative material which meets the criteria of safer antiplaque materials, one of them is red beet root. Red beet root is a safe material because it is usually

consumed as a vegetable and has lots of benefits for human body.<sup>8,9</sup>

Red beet root has antibacterial activity towards gram positive and gram negative bacteria. The highest antibacterial activity in red beet is towards gram positive bacteria. Beet has an antibacterial activity because it contains phenol. The other content of red beet antibacterial are flavonoids, tannins and saponins.<sup>10</sup>

### Material and Methods

This quasi experimental study used post-test only control group design. This study was carried out in Laboratory of Microbiology Faculty of Dentistry, Laboratory of Microbiology and Laboratory of Botany Faculty of Mathematics and Natural Science University of Jember, Jember, Indonesia. This study passed ethical approval by Medical Research Ethic Committee of University of Jember as stated in the letter No.1072/H25.1.11/KE/2016.

The subjects were 27 students of Dentistry Faculty University of Jember, Jember, Indonesia chosen by purposive sampling. The subjects were divided into 3 groups, group 1 was rinsing with aquadest, group 2 was rinsing with chlorhexidine gluconate 0.2% and group 3 was rinsing with red beet root juice. All subjects were given the instruction of how to rinse, scaling was done one week before the research, instructed to use the same technique of brushing their teeth which was roll technique, asked not to use any mouthwash during study, instructed not to eat and drink one hour before the study and did a standard-ize which was rinsing using aquadest for each group.

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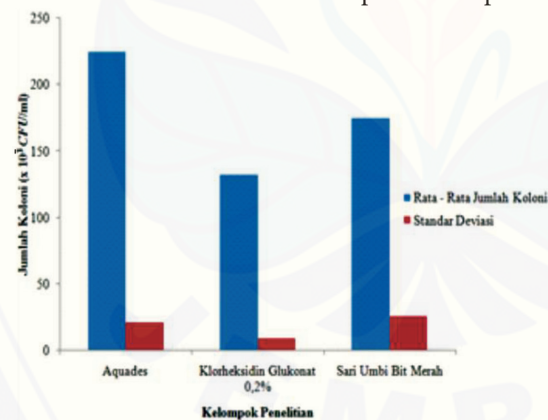
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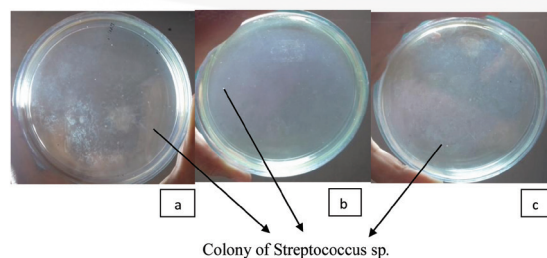
The study done in the Laboratory of Microbiology Faculty of Dentistry University of Jember, Jember, Indonesia at 09.30-10.00 WIB. The time was chosen to decrease the negative impact from circadian rhythm to the flow of saliva.<sup>11</sup> The first day group 1 was rinsing with aquadest, second day the group 2 was rinsing with chlorhexidine gluconate 0.2% and the third day group 3 was rinsing with red beet root juice. Each subjects rinsing about 15 ml solution in 30 seconds. After that, it was discarded and waited for 5 minutes and continued with plaque sampling on the buccal surface of the maxillary first molar tooth about 3 times using excavator from mesial to distal, then it went inside of a test tube containing of 2 ml PZ solution. Then, the plaque sample diluted in  $10^{-3}$  dilution. After diluted, the plaque sample planted in streptococcus jelly medium using pour plate method, then incubated for 24 hours at 37°C. The inoculation result of streptococcus sp were counted using a colony counter.

## Results

Study results the number of colonies of streptococcus sp. as much as  $174.7 \times 10^3$  CFU/ml, rinsing group used chlorhexidine gluconate 0.2% have a number of colonies of streptococcus sp.



**Figure 1** Histogram of the average number of streptococcus sp. in each group



**Figure 2** The results of inoculation of streptococcus sp. in rinsing group A. Aquadest, B. Chlorhexidine gluconate 0.2%, C. Extract of beet root

as much as  $132.1 \times 10^3$  CFU/ml and the number of colonies group that rinsing with aquadest was  $224.4 \times 10^3$  CFU/ml. The average results on the number of colonies of streptococcus sp. each group obtained histogram that can be seen in [figure 1](#).

The fewest number of streptococcus sp. colonies were the rinsing group with chlorhexidine gluconate 0.2% [figure 2b](#), followed by rinsing group used the extract of red beet root [figure 2c](#) and the number of colonies, mostly were in groups rinse aquadest [figure 2a](#). It can be interpreted that the most decreased number in colony of streptococcus sp. was occurred in the group gargling with chlorhexidine gluconate 0.2%, followed by gargling group was the extract of red beet bulb and the latter were group aquadest rinse.

The results of One-Way ANOVA test was obtained significance value of 0 which means less than 0.05. Based on the significant value it can be interpreted that there are differences in the number of colonies of streptococcus sp. significance due to material different mouthwash. Test followed by LSD (Least Significance Different) determined significant difference in all groups.

## Discussion

The results showed that the decrease in the number of colonies of streptococcus sp. In other hand the largest plaque occurred in the positive control group was the group rinsed with chlorhexidine gluconate 0.2% group as much as  $132.1 \times 10^3$  CFU/ml, followed by a group of red beet root juice as much as  $174.7 \times 10^3$  CFU/ml and the number of colonies by aquadest group was  $224.4 \times 10^3$  CFU/ml.

Positive control in this study was 0.2% of chlorhexidine gluconate having the ability to reduce the number of colonies of streptococcus sp. in plaque. Chlorhexidine is a mouthwash that has a broad spectrum against positive and negative gram bacteria. Chlorhexidine has bactericidal characteristic with strong binding mechanism to the bacterial cell membrane, increasing cell permeability, thus starting bacterial leakage of intracellular components.<sup>6</sup> Chlorhexidine resulted in coagulation and precipitation of cytoplasm with the formation of phosphate complexes that include adenosine triphosphate (ATP) and nucleic acids that resulted in cell death. The antimicrobial effects of chlorhexidine is caused by presence of absorption dicationic molecules to the surface of the oral cavity, email, or pellicle.<sup>12,13</sup>

Chlorhexidine has a big antibacterial characteristic. Further, chlorhexidine can help the removal of plaque optimally, however it also has side effects, such as stain in teeth, tongue and materials-dental,



can cause changing in the sensation of taste, burning sensation in the oral mucosa and the occurrence of epithelial desquamation reaction.<sup>7</sup> The use of chlorhexidine gluconate 0.2% in the study resulted in the loss of sensation shortly after gargling by using chlorhexidine gluconate 0.2%.

Red beet root juice extract in this study can reduce the number of streptococcus sp. in plaque. The red beet root are known contain a number of antimicrobial compounds such as phenols, flavonoids, alkaloids, tannins and saponins.<sup>9</sup> The content of the antibacterial in the red beet root is assumed to decrease the number of colonies of streptococcus sp. in plaque. Phenol in red beet roots have high antimicrobial activity. The mechanism of phenol as an antibacterial giving an effect on the bacterial cell membrane, changing the cell membrane and changing the structure of the membrane, thereby increasing the permeability of the cytoplasmic membrane. The increased of permeability of the cytoplasmic membrane resulted in the loss of cellular pH gradient, decreased levels of ATP and the loss of the proton motiveforce, which causes cell death.<sup>10</sup>

The mechanism of flavonoids as antimicrobial is to create a bond with phospholipids in the cell membrane of bacteria by reducing the permeability of the membrane, hence the cells become lysis and cause denaturation of proteins, inhibiting the formation of cytoplasmic proteins, nucleic acids and bonding with ATP-ase in the cell. The damage from cell membrane results in leakage of critical components such as proteins, nucleic acids and nucleotides which are the result of cell permeability disturbance so the cells unable do the activities of life and stunted growth or even death.<sup>14</sup>

The red beet root contains tannins that has antibacterial properties. The mechanism of tannins antibacterial is through reaction with the cell membrane, inactivation of the enzyme and inactivation of the function of the genetic material. The mechanism of tannins as antibacterial agent are inhibiting the enzyme reverse transcriptase and DNA topoisomerase so bacterial cells cannot be formed. The power of tuber red beet antibacterial is saponins. The mechanism of saponins as an antibacterial which can cause leakage of proteins and enzymes from cells. Saponins can be used as an anti-bacterial because its surface as an active substances like detergents, consequently saponins will reduce the surface tension of the wall of bacterial cell and damage membrane permeability.<sup>15</sup>

## Conclusion

The conclusions of this study is rinsing with red beet root juice effectively decreases the number of colonies of streptococcus sp. in dental plaque. The content of phenols, flavonoids, tannins and saponins in red beet root suspected of having a function as an antibacterial.

## Conflict of Interest

The authors report no conflict of interest.

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