Effect of gum Arabic (Acacia Senegal) topical gel application on demineralized enamel hardness

To cite this article: N Paramita et al 2018 J. Phys.: Conf. Ser. 1073 032016

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Effect of gum Arabic (*Acacia senegal*) topical gel application on demineralized enamel hardness

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Abstract. The aim of this study was to analyze the effect of a topical gel containing gum Arabic on demineralized enamel hardness after using a 1% citric acid solution. Bovine teeth (n = 27) were used for this study. Friedman and Kruskal–Wallis statistical tests were used to analyze the data. The results indicated that there was an increase in tooth enamel hardness after the application of the gel; however, the initial hardness could not be completely restored. The highest increase in enamel hardness was observed in the 96 min exposure duration group.

1. Introduction

Tooth enamel is one of the hardest tissues in the human body. However, unlike other hard tissues, teeth cannot biologically, chemically, or mechanically regenerate or repair their structure when damage occurs. Biological damage, such as dental caries, is caused by bacteria that reduce the tooth structure. In addition, the consumption of acidic food and beverages can also cause the reduction of tooth structure. Teeth consist of many layers, of which the outer layer is enamel, followed by dentin and pulp. Damage to the tooth structure due to acid produced by bacteria and food and beverage consumption begins with the softening of the tooth outer layer.

According to the Indonesian Household Survey in 2007, dental caries are an oral disease with a high prevalence (72.1%) in Indonesia [1]. Dental caries represent an infectious disease resulting from the interaction between various factors, such as the host, environment, and microorganisms. The tooth is the host in which dental caries occur. Many studies have reported that *Streptococcus mutans* are the predominant bacteria that cause dental caries [2]. Indeed, *S. mutans* changes the oral debris to become an acid with a very low pH [3]. The acidic conditions of the oral cavity surrounding the teeth then causes demineralization of the enamel surface, which is characterized by softened tooth enamel and tooth structure loss.

The consumption of acidic foods and beverages represents one of the etiologies for the reduction of tooth structure (Zero, 1996). Indeed, the acid in foods and beverages can lead to tooth erosion. In the early stages of erosion, the tooth structure will become soft, followed by the reduction of tooth structures in large quantities. Tooth erosion is the loss of dental hard tissue as a result of a chemical process caused by acid and does not involve bacteria. This acid may be derived from extrinsic factors such as foods and beverages, as well as intrinsic factors derived from human gastric acid [4].
Tooth enamel is an acellular hard structure of the body composed of inorganic components. Hydroxyapatite crystals represent the main inorganic component and are arranged neatly and orderly in tooth enamel. Hydroxyapatite crystals become separated from each other with a cavity containing water and organic material. This dynamic process results from chemical reactions, including enamel dissolution, ion exchange from the surrounding environment, and demineralization-remineralization reactions occurring on the enamel surface [2,3].

Remineralization is the disposition process of minerals in the cavity between hydroxyapatite crystals. Unlike remineralization, demineralization is the dissolution process of the mineral components of the enamel. Demineralization may cause the dilation of cavities between hydroxyapatite crystals, which leads to enamel softening. Remineralization and demineralization are strongly influenced by environmental conditions surrounding the tooth. When the concentrations of calcium and phosphate ions in the environment surrounding the tooth are high, the remineralization process occurs and the demineralization process can be prevented. Otherwise, demineralization occurs when there is a high hydrogen concentration around the surface of the tooth [4,5].

Fluoride is a tooth remineralization agent widely used to prevent further damage to the tooth structure due to demineralization. Fluoride is used in various preparations such as topical preparations and systemic administration. Topical preparations of fluoride can be found in gel and toothpaste, whereas systemic preparations are added to drinking water or can be used as systemic fluoride tablets. Fluoride is a proven chemical agent that provides resistance to acid, leading to the remineralization process. Hydroxyapatite (HA) on the tooth reacts with fluoride to form fluorapatite (FA), which has a higher resistance to demineralization. FA is not as easily dissolved as hydroxyapatite because it has more stable chemical bonds that lower the enamel solubility pH threshold [4]. However, excessive use of fluoride can lead to tooth fluorosis, discoloration, and gastrointestinal disease.

In this global era, technological advances have been used to search for fluoride substitution materials that can be used as alternative tooth remineralization agents. The sap of acacia (gum Arabic) is one of them. Gum Arabic has been widely used as a herbal medicine in Africa to treat various illnesses, such as sore throat, abdominal pain, and other gastrointestinal diseases, eye diseases, bleeding, and fever [6,7].

Gum Arabic is the sap derived from *Acacia senegal* trees originating from Nigeria. Gum Arabic contains high concentrations of calcium ions, which can potentially increase tooth remineralization. The concentration of calcium ions in gum Arabic can replace the calcium ions separated from hydroxyapatite crystals while preventing further demineralization of the tooth enamel. The withdrawal of calcium ions from gum Arabic by hydroxyapatite forms hydrogen bonds, which are more resistant to demineralization. T. Onishi et al (2008) showed that the concentration of unsoluble calcium and phosphate ions in gum Arabic enhances the remineralization of teeth [4,8,9]. This study was conducted on demineralized tooth enamel immersed in distilled water, NaF/ Sodium Fluoride, 1000 ppm gel, or a gum Arabic solution and demonstrated that the minerals formed in the demineralized tooth enamel immersed in NaF 1000 ppm were similar to the demineralized tooth enamel immersed in the gum Arabic solution.

Currently, remineralization agents derived from gum Arabic are not widely used, especially in gel preparations. The gel is created in semi-solid preparations composed of a suspension of particles that penetrate in the liquid [10]. Gel preparations are typically used topically on the surface of the body. Gel preparations have several advantages over other topical preparations, including that they can be spread evenly without pressure, they do not cause marks, and they are easy to apply [11].

In this study, we analyzed the effect of gum Arabic as part of a 20 mg/mL gel on tooth enamel that was demineralized with 1% citric acid. We also determined the differences in enamel surface hardness after gum Arabic 20 mg/mL gel exposure for 16, 48, and 96 min as an alternative dental caries prevention agent.
2. Methods
This research represented an experimental study performed on 27 pieces of bovine teeth divided into three treatment groups, i.e. 16, 48, and 96 min exposure groups. The number of samples in each treatment group consisted of 9 pieces of bovine teeth. All enamel initial hardness values were measured before treatment, followed by enamel soaking in a 1% citric acid solution (pH 4) for 10 min at 37 °C.

Gum Arabic 20 mg/mL gel was made by dissolving 2000 mg gum Arabic powder in 100 mL water using a magnetic stir plate until homogeneous. After the gum Arabic 20 mg/mL solution was obtained, the acidity value of the solution was measured using a pH meter. A 2 M NaOH solution was then added to neutralize the pH, and the solution was filtered using filter paper to obtain a clearer solution. Natrium carboxymethyl cellulose (Na-CMC) was added to obtain the APF 1.23% topical gel consistency.

The gum Arabic 20 mg/mL gel was applied to the enamel surface for a 16 min duration in the first group, 48 min in the second, and 96 min in the third group. Final hardness measurements were made after the entire sample, which had been applied with gel, was rinsed with distilled water.

Enamel surface hardness test results were analyzed using Shapiro–Wilk normality tests. Data that were not normally distributed were then tested by the Friedman, Wilcoxon, Kruskal–Wallis, and Mann–Whitney tests. Statistical tests performed in this study had a 0.05 significance level (p = 0.05) and a 95% confidence level (α = 0.05).

3. Results
Tooth enamel hardness values are shown in Table 1. There was impairment in the enamel hardness value after soaking in a demineralization solution, and a subsequent increase in the enamel hardness value after the application of the gel as divided in the three treatment groups as shown in Fig. 1. Friedman tests were conducted to analyze the enamel hardness value significance for each treatment group with 16, 48, and 96 min exposure. Our results showed that there was a significant difference between the initial hardness after demineralization for the 16, 48, and 96 min exposure groups.

Table 1. The average value of enamel hardness (KHN) before and after demineralization with citric acid, and after gum Arabic 20 mg/mL gel exposure.

| Treatment                          | Exposure Time   |
|------------------------------------|-----------------|
|                                    | 16 minutes (16’) | 48 minutes (48’) | 96 minutes (96’) |
| Initial                            | 347.78 ± 20:43  | 335.67 ± 16:48  | 337.85 ± 18:04  |
| After demineralized with 1% citric acid pH 4 | 130.96 ± 10:39  | 119.56 ± 12:21  | 120.30 ± 10.84  |
| After Gum Arabic 20 mg / mL gel application | 148.22 ± 12:15  | 139.56 ± 22.90  | 167.19 ± 26.94  |

Wilcoxon tests were conducted to analyze significant differences for the three treatment group exposure times, i.e. 16, 48, and 96 min. The Wilcoxon test showed a significant difference between the initial hardness value and demineralization, between the hardness after demineralization and after application of the gel, and also between the value of the initial hardness and after application of the gel, for the three exposure groups.
Figure 1. Comparison diagram of the mean enamel hardness (KHN) for the three exposure groups

The Kruskal–Wallis tests were conducted to analyze significant differences of enamel hardness values in the initial, after demineralization, and after gel application for the three different exposure times. Our results showed that there were no significant differences between the initial hardness value and the hardness value after demineralization for the three exposure groups. However, the results of Kruskal–Wallis statistical tests showed a significant difference between the enamel hardness value between the three exposure groups.

Post hoc analysis using the Mann–Whitney test was used as a continuation of the Kruskal–Wallis test and showed that there were no significant differences in enamel hardness values in the 16 min and 48 min groups. Significant differences were observed for the enamel hardness value between the 48 min and 96 min groups, as well as the 16 min and 96 min groups.

4. Discussion

Study results demonstrate an increase in hardness values, which was significantly different for all three groups, i.e., 16, 48, and 96 min exposure times. Enamel surface hardness values were obtained using the Zwick Knoop micro hardness testing tool (KHN) with load were equal to 50 grams with 5 seconds indentation [12].

Demineralization process simulation was demonstrated with the immersion of tooth enamel in a 1% citric acid solution at a pH of 4. Citric acid was used as a demineralization solution because citric acid is usually present in foods and beverages to produce a fresh and sour taste [13,14]. Elsbury states that citric acid causes faster erosion, particularly at low pH. Indeed, citric acid has a destructive nature to tooth enamel that is two times greater than either nitric acid or hydrochloric acid because of its greater affinity toward calcium. Thus, citric acid causes damage more rapidly to the enamel
hydroxyapatite crystals bonds. The hardness value impairment caused by the loss of appetite crystals ions with a specific sequence in enamel surfaces. The first missing ion is hydroxy, followed by calcium and phosphate [15]. The erosion process starts from the outside and moves to the inside layer and is caused by the loss of hydroxyapatite crystals on the enamel surface so the enamel surface becomes softer and more vulnerable to physical stimuli [16].

Onishi et al. showed that a gum Arabic San-ei 10 mg/mL solution had a remineralization effect similar to 1000 ppm sodium fluoride with the same exposure time. In this study, we used double the concentration of gum Arabic compared with Onishi’s research (i.e., 20 mg/mL). Markus Beyer et al. showed that the addition of gum Arabic mixed into an acidic drink can reduce the erosive effect on tooth enamel.

The remineralization effect of gum Arabic in a 20 mg/mL gel in this study was seen by the increase in the enamel hardness value after exposure for the three exposure times. In the group of 16 min exposure time, the enamel hardness value increased about 13.18% from the enamel hardness value after demineralization. In the group of 48 min exposure time, the enamel hardness value increased about 16.72% from the enamel hardness value after demineralization. In the group of 96 min exposure time, the enamel hardness value increased about 38.98% from the enamel hardness value after demineralization. In addition, Friedman test results showed a significant difference (p < 0.05) for all three groups of exposure time.

Enamel hardness values increased due to polysaccharides and the high concentration of minerals contained in gum Arabic. This can be seen from micro radiological formation of the radiopaque layer in demineralized enamel surfaces [8]. The establishment of a radiopaque layer is due to the increased mineral distribution quantity. The high concentration of minerals (calcium, magnesium, and sodium) contained in gum Arabic can replace the calcium ions dissolved from hydroxyapatite crystals in the enamel surfaces as a result of immersion in the citric acid solution. Calcium, magnesium, and sodium are found in salt form as constituents of the main component fraction of gum Arabic [17].

Another study has shown the formation of a protective coating on the enamel surface in the form of a polymer layer that can reduce the erosive effect of acid exposure on enamel surfaces. The forming process of the polymer layer is caused by the adsorption of the polymers contained in gum Arabic (1-arabinose, δ-galactose, rhamnose, and δ-l-glucuronic acid) on the hydroxyapatite in the enamel surface. The fourth polymer forms an interaction with the hydroxyapatite nanoparticles, which is described as an electrostatic interaction and the formation of hydrogen bonds with Ca ions contained in the enamel surface. Furthermore, hydrogen bonds formed between polymer molecules form a coating layer that protects the enamel surface from the erosive effects of the acid. Both of these mechanisms are macroscopically visible as an increase in hardness [9].

5. Conclusion
It can be concluded that gum Arabic (A. senegal) as part of a 20 mg/mL gel may increase the value of enamel hardness after demineralization with 1% citric acid at pH 4 in all three exposure time groups. Gum Arabic 20 mg/mL gel exposure for 96 min had the highest increase in enamel hardness value compared with 16 min and 48 min exposure times. However, the increased hardness was not sufficient to completely recover the initial hardness.

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