Original Article

Comparison of the microhardness of primary and permanent teeth after immersion in two types of carbonated beverages

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Abstract

Objectives: The consumption of carbonated beverages is one of the etiological factors that cause dental erosion. The purpose of this research was to compare changes in the microhardness of permanent and primary teeth after immersion in two types of carbonated beverages. **Materials and Methods:** This investigation was done on 30 healthy permanent molars and 30 healthy primary canines. Each group of primary and permanent teeth was subdivided into three groups of 10 teeth. The teeth was immersed in 40 ml of each of the three beverages for 5 min. One subgroup was immersed in water (as a control). The next was immersed in Lemon Delster and the last subgroup was immersed in Coca-Cola. The microhardness of enamel was measured using the Vickers method before and after immersion. Finally, the data was analyzed by paired *t*-test, one-way analysis of variance, and *t*-test. **Results:** Microhardness reduction in the primary teeth was significant in both the Lemon Delster and Coca-Cola groups ($P < 0.05$). This reduction was also statistically significant in the permanent teeth ($P < 0.05$). A comparison of the enamel changes in the primary teeth with permanent teeth after immersion in both beverages showed a greater microhardness reduction in the primary teeth in both the experimental groups. **Conclusions:** Coca-Cola and Lemon Delster caused a significant reduction of microhardness in tooth enamel. This reduction was greater in primary teeth than in permanent teeth, and was also greater after immersion in Coca-Cola than after immersion in Lemon Delster.

Key words: *Carbonated beverages, deciduous tooth, hardness, permanent tooth, tooth erosion*

INTRODUCTION

Tooth enamel, the hardest tissue in the body, protects dental tissues against external factors; however, it can be irreversibly destroyed or damaged despite many resistance factors.[1] Acid is one of the factors that can destroy the enamel and can be produced directly from food or by bacteria. Nowadays, various soft drinks, which are widely available to children and adults, play an important role in this process. According to statistics, although the prevalence and severity of dental caries has decreased in children, particularly in developed countries, dental abnormalities—particularly erosion—have increased.[2,3] The main cause of erosion is exposure of the teeth to acid.[4] Common sources of acid include beverages, such as fruit juice, and other non-alcoholic beverages that contain citric

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acid; beverages that contain carbonation; and dietetic beverages. Common causes for exposure to acid include repeated vomiting, gastroesophageal reflux, esophagitis, and swimming in pools that contain nonstandard and high levels of chlorine.[1,5] In one particular study, it was reported that beverages adhere more abundantly to the enamel than saliva and other sugar-free drinks, such as orange juice.[6] Other studies have shown that acidic beverages cause tooth demineralization. Dietetic beverages are categorized as acidic beverages.[7-9]

Different studies have been conducted regarding the effect of different substances such as beverages and acids on the microhardness of primary and permanent teeth separately;[10,11] however, to date, no studies have specifically compared the effect of beverages on the microhardness of primary and permanent tooth enamel. Considering the differences between primary and permanent enamel, effect of beverages on the enamel of permanent and primary teeth enamel can be different. Therefore, the purpose of this study is to compare the effect of two types of beverages on the microhardness of primary and permanent enamel.

The aim of this study was to compare the microhardness of primary and permanent teeth after immersion in two types of beverages, namely, Coca-Cola and Lemon Delster.

MATERIALS AND METHODS

This study was done as an experimental *(in vitro)* study and was conducted on 30 impacted healthy permanent third molar teeth, which were surgically extracted, and 30 canine primary teeth. Sample size was determined based on similar studies.[7,11] The teeth that were included in the study did not have any caries, hypoclassification, erosion, or cracks according to clinical examination (caries was assessed according to the World Health Organization's criteria). The teeth that did not meet the inclusion criteria were excluded from the study. These teeth were placed in new glass containers that were free from any intervening abrasives and that were purchased particularly for this study. The containers were then filled with tap water and were kept at room temperature. The water in the containers was changed twice a week during this period of time to prevent pollution because of surface changes. The surfaces of the teeth were polished and evaluated for the presence of any enamel effects, caries lesions, and cracks using stereomicroscope (Carton Optimal Industries Ltd, Model SCW-E, Thailand) at 40× magnification.

Square labels measuring 5 × 5 mm were stuck to the distal surface of each tooth. The remainder of each dental surface was covered with transparent self-cured acrylic; thus, the surface of all the teeth was the same without considering the size, shape, and group. The samples were kept in cool water to avoid changes occurring during the acrylic hardening process. After the acrylic was cured, the surface of each sample was polished in water with grit tissue 5000 to achieve a flat, hard surface.

The first microhardness measurement of the teeth was obtained with a Vickers microhardness tool (Shimadzu, Model M-g 5037 model, Japan). The best point for force was determined, following which the samples received 50 g of force.

Each group of primary and permanent teeth was subdivided randomly (using sequentially numbered containers) into three groups of 10 teeth, yielding three subgroups for each original group of primary and permanent teeth. The first subgroup was immersed in tap water from region 6 of Tehran as a control group. The second subgroup was immersed in Behnoosh Lemon Delster as an experimental group. The third subgroup was kept in Coca-Cola, a cola beverage, as an experimental group. Based on an announcement from the Ministry of Industries and Mines, these beverages are the most frequently consumed beverages in Iran; in particular, Delster that is produced in the country. All three beverages used came from the same batches, and their pH was measured with a pH meter (Metrohm Ltd., Herisau model CH-9101, City, Switzerland). Table 1 shows the pH of each beverage.

Each of the teeth was immersed in 40 ml of the beverage designated for the subgroup; the teeth were immersed in the beverage, in a graded container immediately after opening, for 5 min. To simulate the conditions under which consumers typically drink the beverages, all the beverages were placed in a refrigerator at a temperature of 9°C at the time of the experiment. During this time, the beverages were slowly stirred.

The surface of each sample was washed after immersion in each beverage. Then, a person who was not

Table 1: pH of beverages

Liquid	pH
Zam Zam cola beverage	−3/26
Behnoosh Lemon Delster	4/02
Tap water	6/67

aware of how the teeth had been divided measured their microhardness. Next, the data were analyzed. The changes in microhardness were measured by paired *t*-test in each group and by one-way analysis of variance (ANOVA) and *t*-test between groups. The SPSS Inc. Released 2007. SPSS for Windows, Version 16.0. Chicago, SPSS Inc. was used for statistical analysis (the statistical consultant was unaware of the beverages used with each group).

RESULTS

The effect of the two beverages on the enamel of the permanent and primary teeth is displayed in Table 2. According to these results, both groups of the teeth (permanent and primary) suffered the largest decrease in the microhardness of enamel after they were immersed in the Zam Zam cola beverage. In addition, the mean changes in both groups of teeth that were immersed in tap water were not significant ($P > 0.05$).

In the primary teeth, there was a significant difference in the rate of loss of microhardness in both the water–Delster and water–Zam Zam Cola groups ($P < 0.05$), whereas this difference was not significant in the primary or the permanent Zam Zam Cola–Delster groups ($P > 0.05$).

In the permanent teeth, there was a significant difference in the water–Zam Zam Cola group ($P < 0.05$). A comparison of the effect of both experimental beverages on the microhardness of the enamel of permanent and primary teeth by independent *t*-test showed that the mean of the microhardness changes was greater in the primary teeth than that in permanent teeth for both Lemon Delster and Coca Cola [Figure 1].

DISCUSSION

Dental erosion and tooth damage because of chemical agents with no interfering bacteria is a growing problem in modern societies. Beverages, in particular carbonated beverages, are one of the most important factors that contribute to these problems.[1]

In this study, the effect of two types of acidic beverages on the microhardness of the enamel of permanent and primary teeth was assessed. Some studies have been conducted in the past to evaluate the effect of pure acid and beverages on the microhardness of the enamel of permanent and primary teeth separately by using different methods, however in this study, for the first time the effect of erosive beverages in these two dentitions were compared.[7-11]

Results showed that these two beverages caused a significant reduction in microhardness of tooth enamel, and the microhardness reduction was greater in the primary teeth than in permanent teeth. There are some differences between the primary and permanent enamel tooth such as differences in thickness and mineralization, and these factors can affect the results.

Results also showed that microhardness reduction was greater after immersion in Coca-Cola than after immersion in Lemon Delster. Different composition

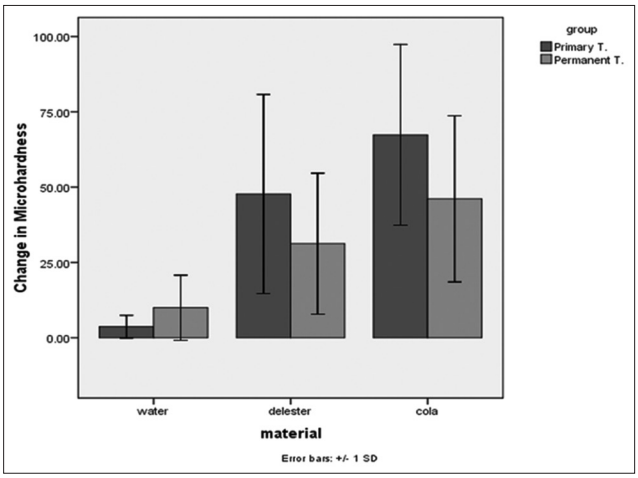

Figure 1: Comparison of the effect of water, Lemon Delster, and Zam Zam cola on permanent and primary teeth

Table 2: Micro-hardness of teeth before and after immersion in three types of beverages

Type of Teeth	Beverage	Sample size	Mean change	SD	*P**
Primary	Water	10	−0.90000	5.34039	0.607
	Lemon Delster	10	−47.73333	33.04273	0.001
	Zam Zam Cola	10	−67.36667	30.00883	0.000
Permanent	Water	10	−2.23333	14.89556	0.647
	Lemon Delster	10	−19.06667	35.06988	0.120
	Zam Zam Cola	10	−46.13333	27.59996	0.001

**(level of significance: 0.05)*

of these two beverages probably plays a role in different effect in their tooth demineralization.

West *et al*. measured the effect of acids and pH on the erosion of dentine and enamel, and showed that citric acid, with a higher pH, had lower erosion ability than phosphoric acid, which has a lower pH.[12] However, based on the results of the present study, the loss of enamel was lower in teeth that were immersed in Lemon Delster, which contains citric acid and has a higher pH than those which were immersed in cola, which contains phosphoric acid and has a lower pH. These differences may be because of the presence of other acid-containing compounds, carbonated beverages, and other ingredients in Lemon Delster that may cause some changes in the initial effect of the acid.[13]

Meanwhile, Lippert *et al*. showed that the softening effect of lemonade and cola, which contain phosphoric acid, on the enamel was greater than the effect of fruit juices, which contain citric acid. These results are consistent with the present results.[14]

However, in the study conducted by Lippert *et al*., the samples were stored in sodium hypochlorite before testing, which caused changes in the properties of the enamel and was considered an intervention.[15]

Fallahinejad *et al*. stated that acidic beverages cause teeth to lose more calcium than the standard and control solutions. They compared 4 types of beverages with 4 standard solutions that had the same acid and pH as the beverages. This indicated that gases and other ingredients in beverages, such as sugar, could augment the effect of erosion in beverages more than that of plain acid.[16] In this research, both spectrometry and photometry were used to measure the amount of calcium uptake. By default, the amount of calcium in tooth enamel is 4/37% of the weight of enamel, and the depth of the missing enamel was evaluated against this baseline; however, in reality, the amount of calcium in tooth enamel varies from person to person.[16]

West *et al*. and Ganss *et al*. used a profilometric method to investigate the effect of acid on the dental tissue. The amount of the roughness, height, and lowness of the surface of teeth was measured with a diamond probe. Regarding this result, important point to note is that contact with acid solutions first leads to a reduction in the microhardness of enamel, followed by the loss of surface tissue. Therefore, it is not an exact method to assess the erosion that is caused by changes in the microhardness of enamel.[12,17,18]

In this regard, Devlin *et al*. used the Vickers hardness method to investigate the effect of beverages on the hardness of tooth enamel. Samples were chosen from the healthy part of decayed teeth and were investigated with the Vickers hardness method. The results of that study showed greater loss of microhardness than the present study. The reasons could be the presence of intervening factors, such as the decayed teeth, from which the samples were chosen, and the effects of cutting the samples.[19] In the present study, healthy primary and permanent teeth were used to reduce the effects of intervening factors and the samples were kept in water without using factors such as heat, cutting, additional pressure, and antiseptic agents, such as sodium hypochlorite. At the time of testing, the teeth were kept for 5 min in the solutions and investigated at a temperature of 9°C (temperature of the beverages in the refrigerator).[20]

According to the average storage time in the mouth (20 s) during the daily consumption of soft drinks and beverages, 5 min can be considered reasonable for the daily use of non-alcoholic carbonated beverages. According to this criterion, the time (1, 2, 3, and 15 h) that was used in Delvin's study is more than the time (5 min) that was used in the present study; therefore, it could be another factor that caused a greater loss of microhardness in Delvin's study.[9,18,21]

Ajami *et al*. also used the Vickers hardness method. They used the enamel blocks for their testing, and their results showed a significant reduction of microhardness in both the permanent and primary tooth enamel after the teeth were immersed in beverages. In the present study, we found that the loss of microhardness in the permanent and primary tooth enamel, and the difference in the reduction, can be attributed to the sample preparation method. In the method reported by Ajami *et al*., the enamel was used without the support of dentin, which does not fit with the normal conditions of the human mouth.[10]

In another study, human models were used to try to bring the test situation as close as possible to the natural conditions of the oral cavity.[22]

It is recommended that additional studies be conducted by bringing the process of immersion in liquid closer to the conditions of the oral cavity and by reducing the immersion time to 20 s. Based on the results of this study, permanent and primary teeth that were immersed in Behnoosh Lemon Delster and Zam Zam cola beverage experienced significant reduction

in the microhardness of enamel. This reduction was greater in teeth that were immersed in Zam Zam Cola than those which were immersed in Lemon Delster; moreover, it was more significant in primary teeth than in permanent teeth.

In this study, we compared the microhardness of primary and permanent teeth after immersion in two types of beverages. One of the limitations of this study was the collection of teeth that met the inclusion criteria

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Conflicts of interest

There are no conflicts of interest.

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