ORIGINAL RESEARCH PAPER
Volume - 12 | Issue - 11 | November - 2023 | PRINT ISSN No. 2277 - 8179 | DOI : 10.36106/ijsr
INTERNATIONAL JOURNAL OF SCIENTIFIC RESEARCH

EFFECT OF SOAKING COW'S MILK ON TOOTH ENAMEL SURFACE ROUGHNESS

Dentistry

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ABSTRACT

Tooth enamel is the outermost part of the tooth structure and is the hardest part of the body. Demineralization and remineralization are natural processes in tooth enamel. Remineralization is the first step in bringing new minerals into a demineralized tooth. Remineralization can be produced by a solution containing calcium and phosphate ions, one of which is milk. Objective: This study aims to determine the effect of soaking pasteurized cow's milk on the surface roughness of tooth email. 32 premolars which were divided into 2 groups consisting of a treatment group and a contrl group. The sample of the treatment group was soaked in pasteurized cow's milk and the control group was given 20 ml of distilled water for 5 minutes. The treatment group was repeated every 6 hours per day for a week, and the controup group was repeated once a day. All sample were measured using an enamel surface roughness tester. Analysis of the data used is bivariant data for determine the differences between the two sample groups.16 teeth soaked in milk decreased the surface roughness of tooth enamel. There was a significant decrease in tooth enamel surface roughness after soaking in pasteurized cow's milk ($p<0.05$). There is an effect of pasteurize cow's milk drink on the surface roughness of tooth enamel.

KEYWORDS

Remineralization, Demineralization, Cow's Milk, Surface Roughness, Tooth Enamel

INTRODUCTION

Tooth enamel is the outermost part of the tooth structure and is the hardest part of the body. Roughness of the enamel surface can facilitate attachment and bacterial colonization which promotes demineralization. Enamel demineralization can occur if the enamel is in a pH environment below 5.5. The pH value plays a role in demineralization because a low pH will increase the concentration of hydrogen ions and these ions will damage the hydroxyapatite of tooth enamel, causing enamel surface hardness. (Farooq, Imran, and Amr Bugshan (2020)).

Tooth erosion is the progressive loss of tooth tissue in tissues that are not mediated by bacteria but are chemically exposed to acid on the tooth surface. The potential for erosion does not only depend on the pH value of each acid. The acid that causes erosion comes from food and drink not a product of the intraoral flora. The erosion potential in erosive drinks is reduced or even eliminated when acidic drinks are enriched with minerals, especially calcium. Research conducted by Vieira, et al. In 2018, stated that worldwide prevalence of dental erosion varies between 6% and 52% in children aged 2 years to 5 years in primary teeth and between 3% to 95% in adolescent permanent teeth. (Vieira A, Chung C, Raffensperger S, Muluk P (2018), Korner , Wiedemeier , Attin , Wegehaupt (2020))

Dental caries is a major oral health disease caused by demineralization of the hard tissue structure of teeth through organic acids produced from fermented carbohydrates by cariogenic bacteria in dental plaque. Demineralization and remineralization are natural processes in tooth enamel. Remineralization is influenced by saliva, oral hygiene, fluoride and other natural protective factors. The stability of the oral environment is affected by demineralization and remineralization. The demineralization process is greater than the remineralization process which causes tooth enamel to become caries. (Al-Ani , Al-Naimi (2020), Dzulfia , Damiyanti , Herda (2016))

One of the things that can overcome the risk of dental caries is by consuming milk. Milk can reduce erosion of tooth enamel because it contains phosphorus, calcium, lactoferrin, phosphate, fat, and natural casein which are good for health and can protect teeth from caries. Research Dzulfia, et al. In 2016, it was shown that cow's milk had an effect on increasing tooth enamel hardness.(Dzulfia , Damiyanti , Herda (2016))

Indonesia is the largest milk producer in Southeast Asia and its products are popular in Indonesia. Singapore, Malaysia, Hong Kong, Brunei and the Philippines. The need for a healthy lifestyle and materials high-quality food is increasing, but mostly met by imported products. So that Indonesia produces cow's milk to meet local and regional needs surroundings.(Dzulfia , Damiyanti , Herda (2016)).

Cow's milk is considered a complete food, providing high-quality protein and essential micronutrients, including vitamins and minerals. Cow's milk has been shown to contain fluoride and can increase bone remineralization. Research conducted by Rahardjo, et al. in the study of Dzulfia, et al. in 2016, proved that topical application using milk had the best protective effect against demineralization compared to the combined application of milk-tea and milk with 0.2% NaF. Research conducted by Damayanti in 2011, in the research of Dzulfia, et al. in 2016 stated that in 100 grams of milk there is 125 milligrams of calcium, this high calcium content is very beneficial for bone health, dental health and children's body growth. (Dzulfia , Damiyanti , Herda (2016), Antunes, I. & Bexiga, R. & Pinto, Carlos & Roseiro, Cristina & Quaresma, Mário. (2022)).

The effect of cow's milk on the surface roughness of tooth enamel has been significantly known to cause remineralization of the tooth enamel surface. Therefore researchers were interested in conducting research on the effect of soaking cow's milk on the surface roughness of tooth enamel and by comparison with the control group.(Dzulfia , Damiyanti , Herda (2016))

METHODS

The type of research used is laboratory experimental research (in vitro) with a pretest-posttest research design with control group design. The research was conducted at the Technical Laboratory of Tarumanagara University. This laboratory was chosen because it has a tool to measure tooth surface roughness, namely a surface roughness tester. The time of the research was carried out in March 2022 – May 2022. The sample in this study were 32 premolars. Crowns were still intact, No caries, No fillings, No white spots. Sampling was done by systematic random sampling technique. Samples were taken at random in a systematic way.

The samples used were premolars that matched the inclusion and exclusion criteria. All tooth samples were cleaned with running water. Samples were washed with running water and stored in saline solution at room temperature. Every two days the saline solution was changed until the time of the study in order to maintain the humidity of the teeth so that no changes occurred. If the sample wants to be used, then the sample is washed first with running water and dried with a tissue or chip blower. The sample was divided into two groups, namely the treatment group and the control group. The treatment group and the pre-test group will be planted on wax and the initial enamel surface roughness will be measured using a surface roughness tester.

Milk drinks (packaged pasteurized cow's milk) and aquadest are first measured in the pH of the container and measured with a pH meter. The sample is immersed in 20 ml of packaged cow's milk drink which is pasteurized in a container for 5 minutes then rinsed with running water and dried with a tissue or chip blower. The sample is immersed again in saline solution and the treatment is carried out in the same way every 6

hours for 24 hours because the sample is analogous to humans who consume cow's milk in the morning, afternoon, evening and night. Samples of the control group were immersed in 20 ml of distilled water for 7 days and replaced every 24 hours at room temperature.

On day 8, the entire treatment group and post-test control groups were cleaned with running water and dried with a tissue or chip blower before being planted in wax. All samples to be measured by roughness surface tester will be planted with wax first. All samples of post-test treatment and control groups that have been waxed will be measured using a roughness surface tester to measure the final surface roughness. The specimen is placed in the roughness tester unit. The tool starts working at the time specified by the tool. The results of the roughness of the specimen will appear through the results on the screen contained in the test equipment in units of Ra (Roughness Average).

RESULTS

The data obtained in this study resulted that cow's milk had an influence on the surface roughness of tooth enamel. Soaking the teeth in milk results in a decrease in the roughness of the surface of the tooth enamel. the results of the Independent T-Test showed that soaking in cow's milk and soaking in distilled water there were significantly different results.

Table 1 - Values of Surface Roughness Treatment Group

No. Sample	Before (µm)	After (µm)	difference (µm)
1	0,025	0,022	-0,003
2	0,109	0,064	-0,045
3	0,198	0,186	-0,012
4	0,251	0,220	-0,031
5	0,174	0,128	-0,046
6	0,111	0,110	-0,001
7	0,137	0,126	-0,011
8	0,173	0,102	-0,071
9	0,240	0,143	-0,097
10	0,281	0,223	-0,058
11	0,141	0,134	-0,007
12	0,332	0,124	-0,208
13	0,121	0,115	-0,006
14	0,109	0,103	-0,006
15	0,143	0,084	-0,059
16	0,223	0,179	-0,034
Average	- 0,04343		

Group 1 was the group that had 16 teeth soaked in pasteurized cow's milk. The enamel surface roughness value obtained by calculating the buccal surface roughness before and after treatment (pasteurized greenfields milk immersion) was measured using a surface roughness tester mutitoyo SJ-210. The result data on the roughness value can be seen from table 1.

Table - Surface Roughness Values for Control Group

No. Sample	Before (µm)	After (µm)	Difference (µm)
1	0.129	0,127	-0,002
2	0,169	0,147	-0,022
3	0,211	0,156	-0,055
4	0,382	0,300	-0,082
5	0,297	0,203	-0,094
6	0,391	0,317	-0,074
7	0,299	0,277	-0,022
8	0,059	0,049	-0,01
9	0,323	0,264	-0,059
10	0,615	0,285	-0,33
11	0,300	0,316	-0,016
12	0,451	0,110	-0,341
13	0,137	0,110	-0,026
14	0,599	0,308	-0,291
15	0,121	0,113	-0,008
16	0,593	0,389	-0,204
Average			-0,1022

The control group was the group that was immersed in 20/ml/sample of distilled water and soaked for 24 hours and then replaced every day for 7 days. The surface roughness value of the enamel was obtained using the surface roughness tester mutitoyo SJ-210. The result data on the roughness value can be seen from table 2.

Table 3 - Results of Paired T-Test Between Before Immersion in Cow's Milk and After Immersing in Cow's Milk

Paired Sample Correlation

	N	Correlation	Sig.
Before Soaking in Cow's Milk-After Soaking in Cow's Milk	16	0.742	0,004

* Sig. <0,05= There is a statistically significant difference

Based on the results of the Paired T-Test between before soaking cow's milk and after soaking cow's milk in table 3. there were significant results, namely P = 0.004 <0.05. In these results, there is a significant difference between before soaking cow's milk and after soaking cow's milk. This explains that soaking teeth with cow's milk can reduce the surface roughness of tooth enamel.

Table 4 - Paired T-test Results Between Before Immersion In Aquatic And After Immersion In Aquatic

Paired Samples Test

	N	Correlation	Sig
Before Soaking in Aquades - After Soaking in Aquades	16	0,762	0,005

*Sig. <0,05= There is a statistically significant difference

Based on the results of the Paired T-Test between before immersion in distilled water and after immersion in distilled water, table 3 shows significant results, namely P = 0.005 <0.05. In these results, there is a significant difference between before and after aquadest immersion. This explains that soaking teeth with cow's milk can reduce the surface roughness of tooth enamel.

Table 5 - Results of Independent T-Test Analysis Between After Immersion in Cow's Milk and After Immersion in Aquades

Independent Samples Test

	Sig.(2 tailed)
Independent T-Test	0,005

*Sig. <0.05= There is a statistically significant difference

Table 3 shows the results of the Independent T-Test statistical test to test whether there is a difference in the mean of the two sample groups that are not related or not paired. In the table above, the sig value is obtained. = 0.005 <0.05, which means that the average tooth surface roughness after soaking cow's milk and after soaking in distilled water there is a statistically significant difference. Based on the hypothesis that has been applied, it can be concluded that the research hypothesis is accepted.

DISCUSSION

This study was conducted to explain the effect of soaking pasteurized cow's milk on the surface roughness of tooth enamel. Tooth enamel is the outermost covering of the anatomical crown of the tooth. Tooth enamel is the hardest part of the human body which consists of more than 96% organic matter, the main component of tooth enamel is hydroxyapatite.

Milk is one type of drink that is often and widely consumed by the community, because milk is a product that contains many benefits. The content of phosphate and calcium ions in milk can affect the remineralization process in tooth enamel. Based on a survey from AC Nielsen Indonesia, Greenfields fresh dairy products controlled 54% of the product market share in the first quarter of 2017. The data is taken from sales records in super-super markets which show that Greenfields fresh milk sales reached 900,000 liters.(Yudhit, Astrid & Imanda, Kholidina, Dewi, Yuli (2019))

The treatment group used was 16 premolars which would be immersed in a container with 20 ml of pasteurized cow's milk (greenfields) for 5 minutes per 6 hours a day. Based on the enamel surface roughness values obtained before and after being treated using a surface roughness tester, it showed a decrease. Furthermore, the results of the statistical test carried out with the paired t-test, there was a value of p = 0.004 (p < 0.05) which showed a significant difference between before and after soaking cow's milk.

The effect of decreasing the surface roughness of tooth enamel is caused by various factors, namely extrinsic and intrinsic factors.

Extrinsic factors that cause a decrease in roughness or smoother occur when consuming drinks that have a low pH. This decrease in roughness occurs when the tooth is in a remineralized state. The lower the pH in saliva, the faster the hydroxyapatite bonds to tooth enamel or the faster the remineralization process.

Saliva had a buffering ability, which allowed it to maintain the pH balance of the oral cavity and prevent tooth demineralization.The demineralization process begins when tooth degradation occurs, when the tooth is exposed to a drink or food with a pH below 5.5 it will dissolve calcium in the tooth so that the hydroxyapatite crystals dissolve or are eroded. Milk can prevent the demineralization process and result in the remineralization process. The protein in milk sticks to the tooth surface, at that time calcium and phosphate ions diffuse below the enamel surface to fill the empty hydroxyapatite crystal space as a result of the demineralization process, when calcium and phosphate ions fill the empty hydroxyapatite space, a remineralization process occurs.(Sukanto, Ibnurrafif , Sulistiyani, Lestari, Pujiastuti, Setyorini, Probosari, Budirahardjo, Prihatiningrum (2023))

Solutions containing phosphate and calcium ions can cause remineralization. According to the results of research from Wisyaningtyas, et al. in 2014, stated that the process of tooth enamel remineralization is due to the presence of calcium and phosphate ions that diffuse from pure soy milk into the microporosity of the enamel. Research conducted by Wisyaningtyas, et al. in 2014, using cow's milk immersion using premolars and the milk will be changed every 8 hours. Remineralization can occur if a neutral pH contains sufficient Ca2+ and PO43- ions. Research results from Vieira, et al. in 2017, stated that milk produces enamel remineralization. (Vieira , Chung , Raffensperger , Muluk (2018), Obeid, Khalaf (2022))

This study used premolars including posterior teeth and used pasteurized milk. The difference lies in the research subject and the type of milk conducted by Fadil, et al. in 2021, using resin modified glass ionomer cement using UHT milk and the comparison is fresh cow's milk. The results were not significant in both milks, but similar to this study, there were significant results of remineralization between the fresh cow's milk group and the control group and UHT milk and the control group. .(Yudhit, Astrid & Imanda, Kholidina, Dewi, Yuli (2019))

In group 2, which is the control group, treated with distilled water resulted in a decrease in roughness or a smoother surface than the results of the paired t-test (table 4) of $p = 0.005$ ($p < 0.05$). The decrease in surface roughness of tooth enamel is caused by the pH factor in the solution that plays a role. The type of solution used for the control group was distilled water, where the composition of the distilled water resembled pure water. Aquades has a pH that is close to normal (pH 7). Research according to Hughes cited by S Kazmi in 2016 explains that pH has an important role in increasing roughness. The smaller the pH in the feeding solution, the higher the roughness level, the higher the pH in the solution, the lower the roughness that occurs.

The results of this study indicate that there are significant differences between the treatment group and the control group due to the phosphate, protein, and calcium content in pasteurized cow's milk. This research is strengthened by research from Amaliyah, et al. in 2021, which showed a decrease in tooth enamel surface roughness due to the protein content in cow's milk.(Farooq, Imran, Bugshan (2020))

CONCLUSION

Based on the results of research that has been carried out, there is a change in the form of a decrease in the surface roughness of tooth enamel after soaking in pasteurized cow's milk drink (greenfields), because the pH of the drink contained in this drink is 6.5. The content in milk is one of the substances that are good for bone remineralization, so it can cause a decrease in tooth roughness caused by:

The immersion time used in this study was short so that there was no change in pH which would cause the pH to become acidic and cause demineralization of the teeth.

Proteins attached to teeth cause calcium and phosphorus ions in cow's milk to diffuse below the tooth enamel surface so that phosphorus and calcium ions can occupy empty or eroded hydroxyapatite crystal spaces, resulting in a decrease in tooth enamel surface and a remineralization process.

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