EFFECT OF ACIDS, FLUORIDES AND BRUSHING ON ENAMEL – AN IN-VITRO STUDY

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ABSTRACT

Objective: *To determine the effect of fluoride, citric acid and brushing on tooth enamel.*

Materials and Methods: *This in vitro experimental study was conducted on 60 extracted premolars at Khyber College of Dentistry and Experimental procedure was performed at laboratory, Physics Department, Peshawar University. All maxillary and mandibular premolar teeth with normal tooth morphology extracted for orthodontic reasons and of both genders were included. Premolar teeth restored with fillings or crowns, pathologically affected teeth with caries, Amelogenesis/Dentinogensis Imperfecta, root canal treated teeth or teeth with surface loss (attrition, abrasion, erosion) were excluded from study. The effect on enamel was determined in term of weight change.Paired t test was applied to see the statistical evidence for difference in weight while Independent samples t test was applied to see the effect of different concentration of fluorides at p-value ≤ 0.05.*

Results: *The weight of enamel decreased after immersion in citric acid (-4.134±139.34 mg), coke (-4.134±138.34 mg) and after brushing (-4.404±111.642 mg) statistically significantly (P<0.05). The increase in weight was statistically significant for 450 ppm fluoride (P=0.017) but insignificant after immersion in 225 ppm fluoride (P=.067).*

Conclusion: *Fluoride should be routine part of oral hygiene measures. In patients who are more prone to cariogenic insults, especially physically handicapped and undergoing orthodontic treatment. The increased concentration of fluoride is recommended to enhance remineralization.*

Keywords: *Acid, Fluoride, enamel, Brushing*

INTRODUCTION

The term tooth loss generally used to describe the loss of the hard tissues of teeth through causes other than caries, trauma or developmental disorders is tooth surface loss.[1] The etiology of the progress of tooth wear is multifactorial. More specifically, etiological factors may be divided into erosion, abrasion, abfraction and attrition.[2]

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Dental erosion is an irreversible loss of hard tissues of tooth due to chemical processes in which microorganisms are not involved.[3] Pathogenesis of dental erosion includes many acids including those regularly found in the human diet for example soft drinks and citric foods. It also includes all acids which originate in the stomach like gastric acids due to regurgitation as well as few drugs that are known to exacerbate erosion of teeth.[4] Early appreciation of dental erosion is imperative to prevent serious irreversible damage to the teeth.[5]

Tooth abrasion is the mechanical loss of tooth hard tissues due to physical causes except teeth.[6] It

would be an exaggeration to assumes that brushing of teeth is absolutely harmless and it has been established for a long time that tooth-brushing has some adverse effects on gingiva and tooth hard tissues. Two types of damage seem to predominate: gingival recession with root surface exposure, and cervical abrasion of cementum and dentin.[7]

Attrition is the loss of tooth substance due to the mechanical action of teeth such as during mastication or due to contact between opposing tooth surfaces. This pattern of dental surface loss can be especially noteworthy in vegetarian persons or persons taking a modern healthy food. Parafunctional habits may also cause attrition.[8] Pathological loss of tooth structure is called abfraction. Biomechanical forces like flexion, compression, or tension cause Abfraction. It may also be caused by chemical degradation; it is observable as V-shaped grooves in the cervical area of a tooth.[9]

The citric acid is used in many dietary forms and can be considered the important part of modern to combat the issue of vitamin C deficiency in prevention of scurvy disease. It is considered deleterious only when PH is below 5.5 leading to mineral loss from enamel.[10] Due to different PH of various cold drinks its erosive activity varies. Among various cold drink coca cola have highest erosion rate.[11]

The action of diet in erosion of the enamel has been widely studied, and as such there are several points of contention among researchers. In 2010 an in vitro study conducted by Kenneth Eagle and Anderson T. Hara found that bleaching the enamel and dentine in 10% carbamide peroxide does not enhance their erosive and abrasive susceptibility. Instead the study found that bleaching did not enhance the sensitivity of enamel to erosive and abrasive tooth surface loss.[12] Similarly, another study in 2014 showed that abrasion greatly depends upon the type of toothbrush bristles used to clean the teeth.[13] On the other hand, in 2006 C.A Hemingway et al found that loss of enamel by erosion is decided by the chemical composition of the erosive medium. The study employed fruit juices with different acidity levels and found that all the media induced erosion in tooth enamel to varying degrees.[4] Another study in 2002 also determined that acidic drinks, both carbonated and fresh, induce erosions in teeth enamel. The study found that drinks above pH 3 induced erosions that could not be repaired by calcium flouride levels of up to 20 ppm[14]

This study helping to determine the effect of acidic dietary fluids on erosion of enamel surface and effect of fluoride as a remineralizing agent. This study is also aiding determine the effect of brushing upon remineralization. This study also allows future researchers to further examine the effect of diet on the health of dental hard tissue. It also provide a basis for practitioners to better preserve the health of teeth as well as increase awareness of oral health among public following the decline in the caries rate. This in turn help public oral health.

The objective of this study was to determine the effect of fluoride, citric acid and brushing on enamel.

MATERIALS AND METHODS

This in vitro experimental study was conducted on 60 extracted premolars at Khyber College of Dentistry and Experimental procedure was performed at laboratory, Physics Department, Peshawar University. All maxillary and mandibular premolar teeth with normal tooth morphology extracted for orthodontic reasons and of both genders were included. Premolar teeth restored with fillings or crowns, pathologically affected teeth like caries, Amelogenesis/Dentinogensis Imperfecta, root canal treated teeth or teeth with surface loss (attrition, abrasion, erosion) were excluded from study.

The extracted premolars were stored and immersed in distilled water to prevent dehydration. The effect on enamel was determined in term of weight change. An electronic precision and analytical balance sensitive to 0.0001g (KERN & Sohn, GmbH, D-72336 Balingen, Germany) was used to weigh the premolar teeth. For ten day the premolar were placed in various media for two minutes twice a day and change in weight was measured. The average weight was recorded for each tooth.

The 60 teeth were randomly divided into 4 groups as follows: Group citric acid (n=12), Group carbonated drink (n=12), brushing group (n=12) and group fluoride (n=24). Fluoride were further divided into two groups (225 ppm and 450 ppm)

1. Group citric acid (CA): 12 teeth were submerged in 120 millilitre of 1% citric acid solution (citric acid monohydrate, batch #K15695742, Merck) for 10 days (2 minutes each day) and then weighted.

2. Group carbonated drink (CD): 12 teeth were submerged in 120 millilitre of a carbonated drink (Coca Cola) for 10 days (2 minutes each day) and then weighted.
3. Group Brushing; Tooth surfaces (buccal, mesial, lingual, distal and occlusal) of 12 teeth were subjected to 20 brushing strokes with manual toothbrush per day (Oral B toothbrush, medium bristles) for 10 days. Each stroke means forward and backward movement of brush on a specific tooth surface. After brushing the teeth were washed with 10 ml of distilled water through water jet from a syringe, dried with absorbent paper and weighed. Each surface of tooth received 20 strokes. Each tooth was brushed for 2 minutes
4. Group F225; 12 teeth were submerged in fluoride solution containing 225 ppm of sodium fluoride (Colgate Plax Antibacterial Mouthwash, sodium fluoride 225 ppm) for 10 days (2 minutes each day) and then weighted.
5. Group F450: 12 teeth were submerged in fluoride solution containing 450 ppm sodium fluoride (Sensodyne Proenamel Daily Mouthwash, sodium fluoride 450 ppm) for 10 days (2 minutes each day) and then weighted.

Data collected through a structured proforma were analyzed through SPSS version 20.0. Mean, standard deviation and confidence interval were calculated for numerical variables like weight of teeth. Paired t test was applied to see the statistical evidence for difference in weight between original weight of the teeth and after immersion in citric acid, fluoride for total of 10 days and after brushing. Independent samples t test was applied to see the effect of different concentration of fluorides i.e. 225 ppm vs 450 ppm. P<0.005 was considered significant.

RESULTS

The distribution of various teeth in various groups is given in figure 1. Each group contains 12(20%) teeth.

The weight of enamel decreased after immersion in citric acid, coke and after brushing. The mean reduction in weight after immersion in citric acid was -4.134±139.341 milligrams (mg), in coke was -4.134±138.34 mg, and after brushing was -4.404±111.642 mg. The decrease in weight was statistically significant for citric acid (95 %CI=-3.2, -5.61, P<0.001), coke(95 %CI=3.13, 7.34, P<0.001) and brushing (95 %CI=-3.12, -5.41, P=0.003). After immersion in 450 ppm fluoride (11.2±13.7 mg) the increase in mean weight was found. The increase in weight was statistically significant for 450 ppm fluoride (95 %CI=2.435,19.965, P=0.017). However the increase in weight of enamel after immersion in 225 ppm fluoride was not statistically significant (95 %CI=-.665, 16.86, P=.067). The details are shown in the Table 1.

When weight of enamel after immersion in 450 ppm and 225 ppm of fluoride was compared, the difference in results was statistically was not significant (P=.0.57). The details are shown in the table 2.

DISCUSSION

The teeth were selected in this study from ex-

Table: 1 Comparison of change in weight after immersion in citric acid, fluoride and after brushing

Procedure	Mean ± SD difference weight (mg)	95% CI	P-value
After immersion in citric acid (n=12)	-4.134±139.341	-3.2, -5.61	<0.001
After brushing (n=12)	-4.404±111.642	-3.12, -5.41	0.003
After coke (n=12)	-4.134±138.34	3.13, 7.34	<0.001
225ppm fluoride (n=12)	8.100±13.81	-.665 16.86	.067
450 ppm fluoride (n=12)	11.2±13.7	2.435, 19.965	0.017

*paired t test; CI, confident interval; ppm, parts per million

Table: 2 Comparison of change in weight after immersion in various concentrations of fluoride

Concentration (ppm)	Mean ± SD (mg)	Mean Diff (mg)	P-value	95% CI
225 (n=12)	8.100±13.81	3.200	0.57	-8.40 to 14.81
450 (n=12)	11.2±13.7			

* Independent t test

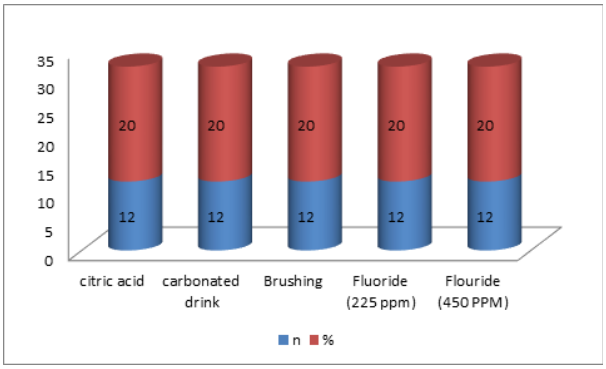

Fig 1: Number of teeth in various categories

tracted premolar teeth at the Department of Oral Surgery, Khyber College of Dentistry, Peshawar. This source provided an easy access to collect a large number of premolars because in Khyber College of Dentistry, a large number of patients are coming for Orthodontics treatment. It has been proposed that a material of newly extracted teeth with a known history and a clinically more genuine enamel surface are preferable.[15] It was decided to use both maxillary and mandibular premolars; these teeth often have a relatively short exposure time to the oral environment since they may be extracted at an early age for orthodontic treatment. Another possibility would have been to use bovine teeth. However, there are differences in structure and in behavior towards clinically related procedures between human and bovine enamel.[15] Since the experimental procedures in the present study are of a clinical nature, we decided to use human teeth.

In the present study, it was clinically done that any tooth which had the sign of wear, erosion and caries were excluded from the selected sample to purely checked the effect of acid, fluoride and brushing on enamel. The results of the current research showed that the surface condition of the premolars before the experimental procedures of brushing and citric immersion was variable with respect to wear, clarity of perikymatae, distinctness of prism ends, distribution of prism free enamel, and presence of a surface coating. The premolar teeth in the this study were selected based on clinical/macroscopic observation. In hindsight, it would have been advantageous to observe the specimens under a dissecting microscope during the selection of the premolar teeth for a better evaluation of the surface condition prior to the treatments.[16]

Most soft drinks are acidic in nature and exposure to these drinks may result in enamel erosion.[17] In the current study, effect of carbonated drink (coke) on the enamel was studied. The effect was measured in term of weight change. When the whole sample was analyzed for the change in weight after immersion in coke and was highly significant (P<0.001). For ten day the premolar were placed in coke for two minutes twice a day and change in weight was measured. Most the change in weight was small but was statistically significant.

A study by Jain et al[18] determine the pH for twenty available brands in the market of soft drinks and the enamel dissolution happens because of immersion in these soft drinks, and the effect of pH on loss of enamel. They concluded that prolonged exposure to soft drinks could lead to significant enamel loss. Our study also showed there is loss of weight of enamel upon exposure to coke/soft drinks. However, the current study is different from the Jain P's study because in our study the effect on enamel was measured in term of weight change while in their study, effect on enamel was measured in term of pH change.

Fluoride(F-) is the negative ion of the fluorine element. Fluorine has largest electronegativity. That is why is very reactive and very rare to found in atomic form and occur as fluoride.[19] Stomach pH is less than the hydrofluoric acid pH so fluoride is fully dissolvable here. In mineralized dental and bone tissues some of the fluoride is become the part of the crystallites, but it can also be more commonly present on crystal surfaces or in their hydration shells. Conventional fluorides in the form of sodium fluoride.[20] Fluoride can stabilized lattice by attracting protons of nearby hydroxy-apatite.[21] It has been reported that fluoride can lead to mineralization defects and

declines enamel hardness. This recommends that fluoride may cause alteration in crystal size, number, shape, or quality by interfering with their formation, as reported for bone crystals.[22]

Previous studies showed that fluoride can remineralized the enamel adequately.[23] In the current study, the effect of fluoride on the enamel was observed after immersion of premolars teeth in sodium fluoride solution of two concentrations i.e. 225 PPM and 450 PMM. These teeth were kept immersed in fluoride solution. The current showed that the change in weight after immersion of teeth in sodium fluoride solution was statistically significant (P<0.05). Stratification for 225 PPM versus 450 PPM showed that 450 PPM was statistically more effective than 225 PPM (P<0.05).

A study by Muhammad et al[24] who determined the effects of fluoride concentration on in-vitro enamel demineralization during exposure to caries-simulating conditions using Scanning Microradiography (SEM). The influence of serially increasing fluoride concentration (0.1–4500 mg/L F-) in the acidic solutions was calculated on the rate of demineralization of enamel. Their results showed that logarithmic linear relationship exist between fluoride concentrations and decrease in demineralization upto 135 mg/L [F−]. Above this level, no further reduction in demineralization was found. Although the methodology of Muhammad et al[24] study is different from the current study but the results are similar that as fluoride concentration increases the loss of mineral from the enamel is decreases.

In the current study, the change in weight after brushing was significant. A study by Waaler et al[25] that after one minute brushing significant change in enamel was found. Waaler et al[25] also used micrograph. On other hand Eisenburger et al[26] found that there was no difference in enamel wear after 12 brushing strokes compared to 500.

It is an in-vitro study, it may not replicate actual oral environment because of the effect saliva and oral heat. So well controlled and large sample in-vivo study is required to further clarify this issue.

CONCLUSION

Enamel lost occurred when exposed to citric acid, coke and brushing at statistically significant level (P<0.05). Fluoride increases enamel weight statistically significant (P<0.05). So, fluoride should be routine part of oral hygiene measures. In patients who are more prone to cariogenic insults, especially physically handicapped and undergoing orthodontic treatment increased concentration of fluoride is recommended to enhance remineralization. Also aggressive brushing should be avoided to preserve enamel. Acidogenic items should be minimized.

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