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UNIVERSIDADE ESTADUAL PAULISTA
"JÚLIO DE MESQUITA FILHO"
Instituto de Ciência e Tecnologia
Campus de São José dos Campos



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# Effect of whitening mouthrinses on color change of stained resin composites

Efeito de enxaguatórios clareadores na alteração de cor de resinas compostas previamente manchadas

**Mariane Cintra MAILART[1], Érica CRASTECHINI[1], Juliana Boa Sorte de OLIVEIRA[1], Carlos Rocha Gomes TORRES[1], Taciana Marco Ferraz CANEPPELE[1], Alessandra Bühler BORGES[1]**

1 - Universidade Estadual Paulista "Júlio de Mesquita Filho", Institute of Science and Technology, Department of Restorative Dentistry, São José dos Campos, SP, Brazil

## ABSTRACT

**Objective:** The present study compared the effect of whitening mouthrinses (WM) on the color change of stained resin composites (RC). **Material and Methods:** Cylindrical specimens (6mm-diameter and 1mm-thickness) were prepared with the following RC (n=60/group): Filtek Z350XT (Z350- methacrylate-based), Admira Fusion (AD- ormocer-based), TPH3 (TPH- methacrylate-based), and Beautifil II (BII- giomer/methacrylate-based). The initial color was assessed with reflectance spectrophotometer using CIE L*a*b* system. The specimens were immersed in staining broth during 14 days, submitted to color evaluation ($\Delta E_1$) and randomly allocated in 4 subgroups (n=15), according to WM adopted: Listerine Whitening (LW-2% hydrogen peroxide), Plax Whitening (PW-1.5% hydrogen peroxide), Bromelain/papain (BP-experimental solution), and Deionized water (DW-negative control). The whitening cycle consisted of RC immersion in WM for 1 min and in artificial saliva for 30 min, simulating 12 weeks, and final color assessment was performed ($\Delta E_2$). Color change data were analysed by ANOVA and Tukey's tests ($\alpha=5\%$). **Results:** After staining, TPH showed the lowest $\Delta E_1$ values and Z350 showed the highest color change (p=0.001). The whitening effect promoted by LW was significantly higher than color alteration obtained with PW ($\Delta E_2$), and BII showed the highest color change values ($\Delta E_2$) after whitening cycle. **Conclusion:** LW exhibited the greatest whitening potential on stained RC, mainly with the Giomer (Beautifill II) and the Ormocer-based (Admira Fusion) materials. Bromelain/papain solution showed no whitening effect on stained RC.

## KEYWORDS

Giomer; Hydrogen peroxide; Ormocer; Resin composite; Whitening mouthrinses.

## RESUMO

**Objetivo:** O presente estudo comparou o efeito de enxaguatórios clareadores (EC) na alteração de cor de resinas compostas (RC) previamente manchadas. **Material e Métodos:** Espécimes cilíndricos (6mm de diâmetro e 1mm de espessura) foram preparados com as seguintes RC (n=60/grupo): Filtek Z350XT (Z350- metacrilato), Admira Fusion (AD- ormocer), TPH3 (TPH- metacrilato), e Beautifil II (BII- giomer/metacrilato). A cor inicial foi mensurada com espectrofotômetro de reflectância utilizando o sistema CIE L*a*b*. Os espécimes foram imersos em um caldo de manchamento durante 14 dias, submetidos a avaliação de cor ($\Delta E_1$) e alocados aleatoriamente em 4 subgrupos (n=15), de acordo com EC adotado: Listerine Whitening (LW-peróxido de hidrogênio a 2%), Plax Whitening (PW- peróxido de hidrogênio a 1,5%), Bromelina/papaína (BP-solução experimental), e Água deionizada (AD- controle negativo). O ciclo clareador consistiu na imersão da RC no EC por 1 min e na saliva artificial por 30 min, simulando 12 semanas, e a cor final foi mensurada ($\Delta E_2$). Os dados de alteração de cor foram analisados pelos testes ANOVA e Tukey ($\alpha=5\%$). **Resultados:** Após o manchamento, TPH apresentou o menor valor de $\Delta E_1$ e Z350 apresentou a maior alteração de cor (p=0,001). O efeito clareador promovido pelo LW foi significativamente maior que o obtido com o PW ($\Delta E_2$) e BII teve a maior alteração de cor ($\Delta E_2$) após o ciclo clareador. **Conclusão:** LW exibiu maior potencial clareador nas RC manchadas. BII apresentou maior alteração de cor em resposta à ação clareadora de ambos enxaguatórios à base de peróxido de hidrogênio testados.

## PALAVRAS-CHAVE

Giomer; Peróxido de hidrogênio; Ormocer; Resina composta; Enxaguatório clareador.

## INTRODUCTION

Esthetic demands in daily clinical practice are increasing over the years. Perfect smile involving whiter teeth and restorations with optical properties similar to the tooth structure are often requested by patients. In this sense, resin composites are widely used for direct restorations in both anterior and posterior teeth, and tooth whitening techniques are commonly performed to minimize tooth discoloration [1, 2].

Traditional modalities of tooth whitening are performed with professional supervision (at-home and in-office techniques). Nevertheless, over-the-counter products such as whitening mouthrinses, painting on gels, toothpastes, and whitening strips are often available to consumers. The whitening mouthrinses are usually composed by low-concentrated hydrogen peroxide solutions and their ability to result in tooth color alteration has been demonstrated in previous studies [3,4].

As an alternative to peroxide, vegetative enzymes such as bromelain and papain have been investigated as stain removal agents [5, 6]. Bromelain is extracted from pineapple and other immature fruits [7]. This substance contains different proteases such as peroxidases, phosphatases, catalases and other substances with phytomedical purposes [7, 8]. Papain is a proteolytic enzyme of cysteine peptidases extracted from papaya plant. The benefit of papain is related to hydrolysis of amide substrate-based with application in industrial sectors, as food industry and biomedical approaches [9]. These enzymes have been previously investigated in relation to tooth whitening [4].

Although the improvements of chemical and mechanical properties of resin composites are expected to result in color stability of restorations, the differences in materials composition, such as type of organic matrix, size and type of particles influence its staining potential [10,11]. The challenges caused by the exposition to saliva, stains, acids and alcoholic food / beverages contribute to the materials discoloration [2, 11-14]. It has been reported that 50% of direct nanohybrid composite restorations presented surface staining after 2 years [15].

The effect of conventional whitening treatments on tooth-colored restorations has been previously shown [16, 17], contributing to reduce staining and favoring the color recovery of resin composites [18, 19]. Nevertheless, the potential of color alteration of the restorations resulting from whitening mouthrinses has not been properly described, although these products are easily available for consumers.

Therefore, this study aimed to compare the effect of whitening mouthrinses on the color change of stained resin composites. The null hypothesis tested was that there was no difference among the effect of the whitening mouthrinses on the color alteration of the stained resin composites tested.

## MATERIAL AND METHODS

### Experimental design

This in-vitro study followed a factorial 4×4 design, considering resin composites (at 4 levels: Z350 – Filtek Z350XT, AD – Admira Fusion, TPH – TPH3, and BII – Beautifil II), and whitening mouthrinses (at 4 levels: LW - Listerine Whitening, PW – Plax Whitening, BP – bromelain/papain experimental solution, and deionized water as the control group).

### Sample size calculation

A power of 80% and significance level of 5% was adopted to determine the sample size. With a standard deviation set at 2 [20] and a minimal difference to be detected of 2.7 (acceptable color difference threshold using CIE L*a*b* system) [21], 14 samples per group were required, considering 4 treatment groups. Thus, 15 samples per group were used. The sample size calculation was performed using an online software (http://estatistica.bauru.usp.br/calculoamostral/index.php).

## Specimen's preparation

The composition of the tested resin composites is described in Table I. Cylindrical specimens (6 mm diameter and 1 mm thickness) were prepared for each material (color A2) using a stainless steel matrix (n=60/material). The materials were inserted in a single increment and a mylar strip was placed on the resin surface to produce a smooth and standardized surface. They were light-cured (Radii Cal curing light, SDI, Victoria, Australia; 440-480nm; 1200 mW/cm$^2$) for 40 s according to manufacturer's instructions.

The specimens were attached to a metal holder and polished with sequential aluminum oxide abrasive papers (#1200, #2400, and #4000; FEPA-P, Struers, Ballerup, Denmark) mounted in a polishing machine (DP-10, Panambra, São Paulo, SP, Brazil) at 600 rpm for 60 s each under water cooling. The specimens were placed in an ultrasonic bath with deionized water for five minutes between each paper change and after polishing procedures to remove residues of abrasive grains.

## Color assessment

The initial color assessment of each specimen was performed using a reflectance spectrophotometer with an integrating sphere (CM 2600d - Konica Minolta, Osaka, Japan), according to the CIE L*a*b* system [22]. The device was adjusted to use the D65 standard light source with 100% ultraviolet light (UV) and specular component included (SCI). The observer angle was set at 2º adjusted to a small reading area (SAV). The color of each sample was measured three times and averaged.

The coordinate values (L*, a*, b*) locates the color of an object in a three-dimensional (3D) color space. The L* axis represents the degree of lightness within a sample and ranges from 0 (black) to 100 (white); a* values correspond to color on the axis of red-green; and b* values correspond to yellow-blue axis.

## Staining

The specimens were stored in individual containers with 2 ml of a staining broth solution (1.5 g of finely ground instant coffee; 1.5 g of finely ground instant black tea, 1.1 g of gastric mucin dissolved into 500 ml of water, 0.33 ml of FD&C Red 40, 0.33 ml of FD&C Yellow 5, 41.6 ml of red wine, 0.125g of methylparaben and 0.075 g of propylparaben) for 14 days (adapted from Wozniak et al., 1991) [23] under agitation. The pH of the solution was measured (pH=6.9), and it was renewed in the 7$^{th}$ day of the experiment [24]. After staining, the specimens were washed with deionized water and a second color assessment was performed with the same specifications described previously. The stained specimens were stored in closed containers (100% relative humidity) preventing their dehydration during color measurement procedures.

The ΔL, Δa, and Δb values were calculated comparing the initial color and after staining assessments, and color alteration was determined ($\Delta E_1$) according to the following formula:

$$\Delta Eab = \sqrt{\left[(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2\right]} \quad (1)$$

**Table I -** Resin composites specifications

| Composite | Manufacturer | Classification (Type of particles) | Filler content (wt %)/Fillers | Resin matrix |
|---|---|---|---|---|
| Filtek Z350XT | 3M ESPE, St. Paul, MN, USA | Nanofilled | 78.5% / Silica, zirconia, aggregated zirconia/ silica | Bis-GMA, UDMA, TEGDMA, Dimethacrylate |
| Admira Fusion | VOCO GMBH, Cuxhaven, Germany | Nanohybrid | 84% / Barium, aluminum silicate glass, silica | ORMOCER® |
| TPH3 | Dentsply Sirona, York, Pennsylvania, USA | Nanohybrid | 60% / Silanized aluminum borosilicate glass, barium glass fluorine aluminum borosilicate silanized, silica | Bis-GMA, EDAB |
| Beautifil II | Shofu Dental Corporation, Kyoto, Japan | Nanohybrid | 83.3% / S-PRG fillers (pre-reacted multifuncional fluoro-boro-alumino-silicate glass particles) | Bis-GMA, TEGDMA |

Bis-GMA: Bisphenol A diglycidyl methacrylate; EDAB: ethane 1,2-diamineborane; ORMOCER: Organically Modified Ceramic; TEGDMA: Triethylene glycole dimethacrylate; UDMA: urethane dimethacrylate.

### Whitening procedures

The stained resin composites were randomly divided, according to $\Delta E_1$ values, into the following four whitening mouthrinse groups (n= 15): LW (Listerine Whitening); PW (Plax Whitening), BP (Bromelain/papain experimental solution), and DW (deionized water – negative control group) (Table II). The whitening cycle consisted of 1 min immersion in 1.5 ml mouthrinses, followed by 30 min in artificial saliva. The specimens were stored in 100% relative humidity overnight. The artificial saliva was composed by 22.1 mmol/l hydrogen carbonate, 16.1mmol/l potassium, 14.5 mmol/l sodium, 2.6 mmol/l hydrogen phosphate, 0.8 mmol/l boric acid, 0.7 mmol/l calcium, 0.4 mmol/l thiocyanate, 0.2 mmol/l magnesium (pH = 7.4) [25].

This cycle was repeated 84 times, which simulates 12 weeks (three months) of whitening mouthrinses use [3]. At the end of the cycle, the specimens were washed with deionized water and the final color assessment was performed ($\Delta E_2$), comparing the values obtained after whitening with the values obtained after staining.

### Statistical analysis

Data were analyzed for normality by the Shapiro-Wilk test. Analysis of variance (ANOVA) parametric test followed by Tukey's test were used. One-way ANOVA was adopted for comparison among resin composites after staining. Aiming to compare the effect of the whitening mouthrinses on the different materials, two-way ANOVA was applied. The significance level used was set at 5%.

## RESULTS

$\Delta L$, $\Delta a$, $\Delta b$ and $\Delta E_1$ values after staining are expressed in Table III. One-way ANOVA revealed significant differences among the resin composites tested after staining ($p<0.05$). Tukey's test showed that Filtek Z350XT presented the higher values of color change ($\Delta E_1$) and yellowing ($\Delta b$). TPH3 presented the lowest color change after staining, as well as lower $\Delta L$ and $\Delta a$ values compared to the other materials. Admira Fusion and Beautifil II presented intermediate color alteration values. All materials tested presented $\Delta E$ above the acceptability threshold ($\Delta E=2.7$).

Regarding whitening efficacy factor, the experimental solution bromelain/papain-based resulted in $\Delta E_2$ values similar to deionized water negative control ($p=0.975$) for all composites tested. The LW mouthrinse promoted higher whitening effect (higher color alteration of the stained materials) than PW ($p=0.016$). For resin composites, BII presented the highest whitening effect ($\Delta E_2$) when compared to the other composites tested ($p<0.05$). Z350 and AD presented intermediate results and did not

**Table II -** Mouthrinses specifications

| Rinse | Composition | Manufacturer |
|---|---|---|
| Listerine Whitening (LW) | Water, 8% alcohol, 2% hydrogen peroxide, sodium phosphate, poloxamer 407, sodium lauryl sulfate, sodium citrate, mint flavor, menthol, eucalyptol, sodium saccharin, sucralose | Johnson & Johnson, São José dos Campos, SP, Brazil |
| Colgate Plax Whitening (PW) | Water, sorbitol, ethanol, 1.5% hydrogen peroxide, poloxamer 338, polysorbate 20, methyl salicylate, menthol, sodium saccharin, CI 42090 | Colgate-Palmolive, São Bernardo do Campo, SP, Brazil |
| Bromelain/papain (BP) | Water, 0.50% bromelain, 0.50% papain, methylparaben, propylparaben | University Laboratory |

**Table III -** Means and standard-deviation of CIE L*a*b* parameters and color change after staining ($\Delta E_1$). Different capital letters show significant differences for the resin composite within each parameter (columns)

| Resin composites | ΔL | Δa | Δb | $\Delta E_1$ |
|---|---|---|---|---|
| Filtek Z350XT | -6.77±1.95 A | 2.17±0.56 B | 3.58±1.87 A | 8.23±1.77 A |
| Admira Fusion | -4.34±1.08 B | 3.39±0.57 A | 2.17±0.86 B | 6.02±1.01 C |
| TPH3 | -2.14±0.66 C | 0.94±0.24 C | 1.87±0.54 B | 3.07±0.55 D |
| Beautifil II | -7.03±1.11 A | 0.55±0.23 D | 2.36±0.95 B | 7.52±1.05 B |
| p values | 0.001* | 0.001* | 0.0000001* | 0.001* |

*asterisks mean significant p values

show significant differences of color change after whitening between them (p=0.99). TPH showed the lowest whitening effect ($\Delta E_2$). Table IV shows the results of the Tukey's test among the different stained materials and mouthrinses tested (Table IV).

## DISCUSSION

The color stability of resin composites represents an important condition in esthetic restorations, and it depends on several factors [26, 27]. The central factors are the composition and proportion of the organic matrix and inorganic content in the material, since they are related to the hydrophilic properties and consequently, staining potential of resin composites [28, 29]. Then, the lower the volume of inorganic fillers, the higher the water sorption extent [10, 11].

TEGDMA is a hydrophilic monomer contributing to water sorption in resin composites. When this monomer is combined with Bis-GMA, a higher water uptake is detected [23, 28]. Since Filtek Z350XT and Beautifil II are composed by Bis-GMA and TEGDMA, this may explain why these materials presented the highest color change after staining ($\Delta E_1$= 8.23 and 7.52, respectively).

The susceptibility to staining may be also explained by the filler's characteristics. Although Filtek Z350XT has 78.5% filler content, this composite presents an aggregated zirconia/silica clusters and these structures may promote gaps among them, favoring staining [11, 24]. On the other hand, TPH3 showed the lowest color alteration values ($\Delta E_1$= 3.07), despite of its lower filler content (60% wt). This material presents barium fillers, which has shown to restrict water/pigments absorption on material surface due to the high reactivity of barium with water [28].

It must be highlighted that, despite the differences among the materials, all resin composites tested presented $\Delta E_1$ values above 2.7 (acceptability threshold) [21], indicating that these restorations would request replacement, repolishing, or whitening for color adjustments [30].

In the present study, the specimens were artificially stained aiming to standardize resin composite discoloration in order to observe the whitening effect of mouthrinses. Then, the specimens were submitted to whitening cycle with the different products. The whitening efficacy was observed only for the LW and PW products. This whitening capacity has is attributed to the hydrogen peroxide component [4]. Thus, the null hypothesis was rejected.

Hydrogen peroxide is able to reduce colored molecules, breaking down the complex pigments structure. As a result of the oxidizing process, chromophores exhibit a decreasing in light absorption, resulting in the perception of a whiter substrate [31, 32]. The LW group showed overall higher whitening efficacy in comparison to PW, which might be attributed to its higher concentration of hydrogen peroxide. Despite the low concentration of peroxide in these products, they were able to change the color of the stained composites, due to the longer period used with mouthrinses compared to professionally supervised whitening treatments.

Bromelain/papain and deionized water (control group) presented similar results, indicating no whitening effect provided by bromelain/papain-based experimental solution. Previous in-vitro studies demonstrated that the incorporation of these enzymes in toothpastes resulted on tooth stain removal [5, 6, 33]. The mechanism of action of vegetative enzymes is related to their proteolytic activity in disrupting the protein portion of the acquired pellicle and, consequently, removing the dye pigments that are attached to the proteins [33]. Nevertheless,

**Table IV -** Means and standard deviation values for each stained resin composite after whitening cycle ($\Delta E_2$)

| Whitening mouthrinse | Resin Composites | | | |
|---|---|---|---|---|
| | Filtek Z350XT | Admira Fusion | TPH3 | Beautifil II |
| Listerine Whitening | 2.61±0.83 Aa | 3.16±0.30 Bab | 2.37±0.39 Ba | 3.66±0.82 Bb |
| Plax Whitening | 2.36±0.95 Aab | 2.82±0.71 Bb | 2.21±0.40 Ba | 3.02±0.37 ABb |
| Bromelain/Papain | 2.33±0.60 Ab | 1.87±0,87 Aab | 1.30±0.47 Aa | 2.40±0.38 Ab |
| Deionized water | 2.23±0.47 Ab | 1.79±0.92 Aab | 1.29±0.44 Aa | 2.40±0.53 Ab |

Groups with the same capital letters refer to whitening rinses comparisons for the same resin and groups with lowercase letters are related to materials comparisons after whitening.

bromelain/papain efficacy is controversial, since no significant tooth color alteration was observed in a previous study testing these actives in solutions [4], as observed in our study with stained composites. Thus, its whitening efficacy may be a result of the association of the enzymatic action with the abrasive action of the toothpastes.

Some resin composites were more susceptible to the whitening effect of the mouthrinses, decreasing $\Delta E_2$ values. Beautifil II presented the most evident whitening potential after mouthrinses application compared to the other composites tested. This resin is classified as a Giomer, an ion releasing material containing surface pre-reacted fluoro-boro-alumino-silicate glass fillers (S-PRG). Despite its high filler content (83.3%), hydrogen peroxide was able to effectively act on giomer material organic matrix [16]. In order to contribute to the ion releasing process, Beautifil II organic content presents hydrophilic monomers, contributing to diffusion of hydrogen peroxide into the organic matrix [34]. TPH3 showed the lower differences in $\Delta E_2$ values after the whitening cycle. Possibly, this condition is related to the limited color change after staining.

The pure Ormocer-based resin composite (Organically Modified Ceramic), Admira X-tra, exhibited intermediate values after staining (Table III). This material is composed by a great amount of filler content (84%), which contributes to a lower water sorption, and consequently is less prone to staining, as observed in a previous study [24]. Additionally, Ormocer has shown higher values of microhardness and degree of conversion, probably due to numerous polymerizable units of ormocer molecule. This increases the number of chemical bonds between the methacrylate groups and contributes for maintenance of its superficial integrity after water storage [24, 35, 36].

Although there were different responses in whitening outcomes depending on the material, all resins exhibited some degree of color change when subjected to peroxide-based products. These mouthrinses resulted in whitening effect superior to the color change acceptability threshold (2.7) for AD and BII materials. Thus, such products might be considered as an alternative to maintenance of restoration color over time, mainly in individuals frequently exposed to colored food and beverages, since these colorants tend to stain restorations more intensely than the tooth [37]. Additionally, the whitening mouthrinses might contribute to potentially reducing restoration premature replacement due to esthetic concerns. Nevertheless, caution should be taken to extrapolate these results to clinical situation, since the in vitro nature of the study presume the exacerbation of the staining conditions, as well as the absence of salivary flow and clearance, diluting the mouthrinse. Additionally, the period of whitening treatment with rinses is long and this might affect patient's compliance.

## CONCLUSIONS

The 2% hydrogen peroxide-based mouthrinse (Listerine Whitening) presented the greatest whitening potential on stained composites, mainly on the Giomer-based (Beautifill II) and the ormocer (Admira Fusion) materials. Bromelain/papain solution showed no whitening effect on stained RC.

## Author Contributions

MCM: Formal analysis, Writing - Original Draft Preparation, Writing - Review & Editing. EC: Investigation, Writing - Original Draft Preparation. JBSO: Investigation, Writing - Original Draft Preparation. CRGT: Methodology, Formal analysis, Writing - Original Draft Preparation, Writing - Review & Editing, Supervision, Project administration. TMFC: Methodology, Formal analysis, Writing - Original Draft Preparation, Writing - Review & Editing, Supervision, Project administration. ABB: Conceptualization, Methodology, Formal analysis, Investigation, Resources, Data Curation, Writing - Original Draft Preparation, Writing - Review & Editing, Supervision, Project administration.

## Conflict of Interest

The authors have no proprietary, financial, or other personal interest of any nature or kind in any product, service, and/or company that is presented in this article.

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## Regulatory Statement

The authors declare that this work does not require the approval of the ethics committee.

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**Alessandra Bühler Borges**
**(Corresponding address)**
Universidade Estadual Paulista "Júlio de Mesquita Filho", Institute of Science and Technology, Department of Restorative Dentistry, São José dos Campos, SP, Brazil.
**E-mail**: alessandra.buhler@unesp.br

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