Teeth Whitening Followed by Remineralization Therapy: 
a Comparative Analysis of Bleaching Systems 
and Remineralizing Agents

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The aim of the study was to determine the most efficient teeth whitening technique based on the study of biochemical composition of oral fluid.

Materials and Methods. The study involved 120 patients divided into three groups, 40 patients in each group, according to a bleaching system used. 30 patients in each group underwent professional teeth whitening with subsequent remineralization therapy, the rest 10 patients having a whitening procedure alone. A biochemical assay of oral fluid was carried out prior to bleaching, immediately after double whitening and remineralization therapy, and 14 days after bleaching. Biochemical assay was applied to determine the acid degree in oral fluid as well as ionized calcium level in oral fluid.

Results. The study revealed a significant increase in calcium ion concentration in oral fluid after teeth bleaching. Maximum calcium ion loss in dental enamel was found when exposed to photobleaching based on 24% hydrogen peroxide concentration; the lowest loss was revealed when using chemical activation and 40% hydrogen peroxide. Remineralization therapy appeared to be effective in all three groups. A maximum effect was found when using the agent based on zinc-substituted carbonate hydroxylapatite Stomsens combined with laser phonophoresis.

Key words: teeth whitening; bleaching systems; biochemical assay of oral fluid; teeth remineralization.

Introduction

Professional tooth bleaching has become widely used in dental practice worldwide that has given rise to its efficiency and safety study. Despite a great number of publications on teeth whitening techniques and possible changes in hard tissues [1–5], many issues in clinical practice still remain open. Currently, the use of certain bleaching systems and safety measures of bleached teeth frequently has no appropriate theoretical grounds.

Whitening quality is known to depend on both: the properties of bleaching systems used, protective techniques of bleached teeth, and enamel composition as well. Enamel acts as a porous membrane: small ions go deeper easily than large molecules, which are absorbed on the surface and can be desorbed without any change in dental enamel crystal form [6, 7].

Surface enamel differs from deep layers by higher mineralization, density, micro-hardness, caries-resistance, higher micronutrient content including fluoride. Surface enamel is less exposed to an acid attack than enamel deep inside parts [7]. In enamel decalcination induced by an organic acid attack, the change of the form, size, and orientation of hydroxylapatite crystals are in progress.

Up to a third of ions in hydroxylapatite can be exchanged. So, calcium ions can be substituted by sodium, silicon, strontium, lead, cadmium, hydroxonium ions and others. Hydroxyl ions can be exchanged for fluoride, chloride ions, and others. The alteration of an optimal carbonate level modifies solubility and strength in mineralized tissues, as well as oral fluid pH due to the substitution of orthophosphate residues in a hydroxylapatite molecule forming carbonated hydroxylapatite.

Calcium citrate in a forward direction has an effect on calcium transport between mineralized tissues and biological fluids. It acts as a calcium transporter from blood to mineralized tissues when hydroxylapatite structures are modeled, and in the opposite direction it acts as chelate in mineralized tissue resorption.

Most researchers consider that the main condition for ions and anions entering the enamel is the osmotic pressure difference of intercellular fluid in the pulp and oral fluid on tooth surface. Since oral fluid is richer in phosphates, calcium ions, and other ions, compared
to interstitial fluids (enamel fluid), ions migrate from the fluid to tooth enamel. The process is complex and can be altered under the influence of many factors: the concentration of substances, enzyme activity, pH level, molecule size, etc. [1]. Therefore, when studying the enamel absorption of inorganic and organic substances, the unavoidable question is the role of oral fluid — the medium a tooth is located, since the substance can enter only as an ionized form, i.e. after being dissolved in a liquid medium.

Thus, the study of biochemical composition of oral fluid, its alteration when exposed to various bleaching systems, will enable to reveal the most efficient whitening technique.

**The aim of the study** was to determine the most efficient teeth whitening technique based on the study of biochemical composition of oral fluid.

**Materials and Methods**

The study involved 120 patients divided into three groups, 40 patients in each group, according to a bleaching system used (for bleaching purposes we have chosen three most popular systems in dental market nowadays, such as Opalescence Xtra Boost (Ultradent Products, USA), Amazing White Professional (Amazing White, USA), and the bleaching system Beyond Polus (Beyond Technology Corp., USA));

- **group 1** — teeth whitening included chemical activation using Opalescence Xtra Boost with 40% hydrogen peroxide concentration;
- **group 2** — teeth whitening being carried out using a photochemical activation system, Amazing White Professional with 37% hydrogen peroxide concentration;
- **group 3** — photobleaching was performed using Beyond Polus with 24% hydrogen peroxide concentration.

30 patients in each group underwent professional teeth whitening with subsequent remineralization therapy, the rest 10 patients having a whitening procedure alone.

Each group, in turn, was subdivided into three subgroups, 10 patients in each subgroup, according to a remineralizing agent applied, which was used after teeth whitening in order to prevent hyperesthesia:

- the first subgroup — teeth were remineralized after bleaching by enamel-sealing liquid (Humanchemie, Germany);
- the second subgroup — remineralization of teeth performed using the agent based on zinc-substituted carbonate hydroxylapatite Stomysens (BioRepair, Italy);
- the third subgroup — remineralization was performed using the agent based on zinc-substituted carbonate hydroxylapatite Stomysens combined with laser phonophoresis.

The study was approved by the Ethics Committee of Privolzhsky Research Medical University and complies with Declaration of Helsinki (2013). All patients gave their written informed consent to research their data. All the individuals under study underwent professional oral hygiene and oral cavity sanation (provisional restoration) before whitening. A biochemical assay of oral fluid was carried out before bleaching, immediately after bleaching and remineralization therapy, and 14 days after whitening. Oral fluid was collected by spitting in sterile vacuum collection tubes.

The biochemical assay included the determination of an acid level in oral fluid, as well as ionized calcium level in oral fluid. The acids were evaluated using gas-liquid chromatography [5]. Calcium ions in oral fluid were quantitatively measured using an ion-selective electrode [4].

The findings were statistically processed using Excel 2010. The reliability of differences was studied by Student t-test and Fisher’s ratio test in a confidence interval over 95%. The differences were considered significant in p<0.05.

**Results**

Immediately after teeth bleaching the patients of all three groups were revealed to have a significant concentration increase in calcium ions in oral fluid in relation to the norm and to the values they showed before whitening (p<0.05), maximum values being found in three group patients: 119.80±0.97 mg/L; p=0.027 (Table 1). The fact suggests that calcium ions are coming out of the enamel crystalline grid while whitening.

By day 14 after the whitening procedure, a reliable decrease was found in calcium ions contained in oral fluid in relation to the values before whitening, however, the values of the latter significantly exceeded those before whitening (p<0.05), it can be due to enamel remineralization processes. The maximum values were also revealed in group three: 96.90±1.88 mg/L; p=0.03 (see Table 1).

The study findings suggest that the bleaching system Beyond Polus has the most pronounced effect on teeth enamel compared to other systems. Amazing White Professional was found in between Beyond Polus and Opalescence Xtra Boost by the effect on a calcium ion level.

Thus, the maximum loss of calcium ions on tooth enamel is under the influence of the bleaching system based on 24% hydrogen peroxide concentration, and the minimum — when using the systems with chemical activation and 40% hydrogen peroxide concentration. The findings provide strong evidence on calcium ions coming out of the enamel crystalline grid and prove the necessity to use remineralization therapy after bleaching.

The analysis of oral fluid indices after whitening followed by remineralization therapy has shown that the use of remineralizing agents results in quantity reduction of calcium ions in all three groups (p<0.05). Insignificant decrease of calcium ions in relation to the data found before remineralization therapy was observed in the
**Table 1**

Calcium ion concentration (mg/L) in oral fluid before and at different periods after whitening (M±m)

| Bleaching system                                      | Follow up periods                  |
|-------------------------------------------------------|-----------------------------------|
|                                                       | Before whitening | Immediately after bleaching | 14 days after bleaching |
| Group 1 — Opalescence Xtra Boost with 40% hydrogen peroxide concentration | 62.80±1.54; p<0.05 | 103.13±1.62; p<0.05 | 73.00±1.87; p<0.05 |
| Group 2 — Amazing White Professional with 37% hydrogen peroxide concentration | 79.03±1.61; p<0.05 | 110.93±1.68; p<0.05 | 89.23±1.99; p<0.05 |
| Group 3 — Beyond Polus with 24% hydrogen peroxide concentration | 65.03±1.50; p<0.05 | 119.80±0.97; p=0.027 | 96.90±1.88; p=0.03 |
| Norm                                                   | 61.01±3.12                                |

**Note.** p — statistical significance of values compared to the norm.

**Table 2**

Calcium ion concentration (mg/L) in oral fluid after bleaching and remineralization therapy (M±m)

| Remineralizing agents | Follow up periods                  |
|-----------------------|-----------------------------------|
|                        | Immediately after remotherapy     | 14 days after bleaching and remotherapy |
|                        | Group 1 — Opalescence Xtra Boost  | Group 2 — Amazing White Professional | Group 3 — Beyond Polus |
|                        | Before whitening                 | After whitening                    | Before whitening       | Before whitening             |
| First subgroup — enamel sealing liquid                 | 104.01±4.07                      | 111.00±4.06                        | 119.40±1.72            | 94.00±3.79                   | 98.80±2.22                      | 107.02±3.11                      |
| Second subgroup — Stomysens                            | 79.90±3.80                       | 109.80±3.57                        | 119.01±1.51            | 75.90±3.80                   | 94.00±3.79                       | 105.34±4.06                      |
| Third subgroup — Stomysens + laser phonophoresis      | 98.80±2.22                       | 113.50±1.54                        | 117.70±1.17            | 72.80±2.22                   | 91.04±2.23                       | 99.31±1.05                       |

**Table 3**

The change of organic acid content (mg/g) in oral fluid at different follow up periods after teeth whitening (M±m)

| Organic acids | Group 1 — Opalescence Xtra Boost | Group 2 — Amazing White Professional | Group 3 — Beyond Polus |
|---------------|----------------------------------|--------------------------------------|------------------------|
|               | Before whitening | After whitening | Before whitening | After whitening | Before whitening | After whitening |
| Butyric acid  | 0.02±0.01         | 0.06±0.04         | 0.028±0.012       | 0.015±0.01       | 0.024±0.01       | 0.017±0.03       |
| Propionic acid| 0.105±0.02        | 0.012±0.001       | 0.241±0.031       | 0.165±0.021      | 0.113±0.022      | 0.08±0.01        |
| Acetic acid   | 0.38±0.012        | 0.062±0.01        | 0.643±0.005       | 0.546±0.008      | 0.473±0.023      | 0.176±0.042      |

The most pronounced decrease in calcium ions was found in patients using Stomysens (79.90±3.80 mg/L; p=0.027), as well as in the third subgroup of group 1 (98.80±2.22 mg/L; p=0.023). 14 days after remineralization, the concentration of calcium ions reduced in all subgroups, however, the most significant decrease was found in the patients of the third subgroup of group 1, who were given the agent based on zinc-substituted carbonate hydroxyapatite as a remineralizing agent in combination with laser phonophoresis (72.80±2.22 mg/L; p=0.023) that was related to the incorporation of calcium ions into the enamel crystalline grid (see Table 2).

Teeth bleaching led to the decrease in the level of all acids in all three groups (p<0.05). In addition, in the first group patients, who used Opalescence Xtra

Thus, remineralization therapy appeared to be effective in all three groups, a maximum effect being found when using Stomysens in combination with laser phonophoresis.

The study of organic acid content in oral fluid in all the patients before whitening procedure showed a significant decrease in an acetic acid level related to the norm (p<0.05). The levels of propionic and butyric acids in oral fluid in all patients before bleaching also reduced although insignificantly relating to the norm (p>0.05). The phenomenon can be due to professional oral hygiene before bleaching (Table 3).
Boost with chemical activation based on 40% hydrogen peroxide concentration, the decrease was more significant compared to both: the norm (p<0.05), and also to their level before whitening (p<0.05). Therefore, a whitening procedure when using different systems is certain to lead to a risk reduction in periodontal and oral mucosa inflammatory diseases, as well as to caries intensity decrease resulted from reduced activity of microorganisms producing acids.

The level of organic acids after remineralization therapy changes slightly.

Thus, the most pronounced decrease of organic acid level in oral fluid after whitening occurs when using the bleaching system Opalescence Xtra Boost with chemical activation based on 40% hydrogen peroxide concentration.

**Conclusion**

The most significant alterations occur when using a photobleaching system Beyond Polus, and the minimal changes — when using a chemical activation system Opalescence Xtra Boost, the alterations consisting in the maximum decrease of an organic acid level, and the maximum increase in calcium ion concentration in oral fluid. The combined use of an agent based on zinc-substituted carbonate hydroxylapatite with laser phonophoresis appeared to be the most efficient for remineralization therapy. The therapy can be employed after any whitening technique.

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