






Does the combination of whitening toothpaste and hydrogen peroxide bleaching increase the surface roughness and change the morphology of a nanofilled composite?

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Aim: To evaluate changes in the surface roughness and morphology of a nanofilled composite following toothbrushing with a whitening (WT) or regular toothpaste (RT), alone or combined with 35% hydrogen peroxide bleaching (HP). **Methods:** Seventy disc-shaped nanofilled composite (Filtek Z350XT) specimens were randomly divided into groups (n=10): WT, RT, TB (without toothpaste – control) or the combinations WT/HP, RT/HP, TB/HP and HP. All groups underwent toothbrushing simulation (60,000 cycles) and bleaching treatment (4 sessions). Mean surface roughness (Ra, μm) was measured before (T_0) and after treatments (T_B). Surface morphology was assessed by scanning electron microscopy (SEM) at T_B . Mean Ra was analyzed using general mixed models and multiple comparisons by the Tukey-Kramer test ($\alpha=5\%$). **Results:** HP caused no surface roughness changes on the nanofilled composite after treatment ($p>0.05$). RT toothbrushing, combined or not with HP, increased the surface roughness ($p<0.05$). WT and WT/HP protocols had no effect on the surface roughness of the composite ($p>0.05$). The nanofilled composite submitted to RT toothbrushing combined with HP (RT/HP) presented substantial surface alterations under SEM, showing deep depressions and round-shaped defects. Toothbrushing with RT combined with the bleaching agent increased exposure of the inorganic fillers. **Conclusion:** WT toothbrushing, regardless of HP combination, or the single HP protocol had no effect on the surface roughness of the nanofilled composite. However, RT combined with HP negatively affected surface roughness and presented the most noticeable surface changes among groups.

Keywords: Tooth bleaching. Toothpastes. Composite resins. Microscopy, electron, scanning.

Introduction

Tooth whitening appears to be a permanent trend among patients due to its high impact on the quality of life and aesthetic self-perception¹. In-office bleaching treatments accelerate color changes compared to at-home techniques² given the use of highly concentrated hydrogen peroxide (HP) available in concentrations up to 40%³.

Over-the-counter products such as whitening toothpastes, powders, and strips are often used to whiten teeth or indicated as adjuvants to a whitening treatment. Their whitening mechanisms differ greatly. Whitening toothpastes, for example, usually present a high-level of abrasiveness to remove dental surface-adhered extrinsic stains⁴. Some contain compounds such as blue covarine or titanium dioxide which are deposited on the dental surface to make it seem brighter and whiter^{4,5}. However, overexposure to these products can harm the enamel or resin composites, increasing surface roughness and wear⁶⁻⁸.

Previous studies on bleaching using highly concentrated hydrogen peroxide have found adverse effects such as morphology alterations, increased surface roughness, and decreased surface microhardness⁹⁻¹¹. Moreover, hydrogen peroxide can interact with either the organic matrix or the inorganic fillers of microhybrid, nanohybrid, and nanofilled resins¹², leading to changes in the composites. Although its mechanism of action on teeth is based on the interaction of reactive oxygen species from the HP reaction with dentin chromophores³, HP could promote an oxy-reduction reaction in a resin-based material inducing surface changes, but would still be unable to reverse the color changes in the enamel¹².

As patients who undergo professional bleaching could present existing restorations¹³, they may use highly abrasive whitening toothpaste, assuming that these can increase the whitening result. Combining over-the-counter products with in-office bleaching could exacerbate the treatment's deleterious effects on the resin surface¹². In some clinical conditions, failing to replace the restoration can lead to biofilm formation¹⁴.

Given this context, this study evaluated the effect of a whitening toothpaste combined with in-office bleaching on the surface of a nanofilled resin. The null hypotheses postulated were that (I) bleaching with 35% HP would not increase surface roughness or change the nanofilled composite's morphology and (II) the combination of whitening toothpaste and 35% HP would not increase surface roughness or change the composite's morphology.

Methodology

Experimental design

Seventy disc-shaped nanofilled composite (Filtek Z350XT, 3M Oral Care, São Paulo, SP, Brazil) specimens (n=10) underwent brushing with whitening (WT), regular (RT) or without toothpastes (TB) alone or combined with bleaching performed with 35%

hydrogen peroxide (HP). The groups were evaluated at baseline (T_0) and after surface treatment (T_B). The variables consisted of surface roughness, measured according to the Ra parameter (mean roughness, in μm), and morphology of the resin composite surface, evaluated by scanning electron microscopy (SEM) at T_B .

Sample preparation

Two increments of a nanofilled composite (Filtek Z350 XT, 3M Oral Care, Sumaré, SP, Brazil, shade Enamel A2) were inserted into disc-shaped Teflon molds (6 mm diameter x 3 mm thickness). A Mylar strip and a glass slide were then pressed onto the top of the sample and a 500 g-load was applied for 10 s. A light-curing device (Bluephase – Ivoclar Vivadent, Barueri, São Paulo, Brazil, 1200 mW/cm² irradiance) was used to cure the composite resin for 20 s at a 1-mm distance from the specimen. Excess resin was removed with a no. 12 scalpel blade. The specimens were polished with descending grits of sandpaper discs (Sof-Lex, 3M Oral Care, St. Paul, MN, United States) for 10 s and discarded after every 5 procedures. Specimens were rinsed with distilled water and ultrasonically cleaned before being stored in a dark environment containing 5mL of artificial saliva – AS (1.5 mM CaCl₂, 0.9 mM Na₃PO₄, 0.15 mM KCl, pH 7.0)¹⁵. All specimens were fabricated and polished by a single operator.

Group division

Specimens were randomly assigned to seven experimental groups (n=10) according to surface treatment (toothbrushing/bleaching):

1. WT: brushing with whitening toothpaste;
2. RT: brushing with regular toothpaste;
3. TB: brushing without any toothpaste (distilled water);
4. WT/HP: brushing with whitening toothpaste followed by bleaching with 35% HP;
5. RT/HP: brushing with regular toothpaste followed by bleaching with 35% HP;
6. TB/HP: brushing without toothpaste (distilled water) followed by bleaching with 35% HP;
7. HP: bleaching with 35% HP without brushing.

Table 1 lists the composition of the restorative material and toothpastes used.

Table 1. Materials used and their respective composition.

| Material | Manufacturer | Composition | Abrasive particles |
|---------------------------------------|--|--|---|
| Nanofilled Resin Z350XT | 3M Oral Care, Sumaré, SP, Brazil | Bis-GMA, UDMA, TEGDMA and bis-EMA monomers. Non-agglomerated and non-aggregated silica (5-20nm) and zirconia (4-11nm). Aggregated silica-zirconia nanoclusters (0.6-10nm). | - |
| WT: Colgate Luminous White (RDA: 175) | Colgate-Palmolive, São Bernardo do Campo, SP, Brazil | Sodium carbonate, water, sorbitol, hydrated PEG-12, silica glycerin, sodium lauryl sulphate, cellulose gum, flavor, tetrasodium pyrophosphate, potassium hydroxide, phosphoric acid, cocamidopropyl betaine, 0.32% sodium fluoride, benzyl alcohol, saccharin sodium, sodium hydroxide, titanium dioxide (CI 77891). | Hydrated Silica |
| RT: Colgate Triple Action (RDA: 68) | Colgate-Palmolive, São Bernardo do Campo, SP, Brazil | Water, sorbitol, sodium lauryl sulfate, sodium monofluorophosphate, aroma, cellulose gum, tetrasodium pyrophosphate, benzyl alcohol, sodium saccharin, xanthan gum, sodium hydroxide. | Calcium carbonate and calcium bicarbonate |

Legend: Bis-GMA: Bisphenol A-Glycidium methacrylate; Bis-EMA: bis-phenol A-methacrylate; UDMA: urethane dimethacrylate; TEGDMA: triethylene glycol dimethacrylate.

Simulated brushing

The samples were fixed in a mechanical brushing machine (MSet, Nucci ME) and 60,000 brushing cycles were performed with 20-mm linear movements, at a frequency of 5Hz and a load of 200g⁷. Thus, 4.5 cycles per second were performed at 37°C. All specimens were brushed with a soft nylon toothbrush (Colgate Twister®, Colgate-Palmolive Company) with a flat head and immersed in a slurry prepared with WT or RT toothpaste and purified water in a 1:3 ratio^{7,16}. The TB and TB/HP groups were brushed with distilled water only. After the brushing cycles, the samples were rinsed in running water, ultrasonically cleaned and stored in AS.

Bleaching protocol

The bleaching gel (HP, 0.01g) was applied to the nanofilled composite of the groups undergoing the bleaching procedure, according to the manufacturer's instructions (Table 2). The resin surface was completely covered by the bleaching gel for 15 minutes and rinsed with distilled water. This procedure was repeated twice in each session, and the other three sessions were conducted at 72-h intervals. The samples were stored in AS at 37°C between sessions⁹.

Table 2. Bleaching gel information.

| Commercial name and Manufacturer | Composition | Manufacturer's instructions |
|---|--|--|
| Whiteness HP (FGM, Joinville, SC, Brazil) | 35% hydrogen peroxide, inert filler, deionized water, dyes, glycol, thickener. pH = 7.0. | HP liquid and the thickener should be mixed in a 1:3 proportion. Gel should be applied and refreshed every 15 minutes. The intense initial carmine coloration should change to transparent in the first five minutes of the reaction. It requires 7-day intervals between each session (up to 4 appointments). |

Surface roughness analysis

Mean surface roughness (Ra) was measured (Surf-Corder – SE 1700 – Kosakalab, Tokyo, Japan) at baseline (T_0) and after brushing/bleaching treatments (T_B) in three different directions, providing the average surface (Ra - μm) of each specimen. The roughness tester used operated with a cut-off of 0.25 mm, a speed of 0.25 mm/s and a measuring length of 1.25 mm¹⁷.

Surface morphology analysis

Two samples from each experimental group were left to dry overnight in an oven and sputter-coated with gold (MED 010. Balzers, Balzer, Liechtenstein). Surface morphology of the nanofilled composite was evaluated by scanning electron microscopy (DSM 940 A – Zeiss, Oberkochen, Germany) operating at 15 KV_a, and images were taken at 500 and 1000 x magnifications.

Statistical analysis

Ra data underwent exploratory analysis, which indicated the need for square root transformation ($\frac{1}{\sqrt{Ra}}$) to meet the normality parameters of parametric statistical tests. After transformation, the data were submitted to mixed models with additional treatment and repeated measures using the Proc Mixed procedure in the SAS software (SAS Institute Inc., Cary, NC, USA, Release 9.2, 2010). Multiple comparisons were performed using the Tukey-Kramer post-hoc test. Significance level was set at 0.05.

Results

Table 3 shows the mean surface roughness (Ra) before and after the treatments. At baseline (T_0), no significant differences were detected among the experimental groups ($p>0.05$). After treatments (T_B), brushing with RT produced significantly higher surface roughness than WT or TB, regardless of combination with bleaching treatment (HP) ($p<0.05$). After treatments (T_B), brushing with RT presented a higher mean surface roughness than the other groups ($p<0.05$). The bleaching treatment alone (HP) did not increase the surface roughness of the nanofilled composite ($p>0.05$), and no differences were observed on the surface roughness of the nanofilled composite when subjected to TB or WT alone or combined with HP ($p>0.05$).

Table 3. Mean and standard-deviation values of surface roughness (Ra) at baseline and after treatments.

| Groups | Baseline (T_0) | After treatments (T_B) |
|--------|---------------------------|----------------------------|
| WT | 0.64 (0.15) ^{Aa} | 0.70 (0.36) ^{Aa} |
| RT | 0.75 (0.19) ^{Aa} | 1.04 (0.43) ^{Bb} |
| TB | 0.63 (0.13) ^{Aa} | 0.57 (0.10) ^{Aa} |
| WT/HP | 0.66 (0.15) ^{Aa} | 0.70 (0.62) ^{Aa} |
| RT/HP | 0.65 (0.17) ^{Aa} | 1.19 (0.54) ^{Bb} |
| TB/HP | 0.70 (0.15) ^{Aa} | 0.75 (0.32) ^{Aa} |
| HP | 0.69 (0.08) ^{Aa} | 0.73 (0.19) ^{Aa} |

Means followed by different letters differ statistically at 5%. Uppercase letters compare time (T_0 and T_B), and lowercase letters compare the group treatments within each time.

Figures 1 and 2 present the representative SEM images of the nanofilled resin surface after the treatments. At low (500x – left side [A]) and high (1000x – right side [B]) magnifications, the RT (Fig. 1) and RT/HP (Fig. 2) groups showed the most predominantly irregular and coarse surfaces compared with the other groups. At higher magnifications, RT/HP (Fig. 2B) showed round-shaped depressions that could also be observed in the WT/HP (Fig. 2B) group, but to a lesser degree. The nanofilled composites subjected to brushing exhibited scratches compatible with the bristles marks, regardless of toothpaste (RT and WT, Fig. 1) or its absence (TB, Fig. 1). The group subjected only to bleaching (HP, Fig. 2) showed irregular surface to a less extent than the RT/HP (Fig. 2) group. However, instead of being irregular or scratched, the surface of HP and TB/HP (Fig. 2) presented structures with characteristics compatible with inorganic particles, as the uniform organic layer formed by the Mylar strip was slightly lost.

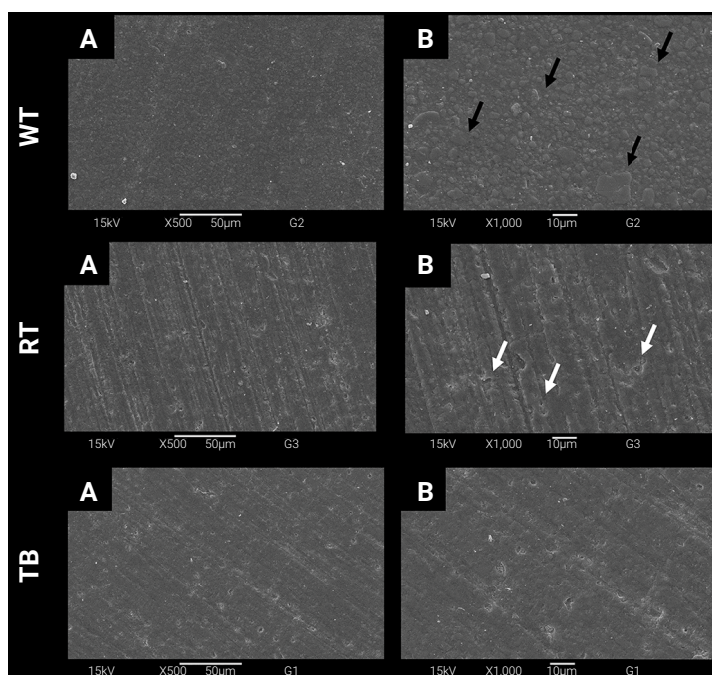


Figure 1. Representative SEM images of the resin surfaces after toothbrushing treatments (T_B) observed under 500x (left side - A) and 1000x (right side - B) magnifications. RT toothbrushing showed deeper depressions and a more irregular surface compared with WT and TB, including a higher predominance of round-shaped defects (white arrows). WT toothbrushing exposed structures compatible with inorganic particles (most visible defects pointed out by black arrows).

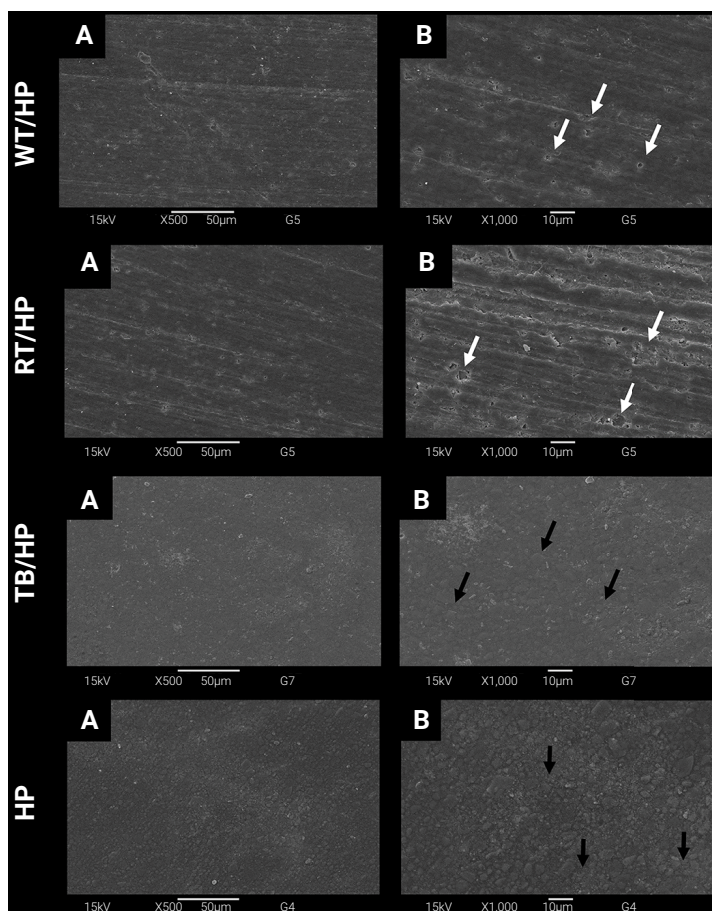


Figure 2. Representative SEM images of the resin surfaces after toothbrushing treatments followed by HP application (T_0) observed under 500x (left side - A) and 1000x (right side - B) magnifications. Combination of toothpastes and 35% hydrogen peroxide bleaching (HP) seemed to increase surface roughness, as round-shaped defects (white arrows) are more pronounced in the WT/HP and RT/HP treatments, with RT/HP presenting the most affected surface. HP application alone or combined with toothbrushing without toothpaste (TB) exposed structures compatible with inorganic particles (black arrows); however, the irregularities promoted by bleaching were more evident when RT toothbrushing was performed before bleaching.

Discussion

SEM analysis showed that 35% HP bleaching treatment, applied according to the manufacturers' recommendations, exposed the inorganic particles in the resin surface (HP and TB/HP groups) but did not significantly increase its surface roughness (R_a). Roughness increased only when 35% HP bleaching was performed after toothbrushing with regular toothpaste (RT); however, RT alone increased the R_a to the same level as RT/HP. Thus, the first null hypothesis was rejected since bleaching with high-concentrated HP did not increase surface roughness but changed the morphology of the nanofilled composite surface.

Findings on the effects of 35% hydrogen peroxide on the surface roughness of composite materials are still controversial^{12,17}, as the composite type, bleaching agent concentration and application protocol vary between studies. Some studies suggest

that at-home or in-office bleaching treatments can increase the surface roughness of microfilled and microhybrid composites^{18,19}. Moreover, evidence shows that dental bleaching with peroxide gels can modify the surface properties of nanofilled resins²⁰. The reason behind this reaction is still unknown, but one hypothesis is that HP could initiate an oxidative breakdown of the polymeric chains²¹. This event would be more pronounced in the unreacted double bounds of the resin²², but would not be limited to them, as HP could react with the carbon single bond of the resin network²³. Additionally, HP free radicals can influence the disintegration of inorganic filler and organic matrix components, probably by facilitating water absorption²¹.

A rougher surface could result in increased biofilm formation, similar to a previous *in vitro* evaluation in which *S. Mutans* and *S. Sanguinis* biofilms increased after 40% HP bleaching²⁴. Thus, a scenario in which bleaching causes changes to the resin surface could affect the long-term clinical performance of restorations. Conversely, other studies corroborate our findings that HP did not significantly increase the surface roughness of the composite^{17,25}. According to Fernandes et al. (2020), 35% HP bleaching did not alter the surface of the nanofilled resin, but it negatively affected the surface roughness of a microhybrid resin²⁶. Differences in bleaching gels (type, concentration, pH and application protocol) and in the resin (characteristics, composition and concentrations of inorganic particles and organic matrix) used may explain the controversial results.

In-office bleaching with peroxide gels is a widespread technique due to patients' high demand for aesthetics and because it represents a conservative and minimally invasive approach to dental structure compared to other restorative treatments¹. However, bleaching procedures usually occur up to the second premolar dentition, which may present existing restorations with different types of resin composite and different clinical service times. The impact of bleaching gels on the resin surface should be considered when deciding on whether to replace restorations. However, bleaching will cause noticeable color changes in the dental structure, leading to a color mismatch between the teeth and the restoration²⁷, which is an important clinical factor to consider when deciding whether to replace restorations. Thus, although HP alone did not significantly increase the surface roughness of the nanofilled composite, its replacement due to color discrepancy with that of the bleached teeth can still occur.

Another factor to consider is that bleaching should be performed on patients with good oral health and, consequently, with frequent toothbrushing habits^{1,2}. The groups that combined toothbrushing/toothpaste and HP bleaching were tested to simulate a clinical scenario¹⁵. Moreover, the trend to use whitening toothpastes as an over-the-counter bleaching option often exposes restorations to supervised and self-administrated bleaching⁴. Even though WT toothbrushing, alone or combined with 35% HP, did not increase the roughness of the nanofilled composite, surface morphology evaluation of the WT/HP group showed small rounded defects that were not detectable in the WT and HP groups. In contrast, the WT and HP groups showed exposure of the inorganic fillers, which is in line with a previous study in which WT was able to remove the organic matrix from the Z350 resin surface²⁸. Hence, we also rejected the second null hypothesis as toothbrushing with whiten-

ing toothpaste following in-office bleaching affected the surface morphology of the composite resin.

In line with our results, a previous *in situ* evaluation found that toothbrushing with Colgate Luminous Whiteness for three months did not increase the surface roughness of Filtek Z350²⁹, but was able to significantly increase the roughness of a nanohybrid composite (Tetric N-Ceram, Ivoclar Vivadent). Although filler size is not a determining factor in the degradation process of the composite, larger filler sizes usually increase the mean surface roughness values²⁹. Tetric N-Ceram nanohybrid has fillers ranging from 40 to 3,000 nm, whereas Filtek Z350 presents non-agglomerated fillers from 5 to 20 nm and agglomerated fillers from 600 to 1,400 nm. Moreover, shape of the filler, distance between fillers, the composite matrix, filler adhesion to the matrix and the degree of conversion also influence the performance of these composites. Even though that *in situ* evaluation performed no bleaching procedures with HP, our investigation found that bleaching did not increase the surface roughness of the composite. Conversely, RT alone or combined with bleaching (HP) increased the surface roughness of the nanofilled composite and resulted in greater surface morphology alterations after treatments.

Toothpaste abrasiveness is measured by the relative dentin abrasivity (RDA) and, according to the International Organization for Standardization (ISO), should not exceed a RDA of 250³⁰. The regular toothpaste (RT) tested here has low abrasiveness according to the RDA classification (68), whereas the WT has a much higher RDA (175), but still within the established ISO parameters. Colgate Triple Action (RT) contains calcium carbonate³¹, a low-abrasive component compared to the hydrated silica present in Colgate Luminous White (WT), which is an intermediate abrasive agent, but more efficient at removing stains than other abrasives due to the adjuvant performance of pyrophosphate²⁹. Aside from the inherent abrasiveness of these components, the prolonged brushing protocol used (60,000 cycles), which could clinically represent up to 6 years of tooth brushing⁵, also influenced the results.

Prolonged simulated brushing may have led to greater polishing and, consequently, to a loss of resin surface volume^{6,8,32}. Since nanofilled resins have smaller particle sizes, the fillers may have been polished in the same proportion as the organic matrix during the brushing process, which could explain the maintenance of the Ra. Despite no increase in Ra for the WT or WT/HP groups, WT/HP presented areas where round-shaped defects could represent the loss of inorganic filler. Thus, these data should be interpreted with caution, and further analysis, *i.e.*, measurement of surface loss, is necessary to confirm the safety of combining whitening toothpaste and in-office bleaching on the surface of nanofilled composites.

Although the prolonged simulated brushing protocol may explain the greater polishing caused by the hydrated silica in the WT groups, the opposite may have occurred with the calcium carbonate and calcium bicarbonate abrasives in the RT-treated groups. This low-abrasiveness toothpaste (RDA 68), used in a prolonged number of brushing movements, may have failed to promote homogenous surface polishing³². This was evident from the significant increase in surface roughness and the SEM evaluation which, in general, showed deep bristle marks and a greater number of round-shaped

defects. More noticeable in the RT/HP group, these rounded defects may suggest that HP could have a synergistic effect with RT toothbrushing, increasing the defects on the surface of the composite. In another study, SEM evaluation showed that hydrogen peroxide application increased the surface porosity of different composites²¹. This reinforces the clinical concern that bleaching could induce an adverse effect when performed on existing restorations.

A recent systematic review showed that a 0.2 μm increase in surface roughness is no longer considered an adequate threshold for predicting biofilm formation, suggesting that topographical changes on the resin surface have a major impact on the clinical formation of bacteria³³. Park et al.¹⁴ (2012) showed that morphological alterations were more important than surface roughness for the accumulation of *S. Mutans* on nanofilled composite (Filtek Z350). Although an increase in roughness of more than 0.5 μm cannot guarantee that resin brushed with RT subjected to in-office bleaching may accumulate more biofilm, the greater predominance of morphological defects detected by SEM evaluation could indicate that these restorations are more likely to develop recurrent caries, depending on the patient's brushing habits and caries risk³⁴.

A viable alternative to overcome this situation could be to polish the restoration after in-office bleaching, a procedure that could increase the resistance of nanofilled composites to degradation caused by HP by-products³⁵. However, the type of polishing system should be chosen carefully³³, since polishers impregnated with silicon carbide particles seem to provide a smoother surface³⁵.

In short, application of 35% HP to the resin surface did not alter the surface roughness of the nanofilled composite, but observing the behavior of bleaching on a resin surface subjected to brushing highlighted that this combination may not be beneficial to the resin structure. Further studies are essential to determine the effects of whitening toothpastes on the surface volume of an existing composite, as this could directly impact on the bleaching action.

Within the limitation of this study, we may conclude that although bleaching with 35% HP influenced the surface morphology of the resin, it did not increase surface roughness alone or combined with whitening toothpaste brushing. However, brushing with a regular toothpaste negatively affected the surface roughness and morphology of the nanofilled composite, regardless of its combination with high-concentrated hydrogen peroxide.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be considered as a potential conflict of interest.

Author contribution

The authors C.B.T and G.M.S.M.M. performed the laboratorial procedures and analyses. The authors C.B.T. and M.K. wrote the initial draft of the manuscript. The authors V.C., F.L.B.A and C.P.T. design the study, supervised the students, and edited the writing. All authors actively participated in the manuscript's findings, revised, and approved the final version of the manuscript.

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