RESEARCH PAPER

Comparative Evaluation of Different Types of Remineralizing Toothpastes on White Spot Lesions: An In Vitro Study

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Abstract

Background: White spot lesions (WSLs) are a frequent complication of fixed orthodontic treatment due to plaque accumulation and enamel demineralization.

Aim: To compare the remineralization efficacy of different commercially available toothpastes on WSLs using surface microhardness analysis.

Materials and Methods: Sixty extracted human premolars were demineralized and divided into four groups: artificial saliva (control), Sensodyne ProEnamel, Vantej (nano-hydroxyapatite), and Amflor (amine fluoride). Surface microhardness (VHN) was measured at baseline, post-demineralization, and post-remineralization. Statistical analysis included ANOVA and Tukey's post hoc test.

Results: Significant reduction in microhardness occurred after demineralization ($p < 0.001$). Maximum remineralization was observed with Amflor (94.7%), followed by Vantej (57.6%) and Sensodyne (29.6%).

Conclusion: Amine fluoride toothpaste showed superior remineralization potential and can be recommended for prevention of WSLs in orthodontic patients.

Keywords: (WSLs)White spot lesion, (amine fluoride)Amflor, , (nano-hydroxyapatite) Vantej.

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Introduction

White spot lesions (WSLs) are early manifestations of enamel demineralization and are commonly associated with fixed orthodontic appliances. The presence of brackets and bands promotes plaque retention, creating an acidic microenvironment that leads to mineral loss from enamel.1-4

The incidence of WSLs in orthodontic patients ranges from 23% to 97%, significantly affecting esthetics and treatment outcomes. 1,2,15

Although saliva contributes to remineralisation, its effect is often insufficient under orthodontic conditions. Therefore, the use of remineralizing agents such as fluoride, nano-hydroxyapatite, and amine fluoride-based toothpastes has gained importance.4-7

This study aims to evaluate and compare the effectiveness of these agents in reversing enamel demineralisation.

Materials and Methods

Study Design

This in vitro study included 60 extracted human premolars.(FIGURE 1)

- Group A: Baseline (pre-demineralisation)
- Group B: Post-demineralization

Group B subdivided into:

- B1: Artificial saliva (control)
- B2: Sensodyne ProEnamel
- B3: Vantej toothpaste
- B4:Amflor toothpaste

FIGURE 1 : Extracted Premolars Used in Study

Specimen Preparation
Teeth were mounted in acrylic blocks with a standardized 4×4 mm window exposed on the buccal surface.(FIGURE 2)

FIGURE NO 2: Acrylic blocks

Surface Microhardness Testing (FIGURE 3)
Vickers microhardness testing was performed at:

- Baseline
- Post-demineralization
- Post-remineralization

Figure 3: Vickers Microhardness Testing Machine

Demineralisation Protocol (FIGURE 4)
Samples were immersed in demineralising solution (pH 4.5) for 10 days to induce WSLs.

Figure 4: Demineralisation Process

Remineralization Protocol

Each group underwent treatment twice daily for 14 days.(Figure 5)

- B1: Artificial saliva
- B2: Sensodyne
- B3: Vantej
- B4: Amflor

Outcome Measure

Primary outcome:

- Surface Microhardness (VHN)
- % Recovery calculated

Statistical Analysis

- Repeated measures ANOVA
- One-way ANOVA
- Tukey post hoc test
- Significance level: $p < 0.05$

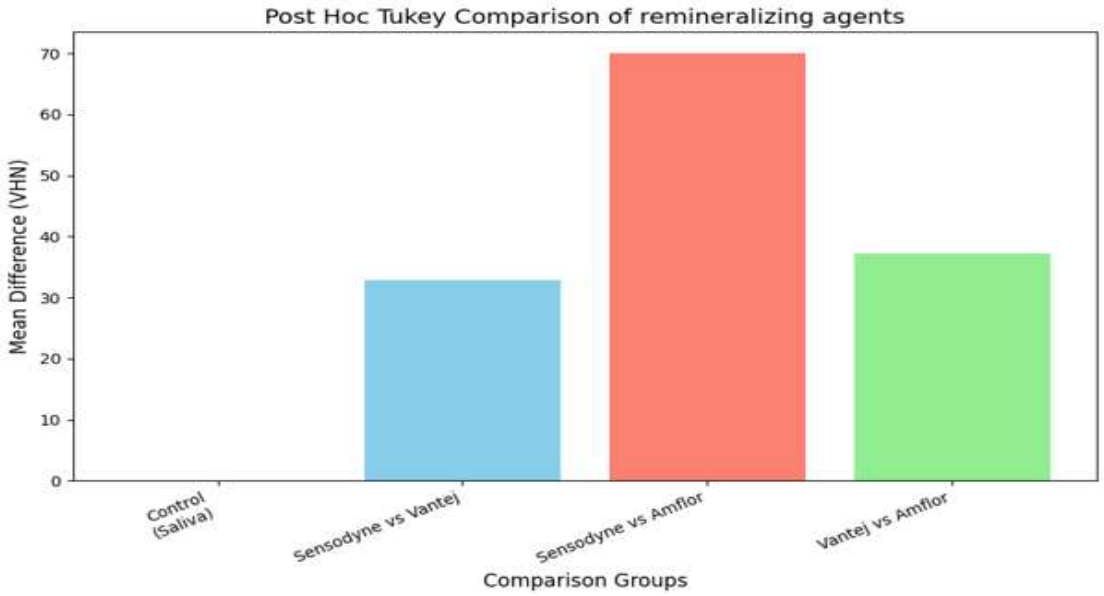
Figure 5- Artificial saliva

Results

A significant reduction in surface microhardness was observed after demineralization ($p < 0.001$). (Table 1)
Post-remineralization:

Table 1: Surface Microhardness Comparison

Group	VHN	% Recovery
Artificial saliva	215	2.8%
Sensodyne	241	29.6%
Vantej	274	57.6%
Amflor	311	94.7%



Discussion

White spot lesions (WSLs) remain one of the most common and undesirable complications associated with fixed orthodontic therapy.1-4 The present in vitro study was designed to evaluate and compare the remineralization potential of different commercially available dentifrices using surface microhardness (SMH) as an objective indicator of enamel mineral content.

The results of this study demonstrated a highly significant reduction in surface microhardness following demineralization, confirming the successful creation of artificial WSLs. The mean SMH decreased from 317.53 ± 20.45 VHN to 208.93 ± 11.62 VHN ($p < 0.001$), which is consistent with previous studies that have reported substantial mineral loss under acidic conditions.5,6,11 This reduction can be attributed to dissolution of hydroxyapatite crystals and increased enamel porosity, resulting in compromised structural integrity.1,4,5,6,11

Importantly, no statistically significant difference was observed among groups at the post-demineralization stage ($p = 0.88$), indicating uniform lesion formation across all specimens. This finding validates the study design and ensures that subsequent differences in remineralization are attributable solely to the treatment modalities rather than baseline variability.6,7,10

Following remineralization, all experimental groups demonstrated an increase in SMH, indicating varying degrees of mineral gain. However, the magnitude of recovery differed significantly among the agents, highlighting the influence of their active components and mechanisms of action.

Among the tested agents, Amflor (amine fluoride) exhibited the highest remineralization potential, with a mean SMH of 311.30 ± 10.80 VHN and 94.7% recovery, approaching baseline values. The superior performance of amine fluoride can be attributed to its surface-active properties, which facilitate enhanced adsorption onto enamel surfaces and formation of a stable calcium fluoride-like reservoir. This reservoir acts as a sustained source of fluoride ions, promoting the formation of fluorapatite, which is more resistant to acid dissolution. Additionally, amine fluoride has been reported to

improve plaque control due to its antimicrobial properties6,7,10, further contributing to its clinical effectiveness.

The Vantej toothpaste (nano-hydroxyapatite) demonstrated moderate remineralization efficacy, with a mean SMH of 274.10 ± 9.30 VHN and 57.6% recovery. Nano-hydroxyapatite particles closely resemble the natural mineral composition of enamel and can penetrate microporosities in demineralized enamel. These particles act as a scaffold for crystal growth, facilitating direct deposition of minerals and restoration of enamel structure. However, the extent of remineralization observed in this study was lower than that of amine fluoride, possibly due to differences in ion release dynamics and depth of penetration.6,7

The Sensodyne ProEnamel group (sodium fluoride-based) showed comparatively lower remineralization, with a mean SMH of 241.30 ± 8.30 VHN and 29.6% recovery. Although sodium fluoride is well established in promoting remineralization, its mechanism primarily relies on ionic exchange and formation of a superficial fluorapatite layer. The lower efficacy observed in this study may be attributed to its relatively limited substantivity and reduced retention on enamel surfaces compared to amine fluoride formulations.6,7,11

The artificial saliva control group demonstrated minimal remineralization (2.8% recovery), confirming that natural remineralization alone is insufficient under experimental conditions. While saliva provides calcium and phosphate ions necessary for remineralization, its effect is often inadequate in the presence of persistent cariogenic challenges, particularly in orthodontic patients with compromised oral hygiene.2,4

Intergroup comparisons using one-way ANOVA revealed a highly significant difference among the remineralizing agents ($p < 0.001$), confirming that the type of dentifrice plays a critical role in enamel recovery. Post hoc Tukey analysis further demonstrated that all pairwise comparisons between the treatment groups were statistically significant, establishing a clear hierarchy of efficacy:4,5

Amflor > Vantej > Sensodyne

The findings of this study are consistent with previous literature emphasizing the superior performance of fluoride-based formulations, particularly amine fluoride, due to their enhanced bioavailability and prolonged action. Similarly, studies on nano-hydroxyapatite have reported its potential as a biomimetic agent, although its effectiveness may vary depending on particle size, concentration, and formulation.8-11

The mixed-effects linear regression model used in this study further strengthened the results by accounting for repeated measurements and inter-sample variability. The significant interaction between time and treatment ($p < 0.001$) indicates that the observed improvements in SMH are primarily driven by the remineralization agents during the treatment phase.

Limitations of the Study

The present study, although methodologically sound, has certain limitations that must be acknowledged. Being an in vitro investigation, the experimental conditions do not fully replicate the complex biological environment of the oral cavity, where multiple dynamic factors such as salivary flow, pellicle formation, oral microbiota, dietary variations, and pH fluctuations continuously influence the demineralization– remineralization process. The use of artificial saliva, while standardized, lacks the complete biochemical composition and protective functions of natural saliva, potentially affecting the extent of remineralization observed.

Additionally, the study evaluated remineralization using surface microhardness (SMH) as the sole outcome measure. Although SMH is a reliable and widely accepted indicator of mineral changes, it primarily reflects surface-level alterations and may not accurately represent subsurface remineralization or lesion depth. Advanced imaging techniques such as transverse microradiography or micro-CT could have provided a more comprehensive assessment.

Another limitation is the relatively short duration of the remineralization protocol, which may not adequately simulate long-term clinical use of dentifrices. The sample size, though statistically sufficient, was limited to extracted teeth under controlled conditions, which may restrict the generalizability of the findings to a broader population. Furthermore, variations in dentifrice application methods, brushing force, and patient compliance—which are critical in clinical scenarios—were not replicated in this study.

Conclusion

Within the limitations of this in vitro study, it can be concluded that all tested remineralizing agents demonstrated the ability to enhance enamel surface microhardness following demineralization. However, the extent of remineralization varied significantly depending on the composition of the dentifrice.

Amine fluoride-based toothpaste (Amflor) exhibited the highest remineralization potential, achieving near-complete recovery of enamel microhardness. Nano-hydroxyapatite toothpaste (Vantej) showed moderate efficacy, while conventional fluoride toothpaste (Sensodyne ProEnamel) demonstrated comparatively lower remineralization. Artificial saliva alone produced minimal improvement, indicating that natural remineralization is insufficient under cariogenic conditions.

The findings highlight that the choice of remineralizing agent plays a crucial role in the prevention and management of white spot lesions, particularly in orthodontic patients. Amine fluoride-based formulations can be considered the most effective among the tested agents for enhancing enamel remineralization.

Further in vivo studies are recommended to validate these results under clinical conditions and to assess the long-term stability of remineralized enamel.

Clinical Significance

White spot lesions are among the most common adverse effects of fixed orthodontic treatment and can compromise both esthetics and long-term enamel health. The findings of this in vitro study suggest that the choice of remineralizing toothpaste significantly influences the recovery of demineralized enamel. Among the agents evaluated, the amine fluoride-based toothpaste demonstrated the greatest remineralization potential, followed by nano-hydroxyapatite toothpaste. These results indicate that appropriate selection of remineralizing dentifrices may help prevent and manage white spot lesions in orthodontic patients. Although further in vivo studies are required to confirm these findings, the present study provides useful evidence to guide clinicians in recommending effective preventive oral care products during fixed orthodontic treatment.

Disclosure Statement

Conflict of Interest: The authors declare that they have no conflicts of interest related to this study.

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Ethical Approval: As this was an in vitro study conducted on extracted human premolar teeth, formal institutional ethical committee approval was obtained where required, and all extracted teeth were collected following institutional guidelines with appropriate consent.

Informed Consent: Not applicable.

Data Availability: The data supporting the findings of this study are available from the corresponding author upon reasonable request.

Author Contributions: All authors contributed substantially to the conception and design of the study, data acquisition, analysis and interpretation, manuscript preparation, critical revision, and approval of the final version.

AI Disclosure: Artificial intelligence (AI) tools were used only for language refinement and grammar improvement during manuscript preparation. The authors take full responsibility for the accuracy, originality, and scientific content of the manuscript.

Data Availability Statement

The data generated and analyzed during this in vitro study are available from the corresponding author upon reasonable request. All relevant data supporting the findings of this study are included within the manuscript. Additional raw data, including surface microhardness measurements and statistical analysis files, can be made available by the corresponding author upon reasonable request.

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