Effect of fluoridated dentifrices on surface microhardness of the enameleroded by Coca Cola

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Abstract

Aims: The purpose of this study was to evaluate the microhardness on the enamel surface eroded by coca cola (An in vitro) treated with four different dental toothpastes after erosion.

Materials and Methods: 120 sound human maxillary first premolars extracted for orthodontic reasons were used in this study and randomly divided into 6 groups 20 teeth samples in each group:artificial saliva (control negative), coca cola then treated with artificial saliva(control positive). While the four remaining groups were eroded by coca cola then the samplestreated by four different dental Dentifrices [T1, T2, T3, and T4 then tested by indentation hardness test. The surface microhardness of the enamel surface was measured with a Vickers hardness tester at baseline, after erosion by coca cola and after remineralization by toothpaste.

Results: The data analyzed statistically using one way ANOVA and Duncan test ($p \le 0.05$) the mean surface microhardness in all groups decreased significantly after eroding with cola drink and increased after remineralization by artificial saliva and fluoridated dentifrice.

Conclusions: Coca Cola reduce the enamel microhardness while fluoridated toothpastes and artificial saliva increase microhardness of enamel, SnF2 dentifrice showed better and higherremineralization aftererosion when compared to other groups.

Key words: Cocacola, Erosion, Artifical saliva, Fluoridated Toothpaste, Microhardness.

Introduction

The human tooth is the hardest part of the human body, with a specific construction and constitution. It is composed of three distinct, highly mineralized and hard tissues (enamel, dentine, and cementum), and the enamel is the most mineralized part of the tooth and is harder than iron, providing the exposed crown with durable cutting and grinding surfaces, and the mature enamel consist of inorganic material makes up about 97 % and the rest is formed of proteins and other components, such as water (Jágr et al., 2014). Currently, it has been observed a significant increase in the prevalence of dental erosion as a consequence of frequent exposure to acid from foods, drinks and gastric juice(Liviaet al., 2013) especially if the chronic exposure to extrinsic/intrinsic acids with a low pH present(Magalhães et al., 2011). Dental erosion is the loss of dental hard tissues caused by nonbacterial acids. Due to acid contact, the tooth surface becomes softened and more prone to abrasion from tooth brushing (Ana et al., 2013). Erosion can be classified according to the source of acid, which could be either extrinsic or intrinsic. Extrinsic acids are mainly from diet or soft drink while example for intrinsic acids is gastroesophageal reflux. The erosion potential of the diet depends mainly on its pH, buffering capacity, titratable acidity, and drinking or consumption patterns (Ganss et al., 2012; Haifeng et al., 2012).

Demineralization of the tooth by erosion is caused by frequent contact between the tooth surface and acids present in soft drinks (Nikita et al., 2014). Lussi et al., (2011)who stated that dental erosion started with initial softening of the tooth surface followed by continuous layer-by-layer dissolution of the dental hard tissue crystals, leading to a permanent loss of tooth volume with a softened layer persisting at the surface of the remaining tissue. Dental erosion is a multifactorial condition. The consideration of chemical, biological and behavioral factors is fundamental for its prevention and therapy. Among the biological factors, saliva is one of the most important parameters in the protection against dental erosion. Saliva is supersaturated with respect to tooth mineral, providing calcium, phosphate and fluoride necessary for remineralization after an erosive challenge (Marilia et al., 2011). The erosion lesion can be divided in two phases: erosion in that there is only softening and erosive wear advanced phase with tooth surface loss (Huysmans et al., 2011; Shellis et al., 2011, Lussi et al., 2011). A more recent benefit provided by some dentifrices is protection against the initiation and progression of dental erosion (Hooper et al., 2007). Dentifrices containing different active

agents (like fluorides or agents with special anti-erosive properties) may be helpful in allowing rehardening or in increasing surface resistance to further acidic and giving some degree of protection against erosion (Ana et al., 2013).

Materials and methods

The experimental design of the study

Grouping the samples:

The total number of samples was (120) samples. This group was subdivided into (6) subgroups, which include:

- **1.** Group 1: control negative group N=20, the samples were immersed in a coca cola solution for 10 minutes at (20±1) C°, this cycle repeated three times daily for 3 days.
- Group 2: control positive group N=20, the samples immersed in the coca cola solution for 10 minutes at (20±1) C°, this cycle repeated three times daily for3 day. Then immersed in artificial saliva, for6 hours were stored at (37±1) C°.
- **3.** Group 3: T1 Dentifrice, Group 4:T2, Group 5:T3, Group 6:T4; N=20, the samples immersed in the coca cola solution for 10 minutes at (20±1) C°, this cycle repeated three times daily cycle for 3day. Then the enamel surface coated by a thin layer of T1 for 5 minutes repeating three times daily for 5 days.

Sample preparation:

Freshly human permanent maxillary first premolar extracted for orthodontic purpose was stored at 4°C in 0.1% buffered thymol solution (pH 7.0) until using it (Isabelaet al., 2011). The enamel was cleaned with non fluoridated pumice and white rubber prophylactic cup using the slow speed hand piece, wiped free of debris and rinse in tap water, then the crowns separated from the roots by using diamond discs then the crowns embedded in self cured acrylic resin. The labial surface was grounded wet using 400, 600, 1200, and 2000 grit silicon carbide paper to obtain flat and smooth surface.

Type 1 Dentifrice	1100 ppmSodium fluoride, silica abrasive
Type 2 Dentifrice	1450 ppm Sodium fluoride, silica abrasive
Type 3 Dentifrice	1450 ppm Sodium monofluorophosphate, silica abrasive
Type 4 Dentifrice	1450 ppm Stannous fluoride, silica abrasive

Active ingredients of fluoridated toothpastes

Artificial saliva composition

The artificial saliva used in this study as a remineralization solution with pH=6.9 and composed of the following:Potassium chloride, magnesium chloride, calcium chloride, dipotassium hydrogen phosphate, potassium dihydrogen phosphate, sodium carboxy methyl cellulose and deionized water(Amaechi et al 1999).

Measurement of enamel surface Microhardness

The surface microhardness (SMH) of the specimens was determined using a Vickers microhardness testing machine(OLPERT, Germany) with a Vickers diamond indenter and 600X objective lens. A load of 300 g was applied to the surface of the specimens for 15 seconds asshown in as shown in figure (1, A). Vicker hardness number (VHN) was measured from three points from each sample (Ambarkova et al., 2013) were equally placed over a circle of 1 mm diameter at the middle third of the specimens. After the diagonal length of the indentation measured by microscope the Vickers values as shown in figure (1, B) converted into microhardness values, and Vickers values converted into microhardness values by using the following equation: HV=1.854 P/d where HV is a Vicker hardness in Kgf/mm2 (Mpa), P is theload in Kgf and d is the length of diagonal in mm (Vesnaet al., 2011).



Figure (1): A: Vickers microhardness tester machine, B: The Vicker indenter

Statistical Analysis

One way analysis of variance and Duncan's multiple range tests ($P \le 0.05$) were performed to evaluate the differences on Microhardness among tested groups.

Results

Tested Groups	Mean±SD	Duncan groups
Control negative	188.35±2.641	Α
Control positive	254.80±3.105	В
T1	321.00±12.312	С
T2	319.50±19.795	С
Т3	336.60±4.751	D
T4	353.10±5.937	Е

 Table (1) Mean values, Standard Deviation and Duncan test among tested groups:

Table (2) demonstrated One way analysis of variance (ANOVA) test which showed that there was significantly different at $p \le 0.05$ of mean microhardness values among tested groups.

Table (2) ANOVA test for the differences on microhardness among tested groups:

Source of variance	Sum of Squares	df	Mean Square	F-value	P-value
Between Groups	387416.242	5	77483.248	752.434	0.000*
Within Groups	11739.350	114	102.977		
Total	399155.592	119			

* a Significant difference existed at $p \le 0.05$



Figure (2): Mean ± Standard deviation and Duncan's multiple range tests of microhardness values of teeth sample groups exposed to coca cola for the 3 day cycle.

Duncan test revealed that MH forT1 andT2 group not significantly different from each other, but both were significantly higher than control negative and control positive group and significantly lower thanT3 andT4. T4 treated group had SMH significantly highest other groups thenT3 treated group significantly higher thanT2,T1, control negative and control positive group, as shown in Table(1) and figure (2).

DISSCUSSION

Soft drinks have many potential health problems, including dental caries and enamel erosion (Majewski, 2001). The most frequent source of the acids is soft drinks like cola, and the acidity of coca cola is the main cause of erosion (Bowen and Lawrence, 2005). In vitro, erosion is greater than in situ erosion, precisely because the saliva protection factor is not present, even with the use of artificial or collected saliva (Westet al., 1999). The cross section microhardness test is widely used to study demineralization and/or remineralization in human teeth or in bovine teeth (Argenta et al., 2003; Queiroz, 2004; Paeset al., 2003; Delbem and Cury, 2002). Microhardness testing is considered to be a relatively simple and reliable method for the provision of indirect information on mineral content changes in enamel (Diamantiet al., 2010). The gain or loss of minerals in enamel as a result of demineralization and remineralisation process can be measured as hardness change. (Vesnaet al., 2011).

The baseline enamel microhardness value of this study was 300±28. VHN after erosion with coca cola, the mean microhardness reduced this may be due to low pH of coca cola which is 2.6 This comes into agreement with Hughes et al. (2000) who stated that the decrease in pH had been associated with increase in dental erosion also in agreement with Dugmore and Rock (2004) which revealed that carbonated drinks especially cola drinks were associated with dental erosion. The authors attributed the greatest erosive potential of the cola drinks to their low pH in comparison to the other non cola carbonated drinks, or due to contain of phosphoric acid, this is an agreement with Lodi et al. (2010) who studied the effect of different carbonated beverages on animals' teeth. Their results revealed that phosphoric acid was much erosive than citric, malic and tartaric acids, as the phosphoric acid has more ability to cheleate calcium. However, the results of the study disagreement by Attin et al. (2005) who revealed that the erosive potential of Sprite is more than that of Coca-Cola. They attributed this result to the severe demineralizing potential of citric acid than that of phosphoric acid.

Artificial Saliva is the most important biological factor affecting the progression of dental erosion, which increases the mean indentation enamel hardness after the demineralization by cola (Marília et al., 2011). The most important factors in the repair of softened enamel are saliva and fluoride. Acids softened enamel can reharden after exposure to saliva or to a remineralization solution and dietary substances and fluoride may enhance the remineralization process (Amaechi and Higham, 2001; Ganss et al., 2001). Calcium and phosphate, as well as an alkaline or neutral environment are prerequisites for remineralization. Calcium and phosphate levels in saliva act as common ions of the minerals in enamel and dentin, resulting in a slower dissolution rate of mineral (Meurman et al., 1996). Devlin et al., (2006) demonstrated that Coca Cola reduced the mean indentation hardness of enamel in the teeth, but the hardness was partially restored with artificial saliva. Lippert et al., (2004) stated that enamel rehardening by saliva was not observed. There are numerous in vitro studies demonstrating the ability of fluoride to protect enamel and dentine against acid attacks (Newbyet al., 2006; Featherstone et al., 1990) Several fluoride compounds are currently available in marketing

dentifrices, including sodium fluoride, sodium monofluorophosphate (MFP), stannous fluoride (SnF2), (Lippert et al., 2009).

In this study have shown that the use of fluoridated dentifrices effective in increasing enamel SMH after demineralization by cola. These findings indicate that the use of the marketed SnF2-based dentifrice provides greater protection against both the initiation and progression of dental erosion compared to the NaF and SMFP products used in these studies, this result come to agreement with Ganss et al., (2011) and Faller et al., (2011) have confirmed the superiority of SnF2 compared to NaF dentifrices to prevent dental erosion; however, its protective potential did not exceed the efficacy of NaF or AmF, (Magalhães et al., 2012), also this disagreement with Lussi et al., (2008) showed toothpaste on erosion failed to find a significant effect or difference when compared to a NaF-containing toothpaste. This study demonstrated directionally better toothpaste formulated with 1,450 ppm F compared with dentifrice formulated at 1,100 ppm F (NaF) (Eversole et al., 2014).

The 1,450 ppm F (as NaF) version of the product performed significantly better than its 1,100 ppm F (NaF) this come in agreement with; Although the results of an in vitro study by Ganss, et al. (2008) suggest considerable differences among NaF, AmF and SnF2, the impact of different fluoride compounds on erosion was not analyzed under clinical conditions yet. The efficacy of fluorides to affect de- and remineralization is related to its concentration and depends on the pH of the fluoride agent. The residual fluoride from dentifrice, present in saliva, had no significant effect on %SMHC (softening) and wear of eroded enamel by cola drinks in agreement with the studies of Lagerweij et al. (2006) and Ponduri et al. (2005), also this disagreement with (Ganss et al., 2011) been discussed that the amount of fluoride available in the dentifrice slurry is not directly related to the possible protective effect of dentifrices against enamel erosion.

CONCLUSIONS

- The hardness of human enamel decreased after exposures to cola drinks.
- Artificial saliva enhances the hardnessof demineralized enamel.
- Enamel reminerilized after uses of fluoridated toothpaste, butSnF2-based toothpaste provides greater protection against both the initiation and progression of dental erosion compared to the NaF and SMFP products used in these studies.

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