EFFECT OF DESENSITIZING OTC TOOTHPASTE PRODUCTS VERSUS FLUORIDE IONTOPHORESIS ON MICROHARDNESS OF DENTIN SURFACE

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ABSTRACT

Objective: This study was designed to compare the effect desensitizing OTC toothpaste products versus fluoride iontophoresis on dentin surface microhardness. Materials and methods: Eighty four extracted human teeth were stored in distilled water at room temperature until use. The teeth were divided according to the remineralizing agents into four treatment groups (Bioactive glass B; n-HAP N; fluoride F & fluoride iontophoresis Fl) 21 teeth each. Each group was further subdivided into three equal subgroups according to the pH cycling period (two weeks (W2), one month (M1) and two months (M2) with 7 teeth for each. Results: The results in comparison to demineralized specimen revealed that Fl group the highest mean surface microhardness value, while the B, N & F groups showed lower microhardness mean values with significant difference. On the other hand, there is an increase in surface microhardness values of B, N, F groups with non-significant difference. Conclusions: Microhardness of dentin is positively affected by the application of remineralizing agent and better with fluoride iontophoresis. Increasing the application rate (pH cycling periods) of the remineralizing agents significantly positively affect the surface microhardness of dentin.

KEY WORDS: Fluoride Iontophoresis, Remineralization, Bioactive glass, Nano-hydroxyapatite

INTRODUCTION

Dental caries is the most common oral disease. The conventional treatment approach for all caries affected teeth includes removal of carious tissues and placement of a restorative material. However, a contemporary approach has been adopted late by the non-invasive intervention of carious lesions. Non-cavitated as well as carious lesions extending up to dentin– enamel junction can be arrested if the cariogenic challenge of the lesion microenvironment is sufficiently controlled or therapeutic agents are applied \(^{(12,13)}\).

Fluoride has been useful as one of the most effective remineralizing agents in dental caries prevention. Therefore, it is necessary to look for alternative, effective non-fluoride agents that help to arrest caries. These recently developed remineralization technologies; include several commercialized calcium-phosphate based systems that have ions in the vicinity of hard tissue. Therefore, it has been suggested to promote mineral regain process in apatite forms \(^{(3)}\).

NovaMin is an ingredient made of bio-active glass particles with a size smaller than 20 microns. It has been assessed for its efficacy in remineralizing dental hard structure while occluding dentinal tubules\(^{(12,13)}\). Nano-sized particles (n-HA) are like the apatite crystal of tooth crystal structure,
crystallinity and morphology (6). Recently, several reports revealed that Nano-hydroxyapatite can remineralize the artificial carious lesions after being added to toothpastes and mouthwashes (7,8).

Manufacturing companies have stated that fluoride iontophoresis (FI) has an effect not only on dentin hypersensitivity but also on caries prevention (9). Consequently, commercial FI devices have been utilized in Korean children with a high-caries risk. However, there is inadequate scientific evidence to sustain the superiority of FI over the conventional over the counter (OTC) products for desensitization of dentin (10). From the previous review and with the existence of few reports on comparison between those agents so, evaluating the effect of these different remineralizing agents and device on surface microhardness of dentin would be helpful to clarify this important aspect of minimal intervention dentistry.

Therefore, the aim of the current study was to compare the remineralization effects through microhardness of FI and OTC. The hypothesis of this study was that FI would have a better remineralizing effect than OTC in topical applications in vitro.

**SUBJECTS AND METHODS**

Three OTC products of remineralizing agents were used in this study, and as are followed:

1. Bioactive glass remineralizing agent in the form of BioMin (calcium sodium phosphosilicate) toothpaste.
2. Nano hydroxyapatite (n-HAP) in the form of Apagard toothpaste.
3. Sodium fluoride in the form of Topex Neutral pH paste.

As well as using the iontophoresis device with acidulated phosphate fluoride (APF), (figure 1). In addition; three additional solutions were prepared: Artificial saliva (0.0016g Na$_2$S, 0.0016g Mg$_2$P$_2$O$_7$, +0.4g NaCl, 0.4g KCl, 0.6g CaCl$_2$, 0.6g NaH$_2$PO$_4$, 4 g Urea, 4 g Mucin, 1L distilled water at pH 7), demineralizing solution (2.2 mM KH$_2$PO$_4$, 2.2 mM CaCl$_2$, 0.05M acetic acid the pH adjusted to 5 with 1 M KOH.) & remineralizing solution (0.9mM NaH$_2$PO$_4$, 1.5mM CaCl$_2$, 0.15M KCl and the pH was adjusted using 5M KOH to 7.0 pH) in Faculty of Pharmacy, Al-Azhar University.

This research was designed to compare the effect of OTC remineralizing products (Bioactive glass, n-HAP & NaF) versus fluoride induction with iontophoresis device on surface microhardness of dentin. A total number of 84 sound non carious human molars were used in this study. Teeth were stored in distilled water at room temperature until use. The teeth were divided into four treatment groups (Bioactive glass $B$, n-HAP $N$, $F$ & $FI$) 21 teeth each. Each group was further subdivided into three equal subgroups according to the pH cycling period (two weeks (W2), one month (M1) and two months (M2) with 7 teeth for each.

All specimens were immersed in demineralization agent for 3 days. Then start to mimic the oral environment (pH cycling) by immersion in re/demineralizing solutions (1$^{st}$ surface treatment, 2 hours remineralization, 2$^{nd}$ surface treatment, 3 hours demineralization, 3$^{rd}$ surface treatment, 2 hours remineralization then artificial saliva overnight). Surface
treatment was done for each respective remineralizing agent as follow: group B the surfaces were brushed with BioMin toothpaste containing NovaMin (bioactive glass) for 3 minutes with artificial saliva and pH cycling and then immersed in artificial saliva. In group N the surfaces were brushed with Apagard paste containing Nano hydroxyapatite for 3 minutes and pH cycling then immersed in artificial saliva. group F the surfaces were brushed with toothpaste containing sodium fluoride for 3 minutes then pH cycling and immersed in artificial saliva.

While in group FI; Fluoride iontophoresis device (Jonofluor, Medical, Via Olivera, Italy) and its Fluoride gel APF was applied. This device consists of; A plastic tray with metal plate in the fitting surface of the try extended above the handle and a disposable grooved sponge adapted in the try that act as a carrier for the fluoride gel. Two electrodes; the positive anode and the negative cathode. Milliampere (mA) monitor display range from (0.2mA-0.4 mA).

A 1.23% acidulated phosphate fluoride (APF) gel 12300 p.p.m. F, pH 3.5 was applied according to manufacturer’s instructions, and electrically charged with 0.4mA, 12V for 4min. Then specimens undergo to pH cycling then immersed in artificial saliva. Artificial saliva was changed daily for two months in the previous protocol; prior each spectrum taken the specimen’s surfaces was rinsed with distilled water.

For assessment of surface microhardness; each tooth was vertically embedded into a special designed mold using self-curing acrylic resin. The occlusal surfaces were ground flat to about 1.5mm to expose the dentin surface. The remineralized dentin specimens in the three different subgroups were examined five times (at base line, after demineralization and after two weeks, one month and two months from the first time of treatment application) by Vickers microhardness testing machine.

RESULTS

A. Microhardness of base line and demineralizing groups:

There was statistically significance at P ≤ 0.05, after demineralization, between base line group (BL) (64.53±1.61) and demineralizing group (DeM) (50.95±1.51), which show statistically significant decrease in surface microhardness, as seen in table (1) and figure (1).

| Remineralizing Material | pH cycling | BAG | n-HAP | NF | IF | P-value |
|-------------------------|------------|-----|-------|----|----|---------|
|                         | Mean ±SD   | Mean ±SD | Mean ±SD | Mean ±SD |    |
| Baseline                | 62.62±1.29 | 64.35±0.73 | 64.48±1.64 | 64.97±1.42 | >0.05ns |
| Demineralized           | 50.66±0.80 | 50.87±0.63 | 50.11±2.54 | 50.88±1.86 | >0.05ns |
| p-value                 | <0.05*     | <0.05* | <0.05* | <0.05* |      |

*: significant (p<0.05)  ns; non-significant (p>0.05)
FIG (2) Bar chart representing microhardness of base line and de-mineralized groups with different remineralizing agents.

B - Effect of remineralizing agents on microhardness:

Regarding all of pH cycle periods (W2, M1&M2); all groups revealed increase microhardness value in comparison to demineralized group. Also, there was statistically significance difference between B, N&F groups on one hand and IF group on the other hand which showed the highest mean value of microhardness On the other hand; there was not statistically significance difference between B, N&F remineralizing groups.

C- Effect of pH cycling periods on microhardness:

Regarding the B, N, F and FI groups the mean microhardness values after two months (M2) revealed statistically significantly highest value followed by one month (M1) and the lowest one was after two weeks (W2).

For Bioactive glass (B) group, there was statistically significance difference between W2 (54.41±1.82), M1 (57.6 ± 1.17) & M2 (60.1 ±1.39) subgroups where, at M2 showed the highest mean of microhardness while W2 showed the lowest mean of microhardness value.

For Nano-hydroxyapatite (N) group, there was statistically significance difference between W2(54.5±1.34), M1(57.88±2.20) & M2 (61.58±1.91) subgroups where, at M2 showed the highest mean of microhardness while W2 showed the lowest mean of microhardness value.

For NaF (F) group, there was statistically significance difference between W2 (54.83±1.74), M1 (58.17±1.44) & M2 (61.67±1.30) subgroups, where M2 showed the highest mean of microhardness while W2 showed the lowest mean of microhardness value.

For fluoride iontophoresis (FI) group, there was statistically significance difference between W2 (57.01±3.83), M1 (60.01±3.17) & M2 (64.67±5.00) subgroups where, at M2 showed the highest mean of microhardness while W2 showed the lowest mean of microhardness value.

TABLE (2) The mean and standard deviation (SD) values of microhardness with different remineralizing agents regarding the pH cycling.

| pH cycling | Remineralizing Material | BAG Mean ±SD | n-HAP Mean ±SD | NF Mean ±SD | FI Mean ±SD | P-value |
|------------|-------------------------|--------------|----------------|-------------|-------------|---------|
| Week 2     | BAG                     | 54.41 ±1.82  | 54.5 ±1.34     | 54.83 ±1.74 | 57.01 ±3.83 | <0.05*  |
| Month 1    | BAG                     | 57.6 ±1.17   | 57.88 ±2.20    | 58.17 ±1.44 | 60.1 ±3.17  | <0.05*  |
| Month 2    | BAG                     | 60.1 ±1.39   | 61.58 ±1.91    | 61.67 ±1.30 | 64.67 ±5.00 | <0.05*  |

*p-value*; significant (p<0.05)  ns; non-significant (p>0.05)
**DISCUSSION**

Microhardness testing considered a relatively simple and reliable method giving an indirect information on mineral content changes in dental hard tissues\(^{(15,16)}\), carious lesions of the dentin have been quantitatively linked to the mineral content of the tissue, so microhardness technique has been applied successfully for the comparative evaluation of the effectiveness of various treatment regimens on incipient carious lesions\(^{(17)}\).

Medical iontophoresis widely used in dermatology because of penetration of therapeutic agents which is maximized through iontophoresis delivery system\(^{(18)}\). In the field of dentistry, the delivery of fluoride ion is still a challenge therefore, iontophoresis system is interesting in dentistry as a fluoride delivery system because of; there may be a histological difference in the site of application between the teeth and skin. Also, the optimal electrochemical properties of FI were not taken into consideration\(^{(19)}\).

Significant difference was found between FI group and the other tested OTC groups during pH-cycling periods which revealed decreased demineralization and/or increased remineralization of the artificial lesions. However, there was no statistically significant difference between those treatment OTC groups.

**TABLE (3):** The mean, standard deviation (SD) values of microhardness with different pH cycling periods regarding remineralizing agents.

| Remineralizing Material | pH cycling | Week 2 Mean ±SD | Month 1 Mean ±SD | Month 2 Mean ±SD | P-value |
|-------------------------|------------|-----------------|-----------------|-----------------|---------|
|                         |            |                 |                 |                 |         |
| BAG                     |            | 54.41 ±1.82     | 57.6 ±1.17      | 60.1 ±1.39      | <0.05*  |
| n-HAP                   |            | 54.5 ±1.34      | 57.88 ±2.20     | 61.58 ±1.91     | <0.05*  |
| NF                      |            | 54.83 ±1.74     | 58.17 ±1.44     | 61.67 ±1.30     | <0.05*  |
| FI                      |            | 57.01 ±3.83     | 60.01 ±3.17     | 64.67 ±5.00     | <0.05*  |
| p-value                 |            | <0.05*          | <0.05*          | <0.05*          |         |

\*; significant \(p<0.05\)  ns; non-significant \(p>0.05\)

**FIG (3)** Bar chart representing microhardness with different remineralizing agents regarding the pH cycling.

**FIG (4)** Bar chart representing microhardness with different pH cycling periods regarding remineralizing agents.
That superior microhardness value of FI group over that of conventional OTC may be attributed to Iontophoresis devices which are more effective at penetrating drug ions into the deeper tissues\(^{(20)}\) while in other groups, the remineralization of carious dentin can occur either by spontaneous deposition of phosphorus, calcium and fluoride ions onto crystallites remnant which remain at the partially demineralized dentin or by incorporation the same ions from treatment of the external sources by (NovaMin, n-HA or NaF)\(^{(21)}\).

Furthermore, the higher microhardness value of IF and F group can be explained by the presence of fluoride which reduce the amount of demineralization and promotes re-deposition of apatite by the formation of high mineral content bands at surface and/or sub-surface area\(^{(4)}\). Another explanation might be attributed to the fact that fluoride was applied to porous, partially demineralized dentin, and so it provides a huge reactive surface area before each period of demineralization and remineralization\(^{(22)}\).

This result agrees with Churchley D et al.,\(^{(23)}\) who reported that the application of sodium fluoride topically may result in remineralization mainly by reducing apatite dissolution through formation of less soluble flour-apatite and increasing surface microhardness.

Calcium sodium phosphosilicate particles which is bioactive glass and commercialized for dental applications, has low bioavailability (water solubility) of calcium and phosphate in vitro which determine the remineralization potential of these products\(^{(24)}\). The increase in surface microhardness in the Nano-hydroxyapatite groups on softened dentin may be related to the ability of Nano-hydroxyapatite to promote remineralization by gradual deposition of the minerals that precipitates and nucleates in the dark zone of demineralization thereby offering complete regeneration of the lost crystallites\(^{(25)}\).

The results of this study revealed that statistically significant mean microhardness values difference was found between pH cycling periods (W2, M1 &M2) which may be related to time factor, as the time increases the rate and amount of nano-hydroxyapatite deposition would increase, at the same time with the deposition of extensive amounts of F, Ca\(^{2+}\) and PO\(^{3-}\) which significantly enhance the remineralization effect\(^{(26)}\).

The mechanism of uptake of fluoride by apatite crystals includes surface adsorption and/or heterionic exchange with hydroxyl ions of hydroxyapatite\(^{(27)}\). Slow release of fluoride over a prolonged period enhanced remineralization and found that the greatest degree of remineralization depth was in the fluoride iontophoresis groups\(^{(28)}\). Furthermore, in case of application of high fluoride concentrations or increase time (pH cycle periods)\(^{(29)}\).

In this study, the FI device mode was set according to the manufacture manual instructions. However, there is insufficient academic evidence regarding the standard conditions for FI. As well as, the fluoride concentration of FI was not concentrated to achieve the maximum treatment effect\(^{(30)}\). In order to maximize the preventive potential, the histological feature of the teeth and the electrical characteristics of the FI device need to be considered. Therefore, further study is needed to determine the electrochemical properties of FI devices, including the applied electric current, voltage and concentration of drug.

**CONCLUSIONS**

According to the circumstances of this investigation, the following conclusions could be reported:

1. Microhardness of dentin is positively affected by the application of remineralizing agents.

Increasing the application rate (pH cycling periods) of the remineralizing agents significantly affect the surface microhardness of dentin.
2. The compositions of remineralizing agents produce different effect on dentin surface.

3. Fluoride iontophoresis was superior to conventional OTC in the microhardness and hence the remineralization of dentin.

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