Original Article

Effect of Zamzam Water on Microhardness of Primary Tooth Enamel After Erosion Induced by Claritin Syrup: An In-vitro Study

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Background: Dental erosion is an irreversible damage to hard tissues in the mouth due to acid dissolution not induced by microorganisms. Oral medications contributed to the etiology of dental erosion. Among these medications, antihistamine-containing syrups were reported to have a considerable erosive effect on hard dental tissues. Objective: The objective was to evaluate the effect of Zamzam water on the microhardness of primary tooth enamel after erosion induced by Claritin™ syrup. Materials and Methods: The present in-vitro study was conducted on 25 primary central incisor teeth. The labial surfaces of the specimens were prepared for microhardness testing. Baseline microhardness was measured first, then the samples were submerged in the erosive agent Claritin™ syrup, for 30 min every 12 h for 12 days. Thereafter, all the teeth were placed in Zamzam water for 30 min every 12 h for 12 days. The microhardness was measured by the Vickers hardness tester and expressed as Vickers microhardness value (VH). The measurements were completed three times for the same sample: baseline, after erosion, and after Zamzam treatment. Data were analyzed statistically using one-way repeated analysis of variance (ANOVA) followed by Tukey’s post hoc test. Results: The primary teeth included in this study showed a significant increase in their surface microhardness after Zamzam water treatment; the mean value of VH was increased from 33.12 ± 1.62 to 89.32 ± 8.52 (P-value <0.0001). Conclusion: Zamzam water could be introduced as a part of the management of dental erosion in the primary dentition.

Keywords: Antihistamine, dental erosion, microhardness, remineralization, Zamzam water

INTRODUCTION

Dental erosion may affect children and negatively impact their quality of life.[1] Severe forms of the disease can be expected if left unrecognized including teeth sensitivity and pulp exposure.[2] Dental erosion is an irreversible damage to hard tissues in the mouth due to acid dissolution not induced by microorganisms.[3] As the pH in the oral cavity reaches its critical value of 5.5, the acids will demineralize, soften, and decrease the microhardness of the enamel surface.[4-6]

The prevalence of dental erosion might vary from one region to another. In Sharjah, United Arab Emirates, about 58.8% of 5-year-old children had dental erosion in 2016.[7] The prevalence in Riyadh, Saudi Arabia was 61% among 3–5-year-old children who had a minimum of one surface affected by erosion.[8] Al-Majed et al.[9]
surveyed 5–6-year-old schoolboys in Riyadh and found that 95% had erosion in at least one primary tooth; 34% of them had pronounced erosion that extended into dentin or pulp. In Jeddah among preschoolers, the prevalence of erosion was 31%.\(^{10}\)

Although dental erosion might occur due to gastroesophageal reflux disease and consumption of acidic diets,\(^{11}\) oral medications could also contribute to the etiology of dental erosion.\(^{4,12,13}\) The severity of erosion was directly related to medicine exposure time.\(^{13,14}\) Claritin™ syrup was considered in this study since it had the most erosive pattern among antihistamine-containing syrups.\(^{12}\)

In the Kingdom of Saudi Arabia, specifically in Makkah, Zamzam well is located. Zamzam water is filtered by microfilters and disinfected by ultraviolet light prior to its distribution to the consumers. Microbiological tests confirmed that there are no signs of microbial growth in the water obtained from Zamzam well.\(^{15,16}\)

Analysis of Zamzam water revealed a high level of fluoride, calcium, chloride, sodium, sulfate, magnesium, nitrate, potassium, bicarbonate, nitrate, and sulfate.\(^{17}\) The four harmful elements, arsenic (As), cadmium (Cd), lead (Pb), and selenium (Se), were found within the accepted quantity that is much below the hazardous level.\(^{18,19}\) The concentration of arsenic detected in the Zamzam water samples was below 10 ppb, that is in consonant with the Saudi Standards, Metrology, and Quality Organization (SASO) for drinking water.\(^{20}\)

Several researches suggested that Zamzam water has preventive and therapeutic potentials. It prevents the formation of renal stones in rats.\(^{21}\) Additionally, it has an anti-cancer effect against lung cancer cells\(^{22}\) and it reduced the effect of gamma irradiation on bone marrow cells.\(^{23}\) *Streptococcus mutans* is considered one of the most cariogenic pathogens in the oral cavity, an experimental probiotic-Zamzam water mouthwash effectively reduces the *S. mutans* count *in vitro*.\(^{24}\)

Dental erosion cannot occur without a disturbance in the salivary pH.\(^{25}\) Management, therefore, should be directed toward buffering the reduced pH in addition to the use of remineralizing agents. Zamzam water has been proposed to effectively manage the initial carious lesions.\(^{26,27}\) Zamzam water exhibits a high buffering capacity due to its high levels of minerals.\(^{28}\)

In this study, we considered the gaps in the literature related to pediatric population applicability. The proposed benefits of Zamzam water were examined on permanent teeth. The magnitude of the effect is expected to be more pronounced in primary teeth.\(^{29}\) Children mostly consume the liquid form of medications as opposed to tablets.\(^{30}\) The aim of the present study was to evaluate the effect of Zamzam water on the microhardness of primary tooth enamel surface after erosion induced by Claritin™ syrup.

**Materials and Methods**

**Study design and eligibility criteria**

The present experiment is a prospective, in-vitro, single-arm repeated measure study. It was conducted on 25 primary central incisors that were extracted due to physiologic mobility or over-retention. The sample size was estimated by G*Power Software Version 3.1.9.2. Released 2014 (Kiel University, Germany). Considering alpha error left at 5%, the effect size is to be measured (d) at 80%, and the statistical power of the study at 85%.

All the teeth were obtained from the Pediatric Dentistry and the Oral Surgery Departments, College of Dentistry, Umm Al-Qura University. The primary central incisors were examined by a stereomicroscope at 40× magnification, and the eligible incisors must be sound. Teeth with visible caries, cracks, or developmental defects were excluded. The selected primary incisors were washed with de-ionized water; debris was removed with acetone. The teeth were stored in de-ionized water with 0.1% thymol to inhibit microbial growth.\(^{32}\)

**Specimens’ preparation**

The specimens were prepared for microhardness testing using acrylic models by mixing the acrylic powder with monomer; the root of each tooth was covered until the cervical line. All the labial surfaces of the teeth were prepared by slight touch using tapered yellow diamond stone bur and polished with disc grit bur using a water-cooled low-speed handpiece to remove the normal structural variations in the surface enamel. This procedure produced a flat surface to allow accurate readings of Vickers indentations.

**pH measurement**

Claritin™ syrup (manufactured by SPIMACO ADDWAEIH, KSA) and Zamzam water bottles (purchased from local market) had labels presenting their ingredients; however, none presented their pH values. Consequently, pH values of both Claritin™ syrup and Zamzam water were measured with a digital pH meter (AD1000, ADWA Instruments Kft., Szeged, Hungary).

**Interventions**

Two sequential interventions were utilized in this study: first by erosive agent and then by Zamzam water. The erosive agent was Claritin™ syrup that
contains 5 mg/5 mL micronized loratadine, used as an antihistamine medication in children. All samples were submerged first in the erosive agent Claritin™ syrup, for 30 min every 12 h for 12 days. The erosive agent was refreshed before each cycle throughout the experiment. Thereafter, all the teeth were washed with de-ionized water and then placed in Zamzam water for 30 min every 12 h for 12 days. After each cycle in Zamzam water, the teeth were washed for 2 min with de-ionized water and kept in de-ionized water to the next cycle to avoid dehydration and the changes in the microhardness.[32] No stirring was performed in all interventions.

**Microhardness testing**

The outcome variable was microhardness measurement of primary incisors performed by the ZHVμ Micro Vickers hardness tester, ZwickRoell Indentec, UK. The measurements were performed three times for the same sample: baseline, after-erosion, and after-Zamzam treatment cycles on the 12th day. All teeth were assessed by Vickers microhardness tester at a load of 50 g for 15 s. The microhardness measurements were expressed as Vickers microhardness value (VH). The personnel committed to testing the samples was kept ignorant about the purpose of the study throughout the multiple measurements.

**Statistical analysis**

Descriptive statistics were calculated as mean and standard deviation (SD) for Vickers microhardness measurements. Differences in the mean values of microhardness at baseline, after erosion, and Zamzam water were compared using one-way repeated analysis of variance (ANOVA) followed by Tukey’s *post hoc* test. Also, the mean difference along with a 95% confidence interval (CI) was reported. ANOVA assumptions were not violated by the data at hand. Data were analyzed by STATA, version 15.1 (StataCorp LP, College Station, TX, USA). The level of statistical significance was set at 0.05.

**Results**

The pH values determined in our study for Claritin™ syrup and Zamzam water were 3.20 and 7.78, respectively.

The mean value of Vickers microhardness for enamel in primary incisors at baseline was 158.36 ± 67.71 [Figure 1]. The microhardness mean value was significantly reduced after the erosion cycle with Claritin™ to 33.12 ± 1.62 [Figure 2]. After Zamzam water treatment, the microhardness mean value was significantly increased to 89.32 ± 8.52 [Figure 3]. The repeated ANOVA model showed a significant difference among the microhardness values at baseline, after-erosion, and after-Zamzam (*P*-value <0.0001).

The adjusted pairwise comparisons showed a significant difference between the microhardness values after-erosion in comparison with after-Zamzam treatment, *P*-value <0.0001, mean difference 56.20 (95% CI: 43.32, 69.08). Also, there was a significant difference between baseline and after-erosion microhardness values, *P*-value <0.0001, as shown in Table 1.
Our study showed that Claritin™ syrup significantly reduced enamel microhardness in primary teeth; the microhardness mean was reduced by 79.09% after 12 days of Claritin™ cycles. Zamzam water significantly enhanced the enamel microhardness after erosion; microhardness increased by 169.7% after 12 days with Zamzam treatment relative to erosion microhardness.

The observed loss of microhardness after Claritin™ syrup use can be explained through mineral loss, citric acid content, high titratable acidity, low endogenous pH, highly viscous consistency, and lack of fluoride and phosphate.[4,33] The pH is a “dominant factor” in any erosive dissolution of enamel[25]; in our study, the pH value for Claritin™ was 3.20, which is less than that reported in previous studies.[4,11,34]

Various studies that used antihistamine syrup, as an erosive agent, showed similar findings to our study. Costa et al.[4] replicated day and night use of antihistamine medication. They found an 89.14% reduction in the microhardness of primary teeth. The study by Valinoti et al.[12] showed a 71.6% reduction in enamel hardness after using simulated erosive oral condition for primary enamel. The study by Gaber et al.[5] showed a 46.66% decrease using the same protocol performed in our study. The slight variation in the magnitude of the reduction between the studies was not profound and can be attributed to differences in the practiced protocol for the intervention and type of teeth (primary vs. permanent).

In our study, we chose the frequency (of every 12 h) and the duration (12 days) of Claritin™ to mimic the use of antihistamines in real life. In addition, repeated exposure to the erosive agent produced more reliable results.[25]

After reduced hardness with Claritin™ syrup, Zamzam water increased the microhardness by more than 150%. Although the mechanism of action is not entirely known, it is expected that high buffering capacity, high mineral content,[28] and high pH played a major role. In the oral cavity, the critical pH has been set at 5.5; Zamzam water had a pH higher than 5.5.[19] In our study, the pH value for Zamzam water was 7.78.

Due to the unique composition and quality, Zamzam water has been investigated in the dental literature for various purposes. Al-Rawi et al.[26] initiated carious lesions in permanent teeth. Then, they used Zamzam water that resulted in a 76.36% increase in the microhardness of enamel. The improvement was less than what is reported in our study, probably for two reasons: the examined teeth were permanent and the overall duration and frequency of Zamzam water treatment were much less than what has been practiced in our study. Another study also used Zamzam water for permanent teeth but with more duration and frequency of exposure than Al-Rawi et al.[26] study. The microhardness was increased by 80.9%.[27]

The limitations of this study included the lack of other intervention arms, using only one measurement for erosion and cautious clinical interpretation. We attempted to measure the effect of Zamzam water on erosion. We did not include other types of water or fluoride-containing vehicles since Zamzam water, relative to other water types, had high mineral content especially fluoride and buffering potential.[28,36] Also, the effect of Zamzam water on the microhardness of demineralized enamel surface was not different from that of sodium fluoride.[26] Moreover, the use of fluoride preparations may supplement rather than antagonize the use of Zamzam water treatment. There are many outcomes to measure changes in the enamel including erosion; microhardness is one of them.[37] It evaluates the softening effect of dental erosion on the tooth structure, particularly in the initial stages.[13,37]

In our study, the enamel microhardness values significantly increased with Zamzam water treatment of the eroded enamel surface with Claritin syrup, however not reaching the baseline values. Further researches are needed to determine the maximum remineralization effect of Zamzam water on the eroded enamel surface through time and to compare the efficacy of Zamzam water with other sources of water, anti-erosive and re-mineralizing agents involved in the prevention of dental erosion.

**Conclusion**

Zamzam water could be introduced as a part of the prevention of dental erosion in the primary dentition.
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**CONFLICT OF INTEREST**
The authors declare no conflicts of interest, financial or otherwise.

**AUTHORS CONTRIBUTIONS**
Not applicable.

**ETHICAL POLICY AND INSTITUTIONAL REVIEW BOARD STATEMENT**
The research proposal of the study was approved by the Ethical Committee (IRB Number: 141-19) at the College of Dentistry, Umm Al-Qura University.

**PATIENT DECLARATION OF CONSENT**
Informed consent was obtained from parents or legal guardians to participate in the study and to gain the extracted teeth.

**DATA AVAILABILITY STATEMENT**
The data that support the study results are available from the corresponding author (Dr Adel Fathi, e-mail: afhussien@uqu.edu.sa) on request.

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