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Comparative Evaluation of Remineralization Capacity Of Two Commercially Available Toothpaste On The Microhardness Of Bleached Enamel Surface: An In Vitro Study

[1]Mahendra Patel, Professor & Head, College of Dental Sciences & Research Centre, Ahmedabad, Gujarat.

[2]Bhavika Bhavsar, Professor, College of Dental Sciences & Research Centre, Ahmedabad, Gujarat.

[3]Ashutosh Kosada, Post graduate, College of Dental Sciences & Research Centre, Ahmedabad, Gujarat.

[4]Nishtha Patel, Reader, College of Dental Sciences & Research Centre, Ahmedabad, Gujarat.

Correspondence Author: Ashutosh Kosada, Post graduate, College of Dental Sciences & Research Centre, Ahmedabad, Gujarat.

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Abstract

Introduction: Bleaching is method of treating tooth discoloration. But it causes some adverse effect like mineral loss from the tooth structure and decreased hardness of the enamel. To overcome this loss of minerals and subsequent microhardness of enamel remineralizing toothpaste are recommended. The aim of the study was to evaluation of remineralization capacity of two commercially avaliable toothpaste on the microhardness of bleached enamel surface.

Methodology: Thirty freshly extracted, sound maxillary incisors were selected and their roots were removed. Then, each tooth crown was embedded in self-cured acrylic resin molds. Specimen were stored in artificial saliva. Initial hardness was measured using vickers microhardness indenter and the teeth are randomly divided into two groups. 22% carbamide peroxide bleaching agent was used to demineralize the teeth and again microhardness was measured. Teeth in group 1(n=15) and group 2(n=15) were treated with Enafix and Sensodyne repair and protect toothpaste for seven days and again microhardness of enamel was measured.

Results and Statistical Analysis: Microhardness decreased in both the groups after bleaching application (% change group1 was 2.34% and group2 was 2.27%). Both remineralizing toothpaste significantly increased mineralization (% change group 1 was 2.73% and group 2 was 3.94%). Test was done using ANOVA with Scheffe's post hoc test and p-value <0.05 was considered as statistically significant. However, difference was not significant(P >0.05).

Conclusion: Both Sensodyne repair and protect and Enafix toothpaste increases microhardness of bleached enamel but Sensodyne repair and protect is more effective for increasing microhardness.

Keywords: Bleaching ,enamel, microhardnes, remineralization.

Introduction

Discoloration of anterior teeth is an esthetic problem that requires effective treatment. Vital bleaching is a viable option to consider when treating intrinsically stained or discolored teeth whose form and integrity are deemed acceptable. Carbamide peroxide introduced by Haywood and Heyman[1] was most widely used for tooth whitening (bleaching) both in professional and in self-administered products. Though the decrease in discoloration has been satisfactory with the application of bleaching agents, there were also adverse effects as the consequence of this

treatment.[2] Prominent side effects of concern included pulpal sensitivity,[3]surface compositional changes,[4]mineral loss from the tooth structure[5] and decreased hardness of the enamel.[6] The decrease in the micro-hardness number has a linear relationship with the mineral loss under the conditions of demineralization.[7] Increased frequency of acid exposure in bleaching tends to alter the total demineralization/ remineralization amounts, resulting in significantly greater amounts of mineral loss.[8] To overcome this loss of minerals and subsequent micro-hardness remineralizing solutions are recommended.[9] Studies have reported that the use of agents such as fluoride and casein phosphopeptide-amorphous calcium phosphate (CPP-ACP) promote remineralization of tooth structure.[10]

The oral fluid mainly comprising of saliva and gingival crevicular fluid is rich in calcium, phosphate and fluoride ions. This results in a dynamic equilibrium between the mineral content of tooth and oral fluid in a neutral pH. In conditions when the pH is neutral, there is the minimal dissolution of hydroxyapatite crystals releasing calcium , phosphate and hydroxyl ions that are already present in the oral fluid. Increase in calcium and phosphate ions in the oral fluid lead to supersaturation of the solution and precipitation of the minerals back onto the tooth surface that can be termed as remineralization. In conditions when the pH of the oral fluid is acidic especially at or below 5.5 termed as critical pH, it becomes undersaturated with respect to phosphate ions . This results in dissolution of hydroxyapatite crystals from the tooth to resaturate the oral fluid, which is termed as demineralization.[11]

The aim of this in vitro study was to evaluate and compare the remineralizing efficacy of two commercially available toothpaste (**Enafix and Sensodyne repair & protect**) on the microhardness of bleached (22%carbamide peroxide) enamel surface.

Materials and Methods

Sample Preparation

Thirty freshly extracted, sound maxillary incisors were selected and their roots were removed. Then, each tooth crown was embedded in self-cured acrylic resin molds, with the labial surface leveled on top and lying flat and parallel to the horizontal plan upon which a 3 mm × 3 mm surface was marked and subjected to the application of bleaching agent and remineralizing agents. The specimens were then kept in artificial saliva to prevent dehydration. The samples were rinsed in water and dabbed dry before subjecting them for baseline hardness test. The baseline microhardness measurements were taken on the labial surface by means of a Vickers indenter with 100 g of force for 30 s by focusing at 10× objective lens, two indentations on each sample away from edge were made and the average of two readings were taken as baseline microhardness measurement.

Materials Used for Preparing Artificial Saliva

Artificial saliva was prepared in Department of biochemistry ,College of dental science and research centre, Ahmedabad, Gujarat. Hence, this study was conducted using artificial saliva to simulate clinical conditions.

- Calcium chloride 0.22g/l
- Sodium phosphate 1.07g/l
- Sodium bicarbonate 1.68/l
- Sodium azide 2g/l
- Distilled water one liter

Procedure for Microhardness Test

Microhardness was tested using Vickers microhardness tester. The test specimens were placed on the stage of the tester and stabilized. Then area to indent was selected by focusing with 10× objective lens. After this, the test was carried out where the indentations were made with a rate of 100 g load for 30 s. The indentation formed was

viewed and measured on the display monitor with 10× objective lens. The average microhardness of the specimen was determined from two indentations to avoid any discrepancy. The procedure was repeated for all the twenty specimens.

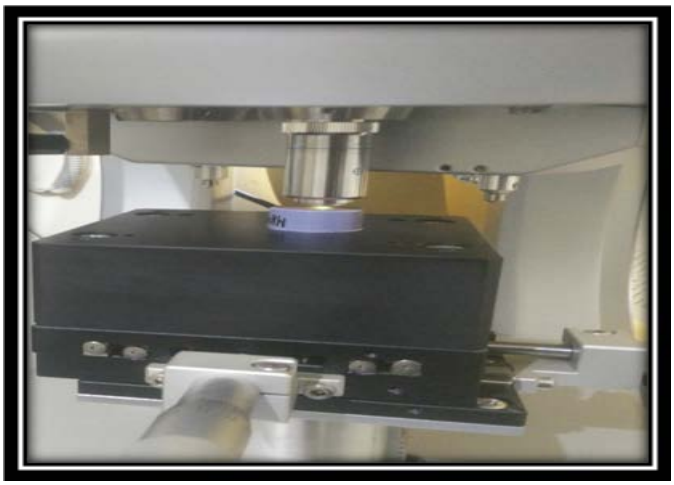

Figure 1: Samples are mounted on Vickers microhardness tester

Preparation of Bleaching Agent

Samples are exposed to 22% carbamide peroxide for 2 h daily for 1-week. After the recommendation time of exposure, daily the teeth were washed with distilled water and placed in artificial saliva for the reminder of 24 h. The procedure was carried out for all specimens for a period of 1-week. The samples were rinsed in water and dabbed dry before subjecting them for hardness test. Again Vickers microhardness test carried out for all the samples and values were recorded.

Figure 2: 22% carbamide peroxide

Figure 3: Bleaching agent application

Remineralization Procedure

The teeth were randomly divided into two group

Figure 4: Remineralizing toothpaste application

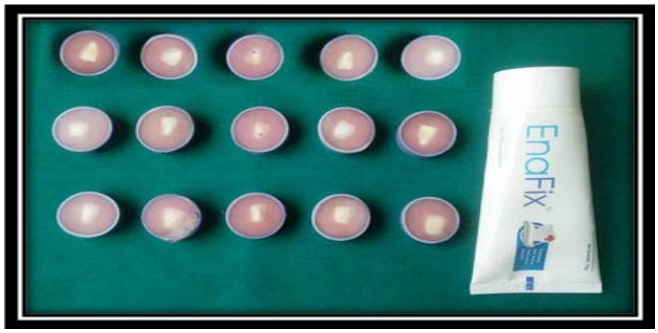

Figure 5: Teeth treated with Enafix toothpaste(Group 1)

Figure 6: Teeth treated with Sensodyne repair & protect toothpaste (Group 2)

Group 1: Enafix toothpaste was applied with cotton applicator tips on the enamel surface of post bleached samples, everyday for seven days with minimum application time of 3-5 minutes. The samples were then washed under water, stored in artificial saliva for seven days after which the samples were tested for microhardness and the values were recorded as described earlier.

Group 2: Sensodyne repair & protect toothpaste was applied with cotton applicator tips on the enamel surface of post bleached samples, everyday for seven days with minimum application time of 3-5 minutes. The samples were then washed under water, stored in artificial saliva for seven days after which the samples were tested for microhardness and the values were recorded as described earlier.

At the end of 7 days, the samples were washed with distilled water and blotted dry. Again the microhardness of the enamel surfaces was measured with the Vickers indenter as previously.The recorded values are subjected to statistical analysis.

Statistical Analysis

The test for significance from initial, after bleaching and after 1-week remineralization was done using **ANOVA with Scheffe's post hoc test** and *p*-value <0.05 was considered as statistically significant.Data analysis was done using statistical package for social science (SPSS 16.0).

Results

A significant decrease in the mean values of microhardness in hardness values (HV) (100 g Load) from initial to after bleaching was seen in both groups [Table 1; P < 0.05]. There was no significant difference in the mean values of microhardness in HV (100 g Load) after bleaching between two groups (P > 0.05). Both groups showed a significant increase in the mean values of microhardness from after bleaching for to after 1-week remineralization. Microhardness decreased in both the groups after bleaching application (% change group1 was 2.34% and group2 was 2.27%) Both remineralizing toothpaste significantly increased mineralization (% change group 1 was 2.73% and group 2 was 3.94%)[table 2]. However, the difference in the mean values of microhardness in HV (100 g load) after 1-week remineralization in group one and group two was not significant (P > 0.05).

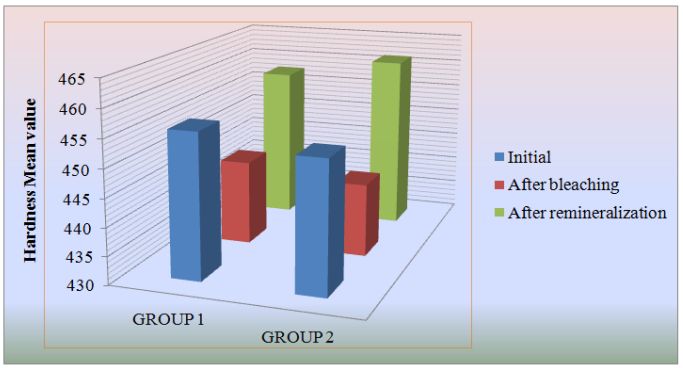

Table 1: Mean hardness at different time intervals.

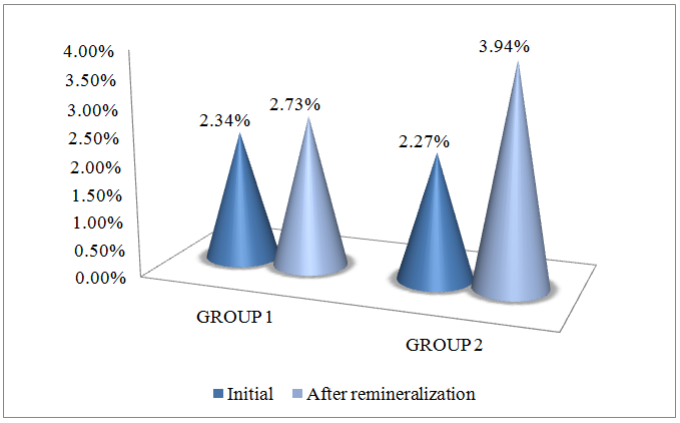

Table 2: Percentage change in microhardness in hardness value (100 g load) after 1-week remineralization

Discussion

Bleaching procedure has been known to alter the enamel matrix because of the pH and strong oxidizing effect of hydrogen peroxide.[12,13] Carbamide peroxide is an adduct of urea and hydrogen peroxide which on contact with water breaks down to urea and hydrogen peroxide. The

chemistry of this agent is based primarily on its ability to generate free radicals in most solvents. The hydroxyl radicals lack one electron, are extremely electrophilic and unstable and will attack most other organic molecules to achieve stability generating other radicals.[14] Hydrogen peroxide bleaching can proceed via the perhydroxyl anion or free radicals depending on pH. As the peroxide diffuses into the tooth, it can react with organic color materials found within the tooth structure leading to a reduction in color. These oxidation reactions can cause alterations in the enamel structure,calcium and phosphate mineral loss, and pulpal sensitivity.[15]

The oral environment can naturally rebuild enamel through a process called as remineralization, to some extent on its own, but the extent of remineralization is controlled by pH and the amount and availability of calcium and phosphate ions.[16]We used artificial saliva for storing the specimens in between bleaching cycles as studies have shown that by doing this there is a slight increase in microhardness after demineralization.[17,18]

Enafix has main remineralizing agent is calcium sucrose phosphate(CaSP). CaSP is a combination of calcium salts of sucrose phosphate esters, blended with inorganic calcium. It soon breaks down into calcium ions, phosphate ions, and sucrose phosphate ions into saliva. It is composed of 10–12% calcium and 8–10% phosphorous (by weight). CaSP permits creation of aqueous solution consisting of very high concentration of calcium and phosphate without occurrence of precipitation. It acts as an ideal carrier for calcium and phosphate. The mechanism of action is by adsorption of sucrose phosphate ions rapidly on the enamel surface, reducing the rate of acid dissolution of hydroxyapatite and quick remineralization by calcium and phosphate ions by common ion effect. Calcium sucrose phosphate decreases tooth enamel demineralization and promotes enamel remineralization. It also inhibits the formation of plaque[19,20]

Sensodyne repair and protect has main remineralizing agent is novamin.Novamin adheres to exposed dentin surface and forms a mineralized layer that is mechanically strong and resistant to acid. There is continuous release of calcium over time, which maintains the protective effects on dentin.[21]When novamin comes in contact with saliva, it rapidly releases sodium, calcium, and phosphorous ions into the saliva that are available for remineralization of the tooth surface. The ions released form hydroxycarbonate apatite (HCA) directly. They also attach to the tooth surface and continue to release ions and remineralize the tooth surface after the initial application. These particles will release the ions and transform into HCA for up to 2 weeks. So these particles will completely transform into HCA.[22]

The comparative studies between Enafix and Sensodyne repair and protect have not been done. So we compared these two Formulations for evaluating their remineralization potential. Microhardness of enamel was significantly and similarly reduced after bleaching in both groups. After bleaching the test products were applied and remineralizing effects of Enafix and Sensodyne repair and protect were compared. Significant improvement in the microhardness was seen after 1-week remineralization in both the groups, confirming usefulness of both agents.Enafix showed a numerically lower increase in remineralization after 1-week remineralization compared to Sensodyne repair and protect. Sensodyne repair and protect is an additional option for improving remineralization in the patients undergoing bleaching.

According to our knowledge, this is the first study comparing remineralizing potential of Enafix versus Sensodyne repair and protect. Kumar et al[23], did an in vitro study to investigate the remineralizing potential of

Casein phosphopeptide amorphous calcium phosphate (CPP-ACP) and bioactive glass(BAG), they found that, both group samples made plugs that sealed the more angular deposits, which appeared to be more compact and intimately attached to the enamel surface on scanning electron microscope analysis. The deposits formed by CPP-ACP in comparison were smaller and amorphous fissures formed by demineralization. BAG plugs created larger, This may explain the high values of hardness for Novamin as compared to Calcium sucrose phosphate in the current study.

Remineralization in the oral cavity is a complex procedure involving a change in pH and replenishment of calcium and phosphate elements. This may not be achieved in the in vitro conditions. We recommend further studies using these products in in vivo conditions. Scanning electron microscopy of enamel surface might add more value to the results. One of the factors, which may alter the hardness, was preparation of specimens, because any tilt or not flat surface may yield a too large indentation and thus a smaller Vickers hardness measurement. Hence two indentations were made to avoid any operational bias and then the average of these two indentations was taken for statistical analysis.

Conclusion

From the above study it can be concluded that, carbamide peroxide bleaching gels had a definite effect of reducing the surface micro-hardness of sound enamel to significant levels. But application of remineralizing toothpaste can remineralize the enamel surface and increase microhardness of enamel to some extent. Both Enafix and Sensodyne repair and protect toothpaste increases microhardness of bleached enamel but Sensodyne repair and protect is more effective for increasing microhardness of bleached enamel. Remineralizing toothpaste is an additional option for improving remineralization in the patients undergoing bleaching.

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