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Effects of black tea tooth staining previously to 35% hydrogen peroxide bleaching

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Aim: To determine if the artificial staining with black tea (BT) influences the enamel microhardness before in-office bleaching and if BT staining is necessary to evaluate the efficacy of bleaching with 35% hydrogen peroxide **Methods:** Enamel/dentin blocks were randomized into groups according to the staining protocol (n=5/group): (CO) control – maintained in artificial saliva solution (AS); (BT_4) immersed in black tea solution for 4 h; (BT_{24}) immersed in black tea solution for 24 h. After the staining protocols, all specimens were kept in AS for one week, followed by bleaching (three sessions of HP application for 40 min). Knoop surface microhardness (kgF/mm^2) was determined at baseline (T_0), after staining (T_1), after 7 days of storage in AS (T_2), and after bleaching (T_3). The color (ΔE_{00}) and coordinate changes (ΔL, Δa, Δb) were measured using a digital spectrophotometer at T_0 and T_3. Data were submitted to one-way (ΔE_{00}, ΔL, Δa, Δb) or two-way ANOVA repeated measures (kgF/mm^2) and Tukey's test (a=5%). **Results:** The staining protocols (BT_4 and BT_{24}) promoted significantly lower microhardness (T_1 and T_2, $p<0.05$) than CO, whereas CO was the only group to maintain microhardness values over time. Bleaching promoted perceptible ΔE_{00} without a significant difference among the groups regardless of the staining protocol ($p=0.122$). CO and BT_4 showed no differences in terms of ΔL and Δa ($p>0.05$), but BT_4 displayed a higher Δb than CO. **Conclusion:** The artificial staining with BT negatively affected the enamel surface microhardness and was not essential to evaluate the efficacy of 35% hydrogen peroxide bleaching.

Keywords: Bleaching agents. Hydrogen peroxide. Staining and labeling.

Introduction

Tooth discoloration is classified based upon its location[1]. Intrinsic color is associated with structural and thickness alterations to dentin during tooth formation or after its eruption[2], while extrinsic discoloration is determined by the adsorption of polyphenolic compounds onto the surface of dental enamel and their interaction with pellicle proteins[3]. Most of these organic chromogens are present in the daily habits of the patients, i.e., intake of foods and smoking[4]. Additionally, enamel defects, dental caries, or restorative materials may facilitate the incorporation of such compounds within the tooth structure[5]. However, it should be noted that the extrinsic staining deposited on the enamel is not resistant to removal with either regular toothbrushing or professional prophylaxis[4,6].

Tooth bleaching remains the most ultraconservative and efficient approach to treat dental discoloration even after prophylaxis[7]. The technique consists of applying carbamide or hydrogen peroxide gels on the enamel surface[8] in concentrations that vary according to the bleaching regimen. The chemical byproducts of the reaction, the reactive oxygen species (ROS) resulting from peroxide decomposition, diffuse through the enamel into the dentin[9], supposedly breaking down the intrinsic staining into smaller molecules. Consequently, teeth look whiter[8].

Several in vitro studies evaluated tooth bleaching protocols' efficacy and adverse effects by using artificial staining on enamel before HP bleaching. One of the reasons for that approach is the necessity of the standardization of tooth color among groups before the bleaching treatments[10,11]. Previous studies reported tooth staining with soft drinks[12], tobacco smoke, coffee[13], red wine[14], and black tea[11]. There is evidence that black tea exhibits a higher staining effect than cola-based soft drinks and coffee[15]. In this regard, Sulieman et al.[3] (2003) validated an artificial staining protocol with black tea specifically for a dental bleaching evaluation. According to the authors, the 24-hour immersion of enamel/dentin specimens in black tea solution did not differ from a 6-day immersion[3]. In other words, an overnight staining protocol would suffice to assess the bleaching efficacy of peroxides.

Nevertheless, staining agents may influence composition and structure other than tooth optical properties. For instance, it has been reported that teeth stained with black tea exhibited lower ROS penetration after highly concentrated HP bleaching than unstained teeth[16]. In addition, long-lasting enamel exposure to solutions or products with a pH lower than 5.5 can trigger enamel demineralization[17]. Since evidence shows that black tea's pH can range from 3.3 to 6.5 in commercial ready-to-drink beverages[18], artificial staining before bleaching may compromise the enamel mineral composition and surface morphology. Moreover, tooth bleaching may modify the enamel composition and structure by decreasing the enamel mineral content[19] and surface microhardness[20], increasing the surface roughness[14], and changing the enamel morphology[11].

Given these facts, artificial black tea staining may interfere with the outcomes and decrease the reliability of the results related to tooth bleaching itself. Although several studies have already investigated the effects of tooth bleaching on enamel sur-

face properties[11,14,19,20], there is no evidence showing if the step of the artificial tooth staining isolated would negatively affect the enamel microhardness and become a confounding variable. Additionally, studies have not indicated whether the specimens' darkening in different exposure times is necessary to determine enamel color change compatible with efficient tooth bleaching. Hence, this study determined the effect of artificial staining with black tea, in short (4 h) or long (24 h) exposures, on enamel surface microhardness and the efficacy of tooth bleaching with 35% hydrogen peroxide (HP) on stained and nonstained teeth. The null hypotheses were that (a) artificial staining would not decrease the enamel surface microhardness and that (b) the staining protocols tested would not affect the color change after bleaching.

Materials and Methods

Experiment Design

Dental blocks were submitted to artificial staining protocols with or without black tea solution (BT):

- CO: Control - without black tea and stored in artificial saliva (AS)
- BT_4: black tea immersion for 4 h (short exposure)
- BT_{24}: black tea immersion for 24 h (long exposure)

After staining, all specimens were stored in AS for 7 d and submitted to an in-office bleaching protocol (35% HP). The color and the surface microhardness of the specimens were assessed before (T_0), after staining (T_1), after storage in AS (T_2), and after bleaching treatment (T_3).

Specimen preparation

Bovine incisors were extracted, cleaned with periodontal scrapers, and stored in 0.1% thymol solution at 4°C for no longer than 30 days. Fifteen teeth without enamel cracks or defects were selected, and the roots were separated from the crowns using a low-speed diamond saw (Isomet, Buehler; Lake Bluff, Illinois, USA) under refrigeration, 2 mm below the cementoenamel junction. Blocks (6 mm × 6 mm and 3 mm thick) were obtained from the central portion of the crown. The dentin was flattened using a rotary polisher (Arotec Ind. Com., São Paulo, SP, Brazil) with silicon carbide paper #600 under water-cooling. The enamel surface was finished with aluminum oxide grit #600 and #1200 and polished using diamond aqueous suspensions (6, 3, 1, and 0.25 µm, Metaldi Supreme, Buehler, Lake Bluff, Illinois, USA). The blocks were ultrasonically cleaned with distilled water for 5 min among each progression in grits.

Artificial Staining and Groups Distribution

The dentin of specimens was isolated using wax to allow the exposure of the enamel surface only. The blocks were randomly assigned to groups according to the black tea protocol (n=5/group): (CO) without BT and stored in AS [1,5 mM Ca; 0,9 mM P; 150 mM KCl e 0,1 M Tris; pH 7,0][21], (BT_4) immersion in BT for 4 h, (BT_{24}) immersion for 24 h.

The sample size was calculated using the software G*Power. The mean and standard deviation values of the enamel surface microhardness reported by Costa et al.[22] (2021) were used to detect the effect size since these authors also used artificially stained enamel to evaluate the action of hydrogen peroxide bleaching. The detected effect size (f = 1.5) was applied on G*Power with a predetermined significance level of 0.05, and a power test of 80%. As a result, five specimens were required for each group.

The black tea solution was prepared according to an adaptation of the protocol described by Sulieman et al.[3] (2003). Two grams of black tea (Dr. Oetker, São Paulo, SP, Brazil) was diluted in 100 mL of boiling distilled water for 5 min, filtered, and cooled. The specimens were immersed in BT (20 mL) at a constant temperature of 37°C. The pH of the solution was checked before and after immersion (4 h: 4.7 – 4.9 and 24 h: 4.6 – 4.9). After staining, the specimens were washed in distilled water and stored in AS for 7 d to allow color stabilization of the specimens.

Bleaching Procedures

In-office bleaching (35% HP, Total Blanc Office H35, Nova DFL, Rio de Janeiro, RJ, Brazil) was performed according to the manufacturer's recommendation: three 40-min sessions at 72-h intervals. The specimens were stored in AS at intervals for seven consecutive days after bleaching.

Surface Microhardness

The mean enamel surface microhardness was obtained by three impressions in the central area of the block, with a Knoop diamond penetrator (Future Tech-FM-1e, Tokyo, Japan), under a static charge of 25 grams for 5 seconds and 100 µm of distance from each other. No differences in kgF/mm^2 among groups were found at baseline (T_0). The microhardness was evaluated immediately after staining (T_1), after stabilization in AS (T_2), and seven days after bleaching (T_3).

Colorimetric Evaluation

The colorimetric measurements on the enamel surface were performed using a digital spectrophotometer (EasyShade, Vita Zahnfabrik, Bad Säckingen, Germany). The equipment was fixed on a platform to allow the tip to face enamel as parallel as possible. The background in which the dental blocks were placed was standardized with an opaque tile. This apparatus was positioned inside a light-controlled chamber. The L*, C*, h*, a*, and b* coordinates were obtained from the average of three measurements. L* represents the luminosity of an object (black to white), while a* and b* represent + red/-green and + yellow/-blue, respectively. The C* parameter determines the chroma of the substrate, and h*, the tooth hue. Because this study aimed to evaluate the effect of bleaching in stained teeth, colorimetric data were collected 7 days after staining and storage in AS (T_2) and 7 days after the last bleaching session (T_3). The color variation was calculated according to CIEDE2000 (ΔE_{00}), with the color difference between T_3 and T_2. The 50:50% perceptibility threshold adopted was 1.2 ΔE_{00} units[23]. The variation in the L*, a*, and b* coordinates were also individually calculated, taking into consideration the same time points mentioned above. The groups exhibited no significant differences in the mean baseline L*, a*, and b* values.

Statistical analysis

The normal distribution (Shapiro–Wilk, p>0.05) and homoscedasticity (Levene, p>0.05) of the data were confirmed. Data were statistically analyzed by one-way (ΔE_{00}, ΔL, Δa, Δb) or two-way repeated-measures ANOVA (microhardness), and Tukey's test detected differences among groups at a significance level of 5%. All analyses were performed by SPSS version 23 (IBM Corp. Released 2015. IBM SPSS Statistics for Windows, Version 23.0. Armonk, NY: IBM Corp.).

Results

Microhardness evaluation. At baseline (T_0), no differences in microhardness were detected among groups (Table 1), but both staining protocols (BT_4 and BT_{24}) decreased enamel microhardness (T_1) in comparison with CO (p<0.001). At this time-point (T_1), BT_{24} exhibited lower microhardness than BT_4 (p<0.001). After storage in AS for 7 d (T_2), the results remained unaltered.

Seven days after bleaching (T_3), CO and BT_4 displayed no differences (p=0.36) but exhibited higher Knoop values than BT_{24} (p<0.05). CO microhardness remained unaltered over time, while microhardness recovery was detected for BT_{24} at T_3.

Table 1. Mean values and standard deviation of microhardness values (kgF/mm²).

Groups	T_0	T_1	T_2	T_3
CO	307.1 (20.5) ^{Aa}	288.5 (14.5) ^{Aa}	286.5 (6.2) ^{Aa}	285.89 (10.74) ^{Aa}
BT_4	307.0 (16.3) ^{Aa}	260.1 (16.9) ^{Bb}	259.5 (17.7) ^{Bb}	267.82 (25.35) ^{Ab}
BT_{24}	310.3 (16.2) ^{Aa}	189.0 (24.4) ^{Cc}	204.6 (25.0) ^{Cc}	224.60 (11.97) ^{Bb}

Means followed by distinct letters are different according to two-way ANOVA repeated measurements and Tukey's test at a significance level of 5%. Capital letters compare staining protocols (columns), and lowercase letters compare time points (lines). T_0: baseline; T_1: after staining; T_2: after 7 days of storage in AS; T_3: after bleaching. CO: control; BT_4: black tea immersion for 4 h; BT_{24}: black tea immersion for 24 h.

Color evaluation. No differences in ΔE_{00} among groups were detected (p=0.122) after bleaching (T_3 -T_2). The mean color differences promoted by bleaching were above the perception threshold (ΔE_{00}>0.8), regardless of the staining protocol (Table 2). BT_{24} showed higher ΔL, Δa, and Δb values than CO but no difference from BT_4 in ΔL and Δb. BT_4 and CO displayed no differences in ΔL and Δa (>0.05) but higher Δb than CO.

Table 2. Mean values and standard deviation of the alteration in Euclidian coordinates (ΔL, Δa, and Δb) and color alteration between color stabilization in AS and bleaching.

Groups	ΔL	Δa	Δb	ΔE_{00}
CO	-2.8 (3.7) ^{B}	-2.3 (1.4) ^{A}	-15.1 (1.8) ^{B}	9.0 (1.1) ^{A}
BT_4	-1.0 (6.1) ^{AB}	-4.2 (1.6) ^{A}	-18.8 (2.1) ^{A}	10.7 (0.2) ^{A}
BT_{24}	6.4 (5.9) ^{A}	-8.6 (1.4) ^{B}	-17.2 (1.0) ^{A}	11.5 (2.9) ^{A}

Means followed by distinct letters are different according to one-way ANOVA and Tukey's test at a significance level of 5%. Capital letters compare staining protocols (columns). CO: control; BT_4: black tea immersion for 4 h; BT_{24}: black tea immersion for 24 h.

Discussion

Black tea staining reduced the enamel microhardness before the bleaching procedures, regardless of immersion time in BT. This outcome remained unchanged even after storage in artificial saliva for 7 days elapsed from the last bleaching session. Therefore, the first null hypothesis was rejected because artificial staining with BT affected the enamel surface microhardness.

Artificial dental staining is a method developed to mimic the natural intrinsic staining that occurs with the deposition of secondary and tertiary dentin along the tooth´s lifetime[3]. The protocol described in 2003[3] has been extensively used by researchers as an attempt to simulate an ideal substrate to be bleached and as an approach to standardize the teeth color of groups[24-26]. Nevertheless, that study[3] failed to report the pH of black tea and its effects on enamel after 24 h of immersion.

To the best of our knowledge, studies investigating the bleaching effect on enamel have not reported the surface microhardness immediately after immersion in the staining solution. For instance, a previous report displayed the microhardness values at baseline and after bleaching while evaluating the effects of light-activated 6% HP on the enamel surface[22,27]. In that study, black tea with an unknown pH and manufacturer was used to immerse dental blocks for 18 h. Since a mean surface microhardness reduction was observed over time, the black tea immersion should also be considered in the interpretation of the results. Bearing this in mind, the time points of the present study allowed a more comprehensive understanding of the real impact of bleaching on the enamel surface. The results showed that 35% HP was not responsible for reducing the enamel surface microhardness, contrary to the black tea solution.

Similar to the tea used in our study, with a pH ranging from 4.6 to 4.9, others have reported the use of BT with a pH lower than 5.5[28-30]. The study of Farawati et al.[30] (2019) investigated the effect of black tea, wine, soda, coffee, and water on the surface properties of enamel submitted to bleaching with carbamide peroxide. The data from that study suggested that only black tea and wine changed the enamel mineral composition. One might say that the presence of fluoride in tea[31] could uphold the level of enamel mineralization; nonetheless, it was demonstrated that the immersion of eroded enamel in a ready-to-drink black tea with 0.760 mg/L of fluoride, in a much lower immersion time than our study, decreases the enamel microhardness significantly more compared to other types of drinks[32]. Additionally, Jameel et al.[29] (2016) showed that a black tea solution (pH 4.9, 0.938 mg/L) negatively impacted not only the surface microhardness but also the enamel roughness. Hence, the existing evidence suggests that the pH of the solutions might play a more important role than the presence of fluoride in the maintenance of the surface properties.

It is important to highlight that group BT_4 was set as an alternative to diminishing the necessary time of staining. Despite the lower immersion time, a significant decrease in KHN was also observed for this group. However, it is noteworthy that this reduction in the microhardness was significantly lower than that of BT_{24}, which may indicate a time-dependent effect of black tea on the enamel microhardness. Even though other protocols could adjust the pH of the staining solution as an attempt to overcome

the low-pH value, raising the pH of this solution could impact the capability of the staining molecules to penetrate the dental structure. Since no studies were found attempting this approach, future studies could investigate its application.

The colorimetric data inferred that the staining protocols did not affect the color change promoted by bleaching. The group stored in artificial saliva showed a similar ΔE_{00} to the BT groups. Thus, the second null hypothesis was accepted since the staining protocols did not affect the color change after bleaching.

According to Paravina et al.[23] (2019), a color change above 5.4 units is compatible with the excellent effectiveness of tooth bleaching, thereby indicating a very highly perceptible outcome. Therefore, the commercial bleaching gel used herein was very effective even without a previous staining step [ΔE_{00} = 9.0 (1.1)]. Additionally, *in vitro* studies using artificial staining may exhibit exacerbated ΔE_{00} results. For instance, different studies evaluating the same bleaching protocols presented higher ΔE_{00} values when teeth were artificially stained[11,13] compared to nonstained teeth[13,33,34].

Differences in the L* and a* coordinate changes among the groups could be explained by the absence (CO) or lower amount (BT_4) of pigments, suggesting a minor challenge for the hydrogen peroxide gel. On the other hand, the high shift in the b* coordinate from b+ (yellow) to b- (blue), independent of the staining protocol, might suggest that the bleaching gel could breakdown the chromogens even without previous artificial staining. These results indicate that artificial staining before bleaching to mimic and standardize the specimens for color evaluation may be discarded since bleaching therapy was effective in nonstained teeth. In this regard, an interesting research approach would be avoiding dental blocks with a high initial luminosity (L*) and low values of the b* coordinate to observe the bleaching effects.

Updated colorimetric systems, such as the color change calculated by CIEDE2000[35,] could overcome the drawbacks of previous calculations (CIELAB or visual shade guides). Moreover, ΔE_{00} adjusts the low influence that the a* coordinate plays on the color of teeth. The Sulieman et al.[3] (2003) study only applied a subjective color evaluation of the specimens while establishing the staining protocol. This fact emphasizes the need for further analysis using appropriate objective color measurements and corresponding updated color and whiteness indexes[7].

A limitation of the present research is the absence of topography analysis and groups with additional BT immersion. However, the purpose of this investigation was to stress the importance of carefully employing artificial staining for bleaching evaluation. Artificial staining should be carefully designed since overexposure and pH might damage the enamel surface and lead to a mistaken interpretation of the HP bleaching effects and enamel mineral content. In addition, excluding artificial staining may not impact bleaching efficacy, and the results could be more reliable and translated to a clinical setting.

In conclusion, artificial staining with black tea showed detrimental effects on enamel surface microhardness, wherein the higher the time of exposure to tea was, the lower the hardness. In addition, bleaching with 35% HP did not decrease surface microhardness in the stained or nonstained specimens, and the staining protocols (4 h or

24 h immersion in black tea) were not essential to detect the bleaching effects. All groups displayed similar color changes after bleaching.

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Conflict of Interest

The authors have no conflicts of interest to declare.

Data Availability

Datasets related to this article will be available upon request to the corresponding author.

Author Contribution

- Samuel da Silva Palandi: methodology, investigation, data curation, writing (original draft, review, and editing);
- Matheus Kury: conceptualization, methodology, formal analysis, data curation, writing (review and editing);
- Mayara Zaghi Dal Picolo: methodology, Investigation, data curation, writing (review and editing);
- Fernando Luis Esteban Florez: conceptualization, funding acquisition, writing (review and editing);
- Vanessa Cavalli: conceptualization, resources, supervision, funding acquisition, writing (review and editing).

References

1. Addy M, Moran J, Newcombe R, Warren P. The comparative tea staining potential of phenolic, chlorhexidine and anti-adhesive mouthrinses. J Clin Periodontol. 1995 Dec;22(12):923-8. doi: 10.1111/j.1600-051x.1995.tb01796. x.
2. Rodríguez-Martínez J, Valiente M, Sánchez-Martín MJ. Tooth whitening: From the established treatments to novel approaches to prevent side effects. J Esthet Restor Dent. 2019;31(5):431-40. doi: 10.1111/jerd.12519.
3. Sulieman M, Addy M, Rees JS. Development and evaluation of a method in vitro to study the effectiveness of tooth bleaching. J Dent. 2003 Aug;31(6):415-22. doi: 10.1016/s0300-5712(03)00069-1.
4. Prathap S, Rajesh H, Boloor VA, Rao AS. Extrinsic stains and management: a new insight. J Acad Indus Res. 2013;1(8):435-42.
5. Żyła T, Kawala B, Antoszewska-Smith J, Kawala M. Black stain and dental caries: a review of the literature. Biomed Res Int. 2015; 2015:469392. doi: 10.1155/2015/469392.

6. de Geus JL, de Lara MB, Hanzen TA, Fernández E, Loguercio AD, Kossatz S, et al. One-year follow-up of at-home bleaching in smokers before and after dental prophylaxis. J Dent. 2015 Nov;43(11):1346-51. doi: 10.1016/j.jdent.2015.08.009.

7. Joiner A, Luo W. Tooth color and whiteness: a review. J Dent. 2017 Dec;67S:S3-S10. doi: 10.1016/j.jdent.2017.09.006.

8. Kwon SR, Wertz PW. Review of the mechanism of tooth whitening. J Esthet Restor Dent. 2015 Sep-Oct;27(5):240-57. doi: 10.1111/jerd.12152.

9. Llena C, Martínez-Galdón O, Forner L, Gimeno-Mallench L, Rodríguez-Lozano FJ, Gambini J. Hydrogen peroxide diffusion through enamel and dentin. Materials 2018;11(9):1694. doi: 10.3390/ma11091694.

10. Gallinari MO, Fagundes TC, da Silva LM, de Almeida Souza MB, Barboza A, Briso A. A new approach for dental bleaching using violet light with or without the use of whitening gel: study of bleaching effectiveness. Oper Dent. 2019 Sep-Oct;44(5):521-9. doi: 10.2341/17-257-L.

11. Kury M, Perches C, da Silva DP, André CB, Tabchoury CPM, Giannini M, et al. Color change, diffusion of hydrogen peroxide, and enamel morphology after in-office bleaching with violet light or nonthermal atmospheric plasma: an in vitro study. J Esthet Restor Dent. 2020 Jan;32(1):102-12. doi: 10.1111/jerd.12556.

12. Prskalo K, Klarić Sever E, Alerić I, Antonić Jelić T, Žaja I. Risk factors associated with black tooth stain. Acta Clin Croat. 2017 Mar;56(1):28-35. doi: 10.20471/acc.2017.56.01.05.

13. Kobayashi RS, Picolo MZD, Kury M, Resende BA, Esteban Florez FL, Cavalli V. Effects of dental bleaching protocols with violet radiation on the color and chemical composition of stained bovine enamel. Photodiagnosis Photodyn Ther. 2021 Jun;34:102194. doi: 10.1016/j.pdpdt.2021.102194.

14. Berger SB, Coelho AS, Oliveira VA, Cavalli V, Giannini M. Enamel susceptibility to red wine staining after 35% hydrogen peroxide bleaching. J Appl Oral Sci. 2008 May-Jun;16(3):201-4. doi: 10.1590/s1678-77572008000300007.

15. Téo TB, Takahashi MK, Gonzaga CC, Lopes MGK. Avaliação, após clareamento, da alteração de cor de dentes bovinos imersos em soluções com elevado potencial de pigmentação. Rev Sul-Bras Odontol. 2010 Oct-Dec;7(4):401-5.

16. de Oliveira Duque CC, Soares DG, Briso A, Ortecho-Zuta U, de Oliveira Ribeiro RA, Hebling J, et al. Influence of tooth pigmentation on H_2O_2 diffusion and its cytotoxicity after in-office tooth bleaching. Oper Dent. 2020 Nov;45(6):632-42. doi: 10.2341/19-013-L.

17. Cury JA. [Fluoride use and caries control as a disease]. In: Baratieri LN, et al. [Restorative dentistry: fundamentals and possibilities]. São Paulo: Santos; 2001. p.31-68. Portuguese.

18. Lun Su Y, Leung LK, Huang Y, Chen Z-Y. Stability of tea theaflavins and catechins. Food Chem. 2003;83(2):189-95. doi:10.1016/s0308-8146(03)00062-1.

19. Cavalli V, Rosa DAD, Silva DPD, Kury M, Liporoni PCS, Soares LES, et al. Effects of experimental bleaching agents on the mineral content of sound and demineralized enamels. J Appl Oral Sci. 2018 Oct; 26: e20170589. doi: 10.1590/1678-7757-2017-0589.

20. Zantner C, Beheim-Schwarzbach N, Neumann K, Kielbassa AM. Surface microhardness of enamel after different home bleaching procedures. Dent Mater. 2007 Feb;23(2):243-50. doi: 10.1016/j.dental.2006.06.044.

21. Queiroz CS, Hara AT, Paes Leme AF, Cury JA. pH-cycling models to evaluate the effect of low fluoride dentifrice on enamel de and remineralization. Braz Dent J 2008;19(1):21-7 doi: 10.1590/S0103-64402008000100004.

22. Costa JLSG, Besegato JF, Kuga MC. Bleaching and microstructural effects of low concentration hydrogen peroxide photoactivated with LED/laser system on bovine enamel. Photodiagnosis Photodyn Ther. 2021 Sep;35:102352. doi: 10.1016/j.pdpdt.2021.102352.

23. Paravina RD, Pérez MM, Ghinea R. Acceptability, and perceptibility thresholds in dentistry: A comprehensive review of clinical and research applications. J Esthet Restor Dent. 2019 Mar;31(2):103-12. doi: 10.1111/jerd.12465 .

24. Thacker M, Chen YN, Lin CP, Lin FH. Nitrogen-doped titanium dioxide mixed with calcium peroxide and methylcellulose for dental bleaching under visible light activation. Int J Mol Sci. 2021 Apr;22(7):3759. doi: 10.3390/ijms22073759.

25. Ferraz NKL, Nogueira LC, Neiva IM, Ferreira RC, Moreira AN, Magalhães CS. Longevity, effectiveness, safety, and impact on quality of life of low-concentration hydrogen peroxides in-office bleaching: a randomized clinical trial. Clin Oral Investig. 2019 May;23(5):2061-70. doi: 10.1007/s00784-018-2607-7.

26. Kyaw KY, Otsuki M, Segarra MS, Tagami J. Effect of sodium fluoride pretreatment on the efficacy of an in-office bleaching agent: an in vitro study. Clin Exp Dent Res. 2018 Jun 29;4(4):113-8. doi: 10.1002/cre2.113.

27. Costa JLSG, Besegato JF, Zaniboni JF, Kuga MC. LED/laser photoactivation enhances the whitening efficacy of low concentration hydrogen peroxide without microstructural enamel changes. Photodiagnosis Photodyn Ther. 2021 Dec;36:102511. doi: 10.1016/j.pdpdt.2021.102511.

28. Simpson A, Shaw L, Smith AJ. Tooth surface pH during drinking of black tea. Br Dent J. 2001 Apr;190(7):374-6. doi: 10.1038/sj.bdj.4800977.

29. Jameel RA, Khan SS, Abdul Rahim ZH, Bakri MM, Siddiqui S. Analysis of dental erosion induced by different beverages and validity of equipment for identifying early dental erosion, in vitro study. J Pak Med Assoc. 2016 Jul;66(7):843-8.

30. Farawati FAL, Hsu SM, O'Neill E, Neal D, Clark A, Esquivel-Upshaw J. Effect of carbamide peroxide bleaching on enamel characteristics and susceptibility to further discoloration. J Prosthet Dent. 2019 Feb;121(2):340-6. doi: 10.1016/j.prosdent.2018.03.006.

31. Hayacibara MF, Queiroz CS, Tabchoury CPM, Cury JA. Fluoride and aluminum in teas and tea-based beverages. Rev Saude Publica. 2004 Feb;38(1):100-5. doi: 10.1590/s0034-89102004000100014.

32. Amoras DR, Corona SAM, Rodrigues Jr AL, Serra MC. Effect of beverages on bovine dental enamel subjected to erosive challenge with hydrochloric acid. Braz Dent J. 2012;23(4):367-72. doi: 10.1590/S0103-64402012000400010.

33. Kury M, Wada EE, Silva DPD, Tabchoury CPM, Giannini M, Cavalli V. Effect of violet LED light on in-office bleaching protocols: a randomized controlled clinical trial. J Appl Oral Sci. 2020;28:e20190720. doi: 10.1590/1678-7757-2019-0720.

34. Kury M, Wada EE, da Silva Palandi S, Picolo MZD, Giannini M, Cavalli V. Colorimetric evaluation after in-office tooth bleaching with a violet LED: 6- and 12-month follow-ups of a randomized clinical trial. Clin Oral Investig. 2022 Jan;26(1):837-47 16. doi: 10.1007/s00784-021-04062-9.

35. Sharma G, Wu W, Dalal EN. The CIEDE2000 color-difference formula: Implementation notes, supplementary test data, and mathematical observations. Color Res Appl. 2005 Feb;30(1):21-30. doi: 10.1002/col.20070.