Summary

Aim: to investigate the remineralization of the artificial white spot lesions under different conditions: in vitro and in situ.

Materials and Methods:
To induce the dental lesions, the bucal surfaces of 20 sound premolar teeth were treated for 1 minute with H3PO4 37%. For the first (in vitro) study, the teeth were divided into two groups: the first one was immersed in artificial saliva, and the second - in artificial saliva with F⁻, at a neutral pH in the first experimental week, and at a low pH (5.5) in the second experimental week. After each week one tooth was removed from each solution for a clinical examination and a chemical analysis of the solutions was made. For the second (in situ) study the teeth were sectioned in two halves, one being fixed in an orthodontic appliance, and the other half being the control one. After two weeks, a clinical examination of the teeth was made.

Results: the remineralization of the white spot lesions in saturated solutions in calcium-phosphate - as the artificial and natural saliva - is a common phenomenon and is stimulated by the presence of fluoride ions, especially at low pH.

Conclusion: the remineralization in situ is significantly more effective than the remineralization in vitro.

Introduction

According to the literature, the „white spot” lesions undergo several changes in the clinic or in the laboratory [1, 3, 13, 14]. It may thus be transformed into a „brown spot” lesion, a remineralized lesion, an arrested lesion, and occasionally it even dissappears completely as a „caries reversal” [2, 4, 6, 7, 10]. Caries progression versus reversal is a delicate balance between bacterially generated acid challenges [5, 8, 9, 11] and a combination of demineralization inhibition and reversal by remineralization [3, 12, 13, 14]. The balance between pathological factors and protective factors - saliva (calcium, phosphate) and fluoride - is delicate [6, 18, 13] being tipped either way several times daily in most people [2, 3, 5].

The aim of the studies is to compare and to evaluate the clinical changes in the macroscopic features of the artificial white spot lesions immersed in artificial and natural saliva with and without increased fluoride content.

Materials and methods

Lesion formation: for both in vitro and in situ experiments, the buccal surfaces of sound premolar teeth extracted for orthodontic reasons were treated for 1 minute with phosphoric acid H₃PO₄ 37% [1, 4, 10] (Figure 1).
After etching, the surfaces were washed and dried. The white spot lesions with an increased porosity have became clinically visible (Figure 2).

In the second experimental week, the pH in both solutions was dropped at 5.5. At the beginning of the experiment the concentration of calcium ions in artificial saliva and of calcium and fluoride ions in the saliva with fluoride was chemically determined. At the end of the first and of the second experimental weeks, the amount of calcium and fluoride ions consumed from the solutions was calculated. Fluoride concentrations were determined with a fluoride-sensitive electrode (Cole-Palmer® Fluoride Electrodes - Cole-Palmer Instrument Company, Illinois, USA) according to the manufacturer's instructions. At the same time, after each week some teeth were removed from each solution for clinical examination.

**Chemical results of the in vitro experiment:**

1. After first week at a neutral pH in the artificial saliva without fluoride ions, all the calcium ions were consumed probably by the formation of hydroxyapatite.
2. The saliva supersaturated with calcium and phosphate provide a driving force for mineral to go back into the teeth.
3. The partially dissolved crystals were probably the „nucleators” for remineralization.
4. In the second solution - with fluoride ions - after the first week at neutral pH there are still calcium ions available, but the concentration of fluoride ions dropped with more than 4 ppm.
5. The existence of calcium ions in the second solution can lead for the idea that in the presence of fluoride ions, calcium ions reacts first with these ions, not with phosphate ions as in the first solution, forming calcium fluoride available for deposition into the lesions.

All the chemical findings are supported by the **clinical results:**

1. After the first week the macroscopic features of the lesions immersed in the solutions are significantly changed (Figure 5): the porosity is decreased - much more for the saliva with fluoride ions - and the colour is modified in yellow - much more for the saliva without fluoride ions.
2. After the second experimental week at a low pH, on the surfaces immersed in artificial saliva - now unsaturated in calcium and phosphate ions - new white spot lesions have appeared (Figure 6).

3. The surfaces immersed in the saliva with fluoride at low pH have a glossy appearance, without any porosity even after prolonged drying with air.

**Conclusion of the in vitro experiment:**

1. All the results confirm the ability of white spot lesions to be turned into arrested or remineralized lesions after immersion in supersaturated solutions in calcium and phosphate.
2. The results are significantly better when fluoride is present and the solution has a low pH.

**For the in situ study,** the teeth were sectioned in two halves (Figure 7).

The experimental procedures used in this study were done with the informed consent of all 20 subjects. The subjects carried a half of their extracted tooth in their mouths, fixed on their orthodontic appliance, the other half being the control one. Two types of appliances were used:

- removable appliances (Figure 8), carried 24 hour/day, including meals;
- functional appliances (Figure 9), earned only 12-14 hours/day (in the night and 2-3 hours in the day time).
In the two weeks of the experiment, the subjects consumed three meals and two snacks a day in a random order, and they used a fluoridated (1000 ppm F) toothpaste twice daily for their teeth and also for cleaning their appliances.

Clinical results of the in situ experiment:

I\textsuperscript{st} case: the lesion fixed in the removable appliance (Figure 10);

![Figure 10](image)

After two weeks the lesion is significantly changed, with a glossy, white, and without porosity appearance (Figure 11).

![Figure 11](image)

II\textsuperscript{nd} case: the lesion fixed in the functional appliance

![Figure 12](image)

After two weeks, the lesion is glossy, with increased hardness (Figure 13).

![Figure 13](image)

**Conclusions**

1. The new surfaces of lesions - glossy appearance, surface increased hardness, missing porosity - are the features of arrested, or even remineralized lesions.

2. The factors responsible for the clinical changes of the white spot lesions in the oral cavity include the supersaturation of the saliva and plaque fluid in calcium and phosphate and of the fluoride from toothpaste.

3. The results are significantly more effective in situ than in vitro conditions, probably because of the cyclic pH conditions in the oral cavity, and because of the role of fluoride on the plaque fluid and composition.

4. Since arrested white spot lesions are a common phenomenon, the general prophylactic measures must seek, as one of the most important goals, to stimulate the factors that will prevent white spot lesions progression and will stimulate their remineralization.
References


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