

COMPARISON THE EFFECT OF PROBIOTIC CHEWING GUM AND PROBIOTIC YOGURT IN REDUCING CARIES RISK OF CHILDREN

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ABSTRACT

Various methods to control caries risk in the mouth have been studied. one of that by consuming probiotics. Probiotics contain of anti-bacterial *reuterin* which can inhibit the growth of cariogenic (antibacterial) bacteria so that it can increase the pH of saliva. This study was an experimental study with a prospective comparative interventional design. The aims of the study was to compare the effectiveness of probiotic chewing gum and probiotic yogurt drink in reducing caries risk of children, by measuring the average of salivary flow rate, salivary pH, and salivary buffer capacity before and after the treatment of drinking probiotic yogurt and probiotic chewing gum. A total of 60 students aged 10-12 years old at primary school of 060925 Harjosari Medan were divided into three groups, consist of group I was the control group, group II was the probiotic chewing gum group, and group III was the probiotic yogurt group. The intervention was carried out for 7 days. The normality test was carried out using the Kolmogorov Smirnov test. The data is not normally distributed, so the analysis uses the *Kruskal wallis test*, to compare salivary pH, salivary flow rate, and salivary buffering capacity between the three groups.. The results showed that there was a very significant difference in salivary pH, buffer capacity, and salivary flow rate before and after the intervention between the control group and the intervention group ($p < 0.05$). The difference in mean of the probiotic yogurt group was greater in pH saliva (1.17 ± 0.58) and buffer capacity (1.36 ± 0.44) while the average difference between the chewing gum group was higher for the salivary flow rate parameter (1.53 ± 0.63). based of Pos Hoc Bonferoni Test. consuming probiotic yogurt was stronger in increasing saliva buffer capacity than probiotic chewing gum, but chewing gum was stronger in increasing the flow rate of saliva than drinking probiotic yogurt. In increasing pH saliva both drinking yogurt and chewing gum have an equally great effectiveness. It can be concluded that consuming probiotic yogurt was more influential in reducing the risk of dental caries than probiotic chewing gum.

Keywords: Probiotic Yogurt, Probiotic Chewing Gum, dental caries risk

INTRODUCTION

Dental caries was still the main problem of dental and oral health problems in the world (Selwitz et al., 2007). Dental caries was chronically progressive because if it is not treated, it will get worse (Tjahja et al, 2002). The prevalence of dental caries severity in Indonesia were still relatively high. Based on basic health survey data of Indonesia In 2018, the prevalence of caries in children aged 10-14 years was 1,8 (73,4%), an increase compared to the 2007 data, which was 43.4% and in 2013 it was 53.2%. As many as 55.6% of Indonesian people aged 10-14 years have problems with their teeth and mouth, and only 9.4% of them have received treatment from dental medical treatment. In the province of North Sumatra, as many as 54.6% of the total population have dental and oral problems, and only 6.7% have received dental medical treatment.

The causes of dental caries in children were consume snacks such as sweet foods, cereal bars, and biscuits, in addition to cariogenic microorganisms in the mouth, and *host* factors, such as the level of salivary secretion and the pH and buffering capacity of saliva (Cagetti et al., 2013).). The leftover food or drink can later form plaque which will affect the pH of saliva and can cause dental and oral disease (Maranatha, 2013).

Various methods to control the risk of caries in the mouth have been studied, one of them was by consuming probiotics. The rationale for the use of probiotics in the maintenance of oral health comes from medical research, where probiotic bacteria can provide health benefits by reducing infections in the gastrointestinal tract including in the oral cavity (Fejerskov et.al., 2015). According to Maqassary (2015), an alternative way to deal with an overly acidic condition in the mouth is by consuming foods that contain probiotics.

In dentistry, the use of probiotic methods aims to replace pathogenic bacteria in the oral cavity and strengthen the natural microflora that causes major oral diseases such as caries, periodontitis, oral candida, and halitosis (Cagetti et al., 2013). Recent clinical studies of probiotics with *Lactobacillus rhamnosus* and *Lactobacillus reuteri* strains have shown that regular intake of probiotics can result in a reduction in the number of *S. mutans* in saliva and plaque (Nikawa et al., 2004). Types of probiotics that are well known to the public are yogurt and chewing gum, which are fermented using *Lactobacillus delbrueckii* sub sp. *bulgaricus* and *Streptococcus salivarius* sub sp. *thermophilus* (Sudhir et al., 2012). The probiotic contains anti-bacterial *reuterin* which can inhibit the growth of cariogenic (antibacterial) bacteria so that it is thought to increase the pH of saliva (Bonifait et al., 2009). Stamatova and Meurman (2009) added that yogurt has antibacterial properties because it contains lactic acid bacteria, hydrogen peroxide, and bacteriocin which can inhibit the increase in the number of pathogens in the mouth. The inhibitory component or production of lactic acid bacteria, bacteriocin, related to the number of pathogens is very important in the general infection defense mechanism.

Research on probiotic yogurt in the field of oral and dental health has been carried out, including the study of Winarmi (2014) which showed that there was a significant difference in the number of *S. mutans* colonies in children's dental plaque after consuming yogurt for 7 days. The study of Javid et al. (2015) showed that the average number of *S. mutans* before consuming yogurt was 2.624 ± 137.73 and after consuming yogurt was 621 ± 69.99 . The study of Oinike et al., (2018) reported that probiotic chewing gum was effective in increasing the pH and salivary flow rate in fifth-grade elementary school children, while in research of Himawan et al., (2018) added that the consumption of probiotic gum can reduce the dental plaque index value salivary *S. Mutans* colony in fifth-grade elementary school students in Ungaran Semarang. Based on this background, the researchers wanted to compare the effectiveness of probiotic chewing gum and probiotic yogurt drink in reducing the risk of caries in children aged 10-12 years, and this case study was at SD Negeri 060925 Harjosari of Medan city.

METHOD

1. Design study

This research was an experimental study with a *prospective comparative interventional* design (Kaliemoorthy et al, 2018). A total of 60 subjects were divided into 3 groups, namely the chewing gum treatment group, the Yogurt drink treatment group, and the control group (not consuming probiotic drinks/candy) with 20 people in each group.

2. Inclusion and exclusion criteria

The inclusion criteria of the sample were not sick, cooperative, the arrangement of the teeth was not crowded or the teeth were lightly crowding and the maximum number of *decay* (tooth cavities) was 4 teeth, while the exclusion criteria were not having tartar or dental plaque with mild criteria, consuming Other types of chewing gum 1 hour before the examination, consuming flavored food and drinks 1 hour before the examination and using mouthwash 1 hour before the examination.

3. Clinical and laboratory measurements

- Preparation phase, consist of Prepare chewing gum and probiotic yogurt drink , and also Explaining the research implementation before asking the respondent to signing the *informed consent* form.
- Research Stage. Saliva collection before the intervention was carried out after the subject of the first study break and the patient had consumed snacks as usual. After 30 minutes rest, the patient is not allowed to eat and drink. Respondents were instructed to sit in an upright position with the head slightly bent forward, to assist saliva collection. Respondents were asked to refrain from speaking and collect saliva in the mouth for 30 seconds (using a stopwatch) then spit saliva into the saliva *collecting pot*, in this case, repeated for up to 10 minutes (Baliga et al, 2013). The collected saliva before the intervention was stored in a sealed, labeled container (with a different sample code for each group), stored in an *icebox* and filled with *ice gel*. Then check for *decay* or carious teeth.
- Implementation of interventions consist of :
 - a) Chewing gum (brand disguised to respondents): consuming probiotic gum for 7 days once a day during the day. After 7 days, the pH and salivary flow rate were measured again.
 - b) Drink probiotic yogurt (brand disguised to respondents) as much as 100 ml: consume yogurt milk by drinking milk and holding it in the mouth for 5 seconds and then swallowing it until it runs out.

- Measurement of salivary pH. Salivary pH was measured using a calibrated single electrode digital pH meter. The entire electrode of the pH meter is submerged in saliva. Then the salivary pH is recorded on the datasheet.
- Measurement of salivary flow rate. Saliva was collected immediately for 5 minutes using a calibrated saliva pot with a 5ml limit. The resultant salivary flow rate obtained was recorded in ml/min.
- Measurement of saliva buffer capacity. It using saliva check buffer kit. A total of 1 mL of saliva was put into a tube containing Dentobuff solution. Then the tube was shaken for 10 seconds. The carbon dioxide content is evaporated for 2 minutes. The color that appears is compared to the color indicator in Dentobuff.

The saliva collection phase after the intervention was carried out by instructing the respondent to use the same technique when taking the initial saliva. The collected saliva was stored in an ice box and immediately brought to the laboratory.

4. Data Analysis

Data analysis was carried out as descriptively and analytically methods. Descriptive analysis was carried out univariately to determine the description of each variable, namely salivary pH, salivary flow rate, and salivary buffer capacity. The normality test was carried out using the Kolmogorov Smirnov test. When the data is normally distributed, the analysis uses the *Anova one-way test*, to compare salivary pH, salivary flow rate, and salivary buffering capacity between the three groups.

RESULT AND DISCUSSION

The implementation of initial data collection was carried out starting from the day I, and day 7. Ph Saliva, buffer capacity, and salivary flow rate was conducted in an integrated laboratory of the Faculty of Medicine, University of North Sumatra, Medan.

The frequency distribution of respondents according to the selected class based on simple random sampling was presented in the following table:

Table 1
Clinical characteristics and Baseline demographics of respondents
demographics of respondents

Amount (n)	Group I Control	Group II Chewing gum	Group III Yogurt	p
	20 People	20 People	20 People	
Gender, n (%)				
- Male	10 (50)	10 (50)	10 (50)	
- Female	10 (50)	10 (50)	10 (50)	
Number of Teeth (Mean \pm SD)	23,50 \pm 0,76	23,95 \pm 0,60	23,50 \pm 0,76	0,056*
Total Caries (Mean \pm SD)	3,90 \pm 0,78	3,90 \pm 0,718	3,90 \pm 0,788	0,769*
Saliva Flow Rate n(%)				
- Medium	15(75)	14 (70)	15 (75)	0,740*
- High	5 (25)	6 (30)	5(25)	
Buffer Capacity n(%)				
- Medium	17 (85)	19 (95)	16 (80)	0,059*
- High	3 (15)	1 (5)	4 (20)	

The respondents were in mix dentition stage with average number of teeth were 23-24 and the average number of caries were 3.8-3.9. It means that the respondents were at the high risk of being affected by caries. The respondents also have poor oral hygiene conditions. According to Motamayel et al, (2013) high caries risk factors was characterized by the presence of two or more caries that have recently occurred or have recurred in the last year, or two or more, including the presence of deep or non-fused pits and fissures, frequent consumption of sugar, salivary flow. poor oral hygiene, insufficient fluoride exposure, pits and fissures caries histories, family history of high caries rate, rampant caries, and presence of radiolucency in the proximal area.

Table 2
Kolmogorov-Smirnov Normality Test

Observation	pValue
- pH before	0,000
- pH after	0,000
- Buffer Capacity before	0,000
- Buffer Capacity after	0,078*
- Salivary flow rate before	0,003
- Salivary flow rate after	0,039

*=Data was normally distributed : $p > 0,05$

Based on the normality test using the *Kolmogorov - Smirnov* on the table show that not all of the data were normally distributed, So to compare the measurement data between the three groups before and after the intervention, the Kruskal Wallis test.

Table 3
The results of the measurement of the average salivary pH, buffer capacity, salivary flow rate after intervention

Variable	Group I (n=20) $\bar{x} \pm SD$	Group II (n=20) $\bar{x} \pm SD$	Group III (n=20) $\bar{x} \pm SD$	pValue
Ph Saliva	7,28 \pm 0,43	7,88 \pm 0,41	8,02 \pm 0,24	0,00*
Buffer Capacity	7,37 \pm 0,44	7,72 \pm 0,43	8,02 \pm 0,24	0,00*
Salivary Flow Rate	7,37 \pm 0,44	7,72 \pm 0,43	8,08 \pm 0,55	0,00*

*Significantly different data: ($n < 0,05$)

Based on table 6 above show that all p values have very significant differences. It was known that the salivary pH variable of the group III has the highest average final pH value (8.02 ± 0.24), while the lowest was group I (7.28 ± 0.43). Based on the saliva buffer capacity parameter, the lowest mean final score was 7.37 ± 0.44 for group I, while the highest was for group III (8.08 ± 0.55). Based on the salivary flow rate variable, the lowest mean score was in group I (7.58 ± 0.46) while the highest was in group II (0.14 ± 0.08).

Table 7
The average comparison between intervention groups using the Pos Hoc Bonfereni Test

Variable	Control Group & Yogurt Group	Control Group & Chewing Gum Group
Saliva Flow Rate	0.015*	0,000*
pH Saliva	0,000*	0,000*
Buffer Capacity	0,000*	0.0032*

*=Significantly related

Based on table 7 above shows that consuming probiotic yogurt was stronger in increasing saliva buffer capacity than chewing gum, but chewing gum was stronger in increasing the flow rate of saliva than drinking probiotic yogurt. In increasing pH saliva both drinking yogurt and chewing gum have an equally great effectiveness.

DISCUSSION

Saliva contains more than 99% water and several complex mineral mixtures. Saliva also contains oral epithelium, microorganisms, and leukocyte products, serum constituents, gingiva crevicular fluid, and food debris. Saliva has a normal pH range of 6.2 – 7.6 with an average of 6.7. Maintenance of pH takes place through 2 mechanisms. First, the flow of saliva removes carbohydrates that can be metabolized by bacteria and remove acids produced by bacteria. Both acidities from drinks and food as well as from bacterial activity are neutralized by the buffering activity of saliva (Baliga et al, 2013).

Data statistical showed that the average pH of saliva before drinking probiotic yogurt milk was 6.85 ± 0.50 and after the intervention changed to 8.02 ± 0.24 or increased by 1.17 ± 0.58 . This increase in salivary pH was greater than the chewing gum intervention (group 2) which increased by 1.03 ± 0.48 . It means that probiotic yogurt milk was better at increasing salivary pH than probiotic chewing gum. The results of this study are inline with the results of research conducted by Sulastri (2018) on students of SDN Puluhan Argomulyo Sedayu Bantul that there was an effect of drinking probiotic yogurt in increasing salivary pH compared to those who do not drink yogurt.

Salivary pH has been widely used as a diagnostic marker in periodontal disease. If the salivary pH is <5.5 in the oral cavity, then there is a subsaturation of Ca^{2+} and PO_3^- ions which cause the solubility of the enamel, which is called demineralization, resulting in dental caries. Caries teeth occur over time with the destruction of enamel crystals by acids produced by bacteria, namely *Streptococcus mutans*. Bacteria use carbohydrates as energy to generate energy in the glycolytic process and produce acid by-products so that if there is a lot of food residue or substrate attached to the teeth, the bacteria will be faster to produce acid which will result in tooth demineralization.

Based on data it is known that the initial value of the buffer capacity measurement of drinking yogurt milk group was 6.66 ± 0.35 , and after the intervention increased to 8.02 ± 0.35 or included in the low-risk category. The measurement of the buffer capacity from the initial to final was 1.36 ± 0.44 . This difference was higher than the chewing gum group (1.14 ± 0.55) and the control group had a difference of only 0.71 ± 0.39 . The function of saliva was the buffer ability of saliva in oral acid. It is also related to viscosity or volume. When pH saliva above to 5.5, the buffer action will occur to process super saturation of Ca^{2+} and PO_4 ions in which the hard tooth tissue can attract these ions so that the remineralization process will occur and the dental caries process will stop or be inhibited. The buffer capacity serves to restore the salivary pH that has fallen due to the entry of substrate into the oral cavity so that the salivary pH returns to its normal value. Salivary pH above 7.0 usually indicates alkalinity. Excessive alkalinity can cause anaerobic conditions similar to acidemia. Baliga et al., (2013). On the other hand, patients with low salivary buffering capacity have less tartar build-up because saliva pH tends to remain acidic for a long time, causing enamel demineralization.

Based on data, also shows that the salivary flow rate after the intervention was the highest in group II who chewed probiotic gum. Initially, the salivary flow rate was 6.79 ± 0.36 and after the intervention, it was 8.33 ± 0.59 or a difference of 1.53 ± 0.63 . This difference is higher than group 3 which has a difference of 1.30 ± 0.58 and the lowest is in the control group of 0.80 ± 0.37 . Probiotics are live microbes that, when consumed in adequate amounts, have beneficial effects on the health of the host, one of which is antimicrobial which results in a decrease in the number of cariogenic pathogenic bacteria such as *Streptococcus mutans* which is an acidogenic bacterium, i.e. bacteria that can produce acid (Fernandez, 2010).

In this study, the method of drinking probiotic yogurt milk that before swallowing, the milk is held and sucked in the mouth for 30 seconds which aims to make the exposure of probiotic bacteria to the oral cavity more intense and longer so that probiotic bacteria can stick to the tooth surface, especially those with caries. Probiotics are first exposed to saliva which mediates contact with the hard and soft tissues of the mouth. With

the provision of the probiotic solution, it is hoped that it can prevent caries which is a focal infection, especially in cavities.

The probiotic drink was chosen because various studies have mentioned that probiotic products can affect the bacteria that cause dental caries, especially *Streptococcus mutans*. The mechanism of probiotics is through the production of antimicrobial compounds, regulates the immune response and removes the adhesion properties of pathogenic bacteria, and replaces them with non-pathogenic bacteria (good bacteria). Probiotics can create a biofilm layer in the oral cavity that acts as a protective layer for disease-fighting tissues and the mouth. Protective layer for tissues against dental and oral diseases. This biofilm layer protects pathogenic bacteria and pathogenic tissues in the absence of a biofilm layer and competes with cariogenic bacteria and periodontal bacteria (Hasslof et al., 2010). Increased pH caused by probiotic bacteria through inhibition of cariogenic bacteria development. The probiotic used in this study was a probiotic drink containing *L. casei*. Bacteriocins produced by *L. casei* have the activity of inhibiting or killing microbes and selective toxicity against pathogenic bacteria.

CONCLUSION

Based on the results of the study, it can be concluded that drinking probiotic yogurt milk is better for increasing the salivary pH value and salivary buffer capacity than chewing probiotic gum and the control group, consuming probiotic chewing gum is better for increasing the salivary flow rate value than the probiotic and control group drinking yogurt milk. There was a decrease in the risk of dental caries after consuming probiotic yogurt milk or probiotic chewing gum.

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