Influence of Bioactive Materials on Whitened Human Enamel Surface

*in vitro study*

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**Objective:**

To investigate the influence of bioactive materials on whitened human enamel surface using Knoop hardness test

**Materials:**

- Five human teeth
- 16% carbamide peroxide, potassium nitrate, fluoride (Opalescence PF, Ultradent)
- 16% carbamide peroxide 16%, potassium nitrate, fluoride, calcium, phosphate (NiteWhite ACP, Discus Dental)
- 15% carbamide peroxide 15%, potassium nitrate, fluoride + potassium nitrate, fluoride, calcium, phosphate (Opalescence PF, Ultradent + Relief ACP, Discus Dental)
- 15% carbamide peroxide, potassium nitrate, fluoride + potassium nitrate, fluoride, calcium, phosphate (Opalescence PF, Ultradent & Relief ACP, Discus Dental)

**Methodology:**

Five human teeth were sectioned into four slices per tooth. Whitening treatments were performed for 14 days according to manufacturers' instructions. Six Knoop hardness measures were taken for each specimen, three before and three after treatments. The data were compared by Student's t-test (α=0.01).

**Results:**

OPF and OPF + Relief ACP presented statistically significant hardness decrease; NiteWhite ACP and OPF & Relief ACP mixed at the time of application showed that enamel hardness was maintained.
Conclusion:

Whitening treatment can lead to alterations in the dental structure. Minimizing or eliminating the alterations in the whitened dental structure could bring benefits to patients. With the purpose of increasing the mineral deposition on the tooth, amorphous calcium phosphate (ACP) biomaterial has been added to toothpastes, mouth rinses, chewing gums, and more recently to whitening products. This study indicates that the use of ACP simultaneously with the whitening treatment is beneficial.
Influence of Five Home Whitening Gels and a Remineralizing Gel on the Enamel and Dentin Ultrastructure and Hardness

*in vitro study*


Universidade de São Paulo, São Paulo, Brazil

**Objective:**

To investigate the influence of calcium phosphate-enhanced home whitening agents on human enamel and dentin surface microhardness and ultramorphology.

**Materials:**

- Ten intact molar crowns
- 15% carbamide peroxide + potassium nitrate + fluoride (Opalescence PF, Ultradent)
- 16% carbamide peroxide + potassium nitrate + fluoride (Whiteness Perfect, FGM)
- Potassium nitrate + fluoride + calcium + phosphate (Relief ACP, Discus Dental)
- 16% carbamide peroxide + potassium nitrate + calcium + phosphate (NiteWhite ACP, Discus Dental)
- 7.5% hydrogen peroxide + potassium nitrate + calcium + phosphate (DayWhite ACP, Discus Dental)
- 7.5% hydrogen peroxide + potassium nitrate + fluoride + calcium (White Class Ca, FGM)

**Methodology:**

Five intact molar crowns were used for ultrastructural analysis and five for microhardness tests. Each resulting coronal structure was cut in slices. After measuring the baseline Knoop Hardness Number (KHN) of the enamel and dentin, the slices were divided into six experimental groups and one control group (n=5). The groups were as follows: G1 = 15% CP; G2 = 16% CP; G3 = Ca and PO4; G4 = 16% CP with Ca and PO4; G6 = 7.5% HP with Ca.

**Results:**

Conventional whitening agents (G1, G2) and the gel with calcium (G6) cause KHN decrease (p = <0.05). The remineralizing and whitening agents with calcium and phosphate (G3, G4, G5) did not change KHN. A change in morphology was observed on dentin surfaces in G1, G2, and G5.
Conclusion:
The results indicated that the gels with calcium and phosphate added did not change the superficial enamel and dentin hardness nor did it change the tooth morphology. The conventional whitening gels that did not have remineralizing agents added to the formulation showed decreases on superficial hardness of enamel and dentin as well as morphological changes on tooth structure.
Effect of Relief ACP on Dentin Microhardness and Surface Morphology

*in vitro study*


**Objective:**

To investigate effects of Relief ACP on dentin microhardness and surface morphology of extracted human teeth compared to that of Satin Finish.

**Materials:**

- 20 dentin specimens
- Relief ACP (Discus Dental)
- Satin Finish (Discus Dental)

**Methodology:**

Twenty dentin specimens were prepared by grinding the enamel from the surface of human molars until the dentin was exposed. The sample surface was measured for Knoop Hardness Number (KHN) with a Leco Microhardness Tester (M-400-H1, St, Joseph, MI). The specimens were randomly assigned to three groups. Group A (N=4) served as the Control (100% humidity). Group B (N=8) was treated with Relief ACP (Discus Dental), while Group C (N=8) received treatments with Satin Finish (Discus Dental). The samples received 28 treatments of 30 minutes each. Prior to each treatment, the samples were immersed in pooled human saliva for 20 minutes. The KHN was measured after the last treatment, and the specimens were then processes for the SEM evaluation. The KHN data were analyzed using the One-way ANOVA and Student-Newman-Keuls methods.
Take-Home Whitening

Benefits of ACP Results:

There were no significant differences in the KHN values among the three groups before and after treatments. The changes in KHN were statistically different (p=0.032); however, there were no significant within-treatment differences for any of the three groups. The SEM evaluation showed deposits inside of the exposed dentin tubule openings in the specimens treated with Relief ACP, and most of the openings appeared fully blocked by the deposits. Such deposits were not observed in the Control samples and they were less evident in the Satin Finish group.

Conclusion:

The treatment with Relief ACP or Satin Finish does not change surface microhardness of human dentin, and treatments with Relief ACP produce deposits inside of the exposed dentin tubule openings.
Fluoride and Potassium Nitrate-Fluoride Whitening Agents: in vitro Caries Study

**in vitro study**


**Objective:**
To evaluate the effects of whitening agents containing amorphous calcium phosphate with fluoride (ACP-Fl) and potassium nitrate with fluoride (KN-Fl) on enamel caries initiation and progression

**Materials:**
- 15 human teeth
- 16% carbamide peroxide, ACP and fluoride (NiteWhite, ACP-Fl, Discus Dental)
- 15% carbamide peroxide with potassium nitrate and fluoride (Opalescence, KN-Fl, Ultradent Products)

**Methodology:**
Fifteen human teeth with sound enamel surfaces were divided into 3 portions. Each tooth portion was assigned to a treatment group: Group 1) No Treatment Control; Group 2) NiteWhite 16% carbamide peroxide, ACP and fluoride (ACP-Fl, Discus Dental); Group 3) Opalescence 15% carbamide peroxide with potassium nitrate and fluoride (KN-Fl, Ultradent Products). The teeth were treated according to the manufacturer’s guidelines followed by synthetic saliva rinsing on a daily basis for 14 days. Control tooth portions were exposed only to synthetic saliva rinsing. A modified ten Cate solution was used for in vitro enamel caries initiation and progression. The teeth were treated prior to lesion initiation and before lesion progression. Longitudinal sections were taken after the lesion initiation period and the lesion progression period for polarized light study and statistical analysis (ANOVA, DMR).

**Results:**
For both the lesion initiation and progression periods, significant differences were found between ACP-Fl and KN-Fl.

Mean lesion depths:
- **Lesion Initiation Period:** Control 156±17um; KN-Fl 95±12um (P<.05); ACP-Fl 72±14um (P<.05)
- **Progression Period:** Control 306±29um; KN-Fl 172±18um (P<.05); ACP-Fl 108±14um (P<.05).
Fluoride and Potassium Nitrate-Fluoride Whitening Agents: in vitro Caries Study

Mean Lesion Depths

<table>
<thead>
<tr>
<th></th>
<th>Group 1: Control</th>
<th>Group 2: Nite White — 16% carbamide peroxide, ACP and fluoride</th>
<th>Group 3: Opalescence — 15% carbamide peroxide with potassium nitrate and fluoride</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lesion initiation period</td>
<td>156±17um</td>
<td>72±14um (P&lt;.05)</td>
<td>95±12um (P&lt;.05)</td>
</tr>
<tr>
<td>Lesion progression period</td>
<td>306±29um</td>
<td>108±14um (P&lt;.05)</td>
<td>172±18um (P&lt;.05)</td>
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</tbody>
</table>

Conclusion:

Fluoride-containing whitening agents significantly reduce the susceptibility of enamel surfaces to in vitro caries initiation and progression compared with matched no treatment controls. When both ACP and fluoride (ACP-Fl) are present in the whitening agents, caries resistance was markedly improved over the whitening agent containing fluoride, but lacking ACP.
Effect of Take-Home Whitening Agent on Enamel Microhardness

*in vitro study*


**Objective:**

To evaluate the effect of remineralizing agents such as fluoride or amorphous calcium phosphate (ACP) during bleaching on surface enamel microhardness.

**Materials:**

- 16% carbamide peroxide with ACP (NiteWhite ACP, Discus Dental)
- 15% carbamide peroxide with fluoride (Opalescence PF, Ultradent)
- 9% hydrogen peroxide with fluoride (Tres White, Ultradent)

**Methodology:**

Nine human central incisors were cut in half longitudinally for a total of 18 surfaces. The sections were embedded into acrylic with the facial surfaces exposed. The prepared specimens were randomly divided into three groups (six specimens each), and assigned treatment with one of the three whitening agents being tested. Group 1 was treated with 16% carbamide peroxide with ACP (NiteWhite ACP); Group 2 was treated with 15% carbamide peroxide with fluoride (Opalescence PF); Group 3 was treated with 9% hydrogen peroxide with fluoride (Tres White).

Before bleaching was initiated, baseline data were collected on the Vickers surface hardness (VHN) of the enamel using a MicroMet 2100 series tester (Buehler, Lake Bluff, IL). Six hardness tests were conducted with 500 gm load for 15 seconds on each specimen for a total of 36 measurements per group. The specimens then underwent six consecutive one-hour cycles of bleaching with the assigned whitening agent. At the completion of the bleaching, another six enamel hardness readings were taken for each specimen. The data gathered were analyzed using ANOVA with p < 0.05 for significant differences.
Results:
All whitening systems produced a decrease in enamel hardness after six bleaching cycles. The VHN of Group 1 (NiteWhite ACP) decreased by 5.57%, the VHN of Group 2 (Opalescence PF) decreased by 10.49% and the VHN of Group 3 (Tres White) decreased by 11.91%. The decrease in enamel hardness for Group 1 specimens treated with 16% carbamide peroxide with ACP (NiteWhite) was significantly less than the decrease seen in other groups.

Conclusion:
Bleaching with NiteWhite ACP resulted in the least decrease in hardness compared with Opalescence PF and Tres White under the conditions of this study.
Effect of Remineralizing Agents on Enamel Microhardness After Bleaching

in vitro study


Objective:
To determine the potential of remineralizing agents to increase microhardness of enamel after bleaching

Materials:
- Five human incisors
- 15% carbamide peroxide (Opalescence, Ultradent)
- MI Paste (Ultradent)
- Relief ACP (Discus Dental)

Methodology:
Five human incisors were sectioned in half superior inferiorly. The halves were then mounted on cold cure acrylic for ease of manipulation. Six VH readings were then done per specimen on Micromet 2100 (Buehler, Lake Bluff, IL) with 500 gm load to establish baseline. Six cycles of bleaching with 15% carbamide peroxide (Opalescence, Ultradent) were then performed. Each cycle lasted one hour prior to placement of new solutions. The teeth again were tested for VH hardness with six readings per specimen. Upon completion of bleaching, the halves of the teeth were divided into either Group A (MI Paste, Ultradent) or Group B (Relief, Discus Dental). The systems were applied for 30 minutes then rinsed with tap water. The specimens again were tested for VH hardness with 6 readings per specimen. The procedure was repeated for a total of three remineralization cycles. All data gathered were analyzed using ANOVA with p<0.05 for significant differences.

Results:
After six cycles of bleaching, there was a significant decrease in hardness on all specimens from a VH of 313.1 to 280.8. After three applications of remineralization agents in both groups, all hardness measurements returned to baseline.

Conclusion:
There was a statistically significant decrease in enamel hardness for the bleaching system tested in this study. The application of remineralization agents reversed the decrease in enamel hardness to baseline after three applications.
A 180-Day Clinical Investigation of the Tooth Whitening Efficacy of a Bleaching Gel with Added Amorphous Calcium Phosphate

in vivo study


1Martin Giniger & Company, New York, NY, USA, 2Discus Dental, Culver City, CA, USA

Objective:
To determine if there are any significant long-term clinical benefits or side effects caused by the addition of amorphous calcium phosphate (ACP) to a professional 16% carbamide peroxide bleaching gel.

Materials:
• 16% carbamide peroxide gel containing ACP (NiteWhite ACP, Discus Dental)
• 16% carbamide peroxide gel without ACP (NiteWhite Excel 3, Discus Dental)

Methodology:
This study examined the effect of bleaching gel with added ACP in a subset of subjects (n=27) from a previously published short-term (n=50) study, in which two groups were assigned to use either an experimental ACP-containing gel or a similar “control” gel. Both groups used the product for four hours (or overnight) daily for 14 days. In the present study, the long-term ACP effects on tooth color, gingival health and three measures of dentinal hypersensitivity at post-treatment days +90 and +180 were assessed.

Results:
In the previously published study, the difference in tooth whitening efficacy at day +5 between the test group and the control group was only 0.19 shades relative to baseline, and was not statistically significant. In the present study, the differences between the groups had almost doubled at day +90, and were calculated to be 0.34 shades (statistically different t-test p=0.002). Furthermore, the differences had more than doubled again at day +180, with the ACP group subjects’ teeth being 0.78 shades lighter than the control group’s teeth (statistically different t-test p=0.002). Considered as a percentage, at day +180 the ACP group had retained nearly 10% more of their original whitening treatment result compared to control. There were no other significant differences found between the two groups. Tooth sensitivity, soft tissue health and gingival health remained similar to baseline levels.
Conclusion:
This study demonstrated that the 16% carbamide peroxide product with ACP offers 10% better long-term (six months) whitening efficacy than the traditional bleaching gel tested. The long-term safety of the product has also been demonstrated, as there were no adverse gingival or other effects seen at either day +90 or day +180.
Whitening Agents with ACP: Enamel Caries Formation and Progression

*in vitro* study


**Objective:**
To evaluate the effect of whitening agents containing amorphous calcium phosphate (ACP) on human enamel caries formation and progression

**Materials:**
- 15 human teeth
- 9.5% hydrogen peroxide ACP (DayWhite Excel 3, Discus Dental)
- 6% hydrogen peroxide ACP (NiteWhite Turbo, Discus Dental)
- 16% carbamide peroxide ACP (NiteWhite, Discus Dental)

**Methodology:**
Fifteen teeth with sound enamel surfaces were divided into four portions. Each tooth portion was assigned to a treatment group: Group 1) No Treatment Control; Group 2) DayWhite Excel 3 9.5% hydrogen peroxide ACP; Group 3) NiteWhite Turbo 6% hydrogen peroxide ACP; Group 4) NiteWhite 16% carbamide peroxide ACP. The teeth were treated according to the manufacturer’s recommendations followed by synthetic saliva, on a daily basis for 14 days. Control tooth portions were exposed only to synthetic saliva. A modified ten Cate solution was used for *in vitro* enamel caries formation and progression. The teeth were treated prior to lesion formation, and before lesion progression 1 and lesion progression 2 periods. Longitudinal sections were taken after lesion formation, lesion progression 1 and lesion progression 2 periods for polarized light study and statistical analysis (ANOVA, DMR).
Results:

Mean lesion depths were:

- **Lesion Formation Period:** Control 108±15um; DayWhite 93±11um; NiteWhite Turbo 48±7um (P<.05); NiteWhite 16% 105±12um.

- **Progression Period 1:** Control 171±18um; DayWhite 126±13um (P<.05); NiteWhite Turbo 96±9um (P<.05); NiteWhite 16% 132±12um (P<.05).

- **Progression Period 2:** Control 228±20um; DayWhite 165±17um (P<.05); NiteWhite Turbo 129±11um (P<.05); NiteWhite 16% 152±16um (P<.05).

<table>
<thead>
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<th>Group 1: Control</th>
<th>Group 2: DayWhite Excel 3 - 9.5% hydrogen peroxide ACP</th>
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</table>

Conclusion:

Whitening agents containing calcium phosphate have a reduced susceptibility to in vitro enamel caries lesion initiation and progression.
Tooth Surface Enhancement by a 16% Carbamide Peroxide Take-Home Bleaching Gel Containing ACP

*in vivo study*


**Objective:**
To evaluate the effectiveness of a dual-barrel, 16% carbamide peroxide equivalent, take-home bleaching gel containing amorphous calcium phosphate (ACP) in enhancing tooth surface smoothness and gloss.

**Materials:**
- 16% carbamide peroxide equivalent gel containing ACP (NiteWhite Excel 3 ZCP, Discus Dental)

**Methodology:**
Ten healthy adults wore a custom tray containing the test ACP gel for a minimum of four hours daily (or overnight) for two weeks. Evaluations of tooth color, surface roughness and gloss were made at baseline, one week, two weeks and +five days post treatment. The surface gloss index (SGI) and surface roughness index (SRI) measurements were performed by a single experienced examiner who evaluated the anterior teeth. The SGI utilizes a six-point subjective scale, and the SRI uses a four-point scale. For the tooth color evaluation a 16-point Vita Shade Guide score index was used. Mean “before” and “after” tooth color scores, SGI scores and SRI scores were calculated. Mean differences between baseline values and after-treatment values were also reported. A t-test was used to determine significant differences with alpha set at 0.05.

**Results:**
Compared to the baseline control values, the ACP gel showed a significant ($p < 0.01$) longitudinal percent improvement in tooth surface gloss and roughness at one week (SGI = 10.1%; SRI = 6.15%) and two weeks (SGI = 22.4%; SRI = 15.4%). Tooth color was also improved significantly compared to baseline, reaching a maximum shade change of 8.13 ($\pm 1.02$) units at day 14 (t-test, $p < 0.0001$). At the +five days post-treatment evaluation, no significant changes in tooth color, SGI or SRI were found compared to day 14 results ($p < 0.0001$).
**Conclusion:**

The dual-chambered, 16% carbamide peroxide equivalent, ACP-containing bleaching gel has superior tooth whitening and surface properties that include excellent tooth-whitening ability (8+ Vita Shade Guide improvements from baseline) and improving teeth luster (significantly increased enamel gloss and decreased enamel roughness compared to baseline).
The Clinical Performance of Professionally Dispensed Bleaching Gel With Added Amorphous Calcium Phosphate

in vivo study

Giniger M¹, MacDonald J², Ziemba S³, Felix H⁴; The Clinical Performance of Professionally Dispensed Bleaching Gel With Added Amorphous Calcium Phosphate, JADA, 136, 2005.

¹Martin Giniger & Company, New York, NY, USA; ²Discus Dental, Culver City, CA, USA

Objective:
To measure how the addition of amorphous calcium phosphate (ACP) to a professionally dispensed 16% carbamide peroxide equivalent bleaching gel affects tooth color and dentinal hypersensitivity.

Materials:
• 16% carbamide peroxide equivalent gel with calcium and phosphate
• 16% carbamide peroxide equivalent gel (NiteWhite Excel 3 Regular, Discus Dental)

Methodology:
Fifty healthy subjects were enrolled in a parallel, double-blind, two-cell randomized clinical study. All subjects had maxillary anterior tooth discoloration equivalent to or darker than Vita shade 3. A baseline examination was performed which included medical history, oral soft tissue examination, Vita shade tooth color scoring, Gingival Index scoring and self-reported sensitivity scoring. Subjects were randomized into test and control groups that were balanced with respect to age, sex, tooth color and sensitivity scores. The subjects were instructed to use the gel they were given (according to the manufacturer’s instructions for the commercially-available control product) once a day for a minimum of three hours or overnight for 14 days. Clinical re-examinations were performed on days 3, 7, and 14 as well as +5 days post-treatment.

Results:
The test group demonstrated significantly lower (P < .05) mean thermal sensitivity scores with baseline (day 14: 0.21 versus 0.31; day +5: 0.06 versus 0.18). The test group also showed substantially lower (P < .05) tactile sensitivity scores (day 14: 0.26 versus 0.48; day +5: 0.06 versus 0.19). At the conclusion of the study twice as many subjects were free of thermal sensitivity in the test group (80%) compared with the control group (40%) (P < .001). There was a similar significant percentage difference for tactile sensitivity. Both groups demonstrated equivalent and significant tooth color enhancements as compared with baseline (control: -7.73 shade change versus test: -8.12 shade change; P < .05).
Conclusion:
This study demonstrates that ACP could be added to a 16% carbamide peroxide equivalent bleaching gel and result in a significant reduction of clinical measures of dentinal hypersensitivity, both during and after treatment.
Microhardness and Ultramorphological Changes after Whitening: Calcium and Phosphate Benefits

*in vitro study*


Objective:

To investigate the influence of home bleaching agents with and without calcium and phosphate on human enamel and dentin surface microhardness and ultramorphology.

Materials:

- Five human molars
- 15% carbamide peroxide with potassium nitrate and fluoride (Opalescence PF, Ultradent)
- 16% carbamide peroxide with potassium nitrate and fluoride (Whiteness Perfect, FGM)
- 16% carbamide peroxide with potassium nitrate and ACP (NiteWhite ACP, Discus Dental)
- 7.5% hydrogen peroxide with potassium nitrate and ACP (DayWhite ACP, Discus Dental)
- 7.5% hydrogen peroxide with potassium nitrate, fluoride, and calcium (White Class Ca, FGM)

Methodology:

Five intact human third molars were sectioned into five wafers. All wafers had their baseline Knoop Hardness Number (KHN) of the enamel (E0) and dentin (D0) measured. Each wafer of the tooth was assigned to a group (n=5). G1=15% carbamide peroxide (CP) (Opalescence PF-Ultradent) applied for four hours per day; G2=16% CP (Whiteness Perfect, FGM), G3=16% CP with Ca and PO4 (NiteWhite ACP-Discus Dental), G2 and G3 followed same application time of group 1; G4=7.5% hydrogen peroxide (HP) with Ca and PO4 (Day White ACP-Discus Dental); G5=7.5% HP with Ca (White Class Ca-FGM), G4 and G5 were applied for one hour per day. After each session of bleaching treatment, specimens were stored in distilled water (37°C). The products were applied for two weeks, according to manufacturers’ instructions. Subsequent measurements of KHN were taken in the enamel (E1) and dentin (D1). Two specimens from each group were selected for ultramorphological investigation after final tests.
Results:
Conventional bleaching agents and the gel with calcium (Group 1 and Group 2) caused KHN decrease. Bleaching agents with calcium and PO4 (Group 3 and Group 4) did not change KHN. The obvious change of morphology was observed on enamel and dentin surfaces in Group 1, Group 2 and Group 5.
Dentin

Original magnification: 10,000X
Insets: 20,000X

Conclusion:

The bleaching agents with calcium and ACP did not change the superficial enamel and dentin microhardness and ultramorphology. Conventional bleaching agents and the gel with calcium caused microhardness decrease and ultramorphology changes on enamel and dentin.
Whitening Treatment Combined With Bioactive Materials

*in vitro study*


**Objective:**

To investigate the influence of bioactive materials on whitened surfaces and dentin using Knoop hardness test

**Materials:**

- Eight human teeth
- 15% carbamide peroxide, potassium nitrate, fluoride (Opalescence PF, Ultradent)
- 16% carbamide peroxide, potassium nitrate, fluoride, calcium, phosphate (NiteWhite ACP, Discus Dental)
- 15% carbamide peroxide, potassium nitrate, fluoride & glass-ceramic crystalized glass $P_2O_5$ –$Na_2O$–$CaO$–$SiO_2$ (Opalescence PF, Ultradent & Biosilicate, Vitrovita)
- 15% carbamide peroxide, potassium nitrate, fluoride + potassium nitrate, fluoride, calcium, phosphate (Opalescence PF, Ultradent + Relief ACP, Discus Dental)

**Methodology:**

Eight human teeth were sectioned into five wafers per tooth and divided into five experimental groups (n=8). The specimens were treated and mounted in intra-oral palatal retainers. Whitening treatments were performed for 14 days according to manufacturer’s instructions. Six Knoop hardness measures were taken for each specimen, three before and three after treatments. The data were compared by Student’s *t*-test ($\alpha = 0.05$).

**Results:**

Opalescence PF caused hardness decrease on enamel and dentin ($p<0.05$). NiteWhite ACP and the bioactive materials had a positive influence on the hardness of bleached enamel and dentin, except for the effect on enamel of the Biosilicate material when applied for five minutes one time per week, which showed a decrease in KHN.
Conclusion:

Whitening treatment can lead to alterations in the dental structure. Minimizing or eliminating the alterations in the whitened dental structure could bring benefits to patients. Adding bioactive materials to whitening treatments can minimize or eliminate the alterations in the dental structure.