

Preliminary report concerning the influence of simulated tissue liquid upon the Calcium Phosphate system – an In Vitro study

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Abstract: *The aim of the authors is in conditions of in vitro experiment to make a preliminary study of the effect of simulated body fluid on the calcium phosphate system – K_2HPO_4 and $Ca(NO_3)_2$. The K_2HPO_4 and $Ca(NO_3)_2$ system was subjected to in vitro study where calcium phosphates are received on the dentinal tubules and it was monitored the effect of simulated body fluid (SBF) by Kokubo upon the processes of transformation of dried sample of brushite ($CaHPO_4 \cdot 2H_2O$). The pilot study showed necessity of additional in vitro and in vivo experiments to refine the composition and concentration of applied substances to achieve the effect on dentin mostly congruent with the natural regenerative activity of tooth structure.*

Keywords: Simulated tissue liquid, Calcium Phosphate system, In vitro study.

1. Introduction

The treatment of prepared dentin with various medications in older sources [2, 4, 5] is summarized as "disinfection" and "sterilization". A number of authors [8, 9, 10, 11] are focusing on searching of a substance to provide a reliable and lasting isolation, easy to apply, with no toxic effect and possessing bacteriostatic or bactericidal properties. Recommended are more than 30 different substances, but none has been designated to be both effective and safe.

Contemporary research [1, 3, 6, 7, 12, 13] is oriented towards materials that enable increasing of regenerative capacity of tooth structures.

Based on the hydrodynamic theory which states that any reduction in the functional radius of dentinal tubules reduces the degree movement of tubular liquid and affects the sensitivity, Greenhill, J., D. Pashley [7] examined the ability of 28 desensitizing agents on the extent of movement of the tubular liquid in the dentine in vitro. The experimental results indicate that the major change rate of the hydraulic conductivity before and after treatment ranges from 0% for the calcium nitrate and calcium chloride to 98.4% for calcium oxalate.

In the study of natural regenerative activities of tooth structures it is interesting to investigate the application of calcium phosphates, as a regenerative substances.

The study of natural regenerative activities of tooth structures incorporates a thorough analysis of the application of calcium phosphates as a regenerative substances.

2. Aim

The authors set themselves the goal in conditions of an in vitro experiment to investigate the effect of simulated body fluid on calcium phosphate system – K_2HPO_4 and $Ca(NO_3)_2$.

3. Material and Methods

It was subjected on laboratory tests in vitro the system K_2HPO_4 and $Ca(NO_3)_2$, that produces calcium phosphates on the dentinal tubules:

- Primary calcium phosphate precipitation was obtained by rapid mixing of alcoholic solutions of $Ca(NO_3)_2 \cdot 4H_2O$ (1M) and K_2HPO_4 (0.6 M). The resulting calcium phosphate sediment was filtered off and dried at room temperature.
- The phase composition of the obtained samples was controlled by a powder X-ray diffraction on a device DRON-UM1 with filtered CoK_{α} radiation – the Ca/P ratio was determined analytically.
- It was traced the influence of simulated body fluid (SBF) by Kokubo on transformation processes of the dried sample of brushite ($CaHPO_4 \cdot 2H_2O$) for 6, 24, 72 hours and 7 days at 37°C.

4. Results and Discussion

There were carried out physico-chemical studies of the obtained by the interaction between $Ca(NO_3)_2$ and K_2HPO_4 calcium phosphate precipitates. Through the X-ray diffraction analysis it was demonstrated that the precipitated product is a brushite $CaHPO_4 \cdot 2H_2O$ (with Ca/P ratio of 1.22). The subsequent biomimetic phase transformation of calcium phosphate into Simulated Body Fluid (SBF) was made in order to model the processes of phase transformation that would occur in vivo in the dentine. Presented on Figure 1 diffractograms showed that for the detected period of up to 72 h at the SBF kinetic transformation the primary phase undergoes a transition from DCPD to apatite structure and nanometric calcium phosphates with apatite structure are obtained - triclinic octacalcium phosphate $Ca_8(PO_4)_4(HPO_4)_2 \cdot 5H_2O$ (Ca/P=1,33).

Diffractograms obtained after 7 days, confirming the fine grainy structure.

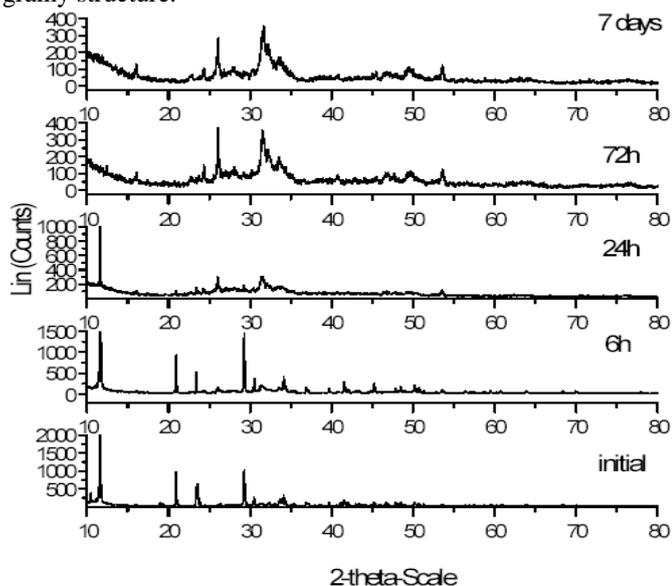


Fig. 1: Diffractograms of dried sample of brushite, which stayed for 6, 24, 72 hours and 7 days in SBF.

5. Conclusion:

The survey showed that both the composition and concentration of applied substances should be further refined in order to reach a clearer concept for their practical application.

Biomimetic transformation of calcium phosphate in SBF and the results obtained are hopeful that in vitro and in vivo experiments in this direction will allow to influence upon the dentine mostly congruent with the natural regenerative activities of tooth structures. The approach is reliable and the more profound comprehensive research in this direction would lead to the refinement of methods and tools for biomimetic mineralization.

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