Dental Sensitivity and Color Change in Patients Undergoing Dental Bleaching with Application of Violet Light

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ABSTRACT

Objectives: Evaluate the effects of Violet Light Illumination (VLI) alone or combined with 10% carbamide peroxide and 35% hydrogen peroxide gels on Dental Sensitivity (DS) and tooth-color difference.

Materials and methods: 15 patients were divided into V group (n=5; dental bleaching using only VLI), VCP group (n=5; VLI and 10% carbamide peroxide gel) and VHP group (n=5; dental bleaching using VLI and 35% hydrogen peroxide gel). Tooth color was assessed by using ΔE and Δ blue of a calibrated colorimeter. Colorimetric measurements and DS scores were recorded before the initial session (T0), after 1st session (T1) and after 2nd session (T2). At each session, DS scores were taken in a visual scale from 0 to 10 and their sum for all teeth evaluated in a given patient was used as that patient's DS score. Kruskal-Wallis and two-way ANOVA tests were used to compare ΔE between groups. Intrapulpal heating of experimental conditions was assessed *in vitro*.

Results: We observed 100% DS reduction for the VCP and VHP groups, whereas the V group exhibited a reduction of >95.6% after T1 and >96.7% after T2. Intrapulpal heating of 2°C for VLI conditions of this study. ΔE was higher for the V group, which indicates this group had a more pronounced whitening effect, followed by the VCP and then by the VHP groups. ΔE and Δ blue exhibited the same trend as the blue readings for each group. No statistically significant differences were found for ΔE between groups.

Conclusion: Application of VLI with or without peroxide gels had a positive effect in obtaining improved in-office dental bleaching. Violet light promoted "desensitization" by decreasing DS over dental bleaching sessions.

Clinical relevance: The effect of light application alone has been scarcely investigated in dental bleaching. Our study provides a detailed evaluation of DS and tooth-color difference as well as possible sources of observed effects according to intrapulpal temperatures and light propagation in tooth tissue.

Key Words: Dental bleaching, Violet light, In-office bleaching, Light-activation, Tooth color, Tooth sensitivity, Colorimetry, Dental sensitivity, Color difference, Whitening.

Introduction

Dental bleaching is a technique used to improve the teeth aesthetical appearance. This aesthetic procedure gives the patient a natural white-teeth smile and keeps their original anatomic shape. Dental bleaching delivers a youthful smile to adult patients without the need of dental surface abrasion and later prosthetic intervention with restorative materials. Thus, dental bleaching may be considered a conservative and minimally invasive treatment in aesthetic odontology.

Dental bleaching became popular after 1989, when Haywood and Heymann published a pioneer study about a homemade application of peroxide carbamide with this purpose [1]. After that, many other researches to investigate the effects, improve techniques and optimize the procedure were done. Currently, a source of light is used to accelerate the dental bleaching process [2]. New studies regarding dental bleaching have aimed at decreasing the unwanted effects caused by peroxides [3,4].

There is much controversy in literature regarding the need of light application in dental bleaching, as combining peroxides and illumination achieved similar results compared to peroxides alone [5,6]. However, the effect of light application alone was scarcely investigated. For a long time, blue light was combined with peroxides when performing in-office dental bleaching. Application of violet light in dental bleaching emerged as a new source of visible light capable to optimize assisted dental bleaching [7]. *In vitro* studies show that violet light was efficient in promoting dental bleaching in bovine teeth even without using peroxide [7]. Other laboratorial studies show the effect of laser light and LED with violet wavelength promoting the break-up of stains in a solution pigmented with rhodamine and pastilles of hydroxyapatite [8,9].

Dental sensitivity (tooth pain) is frequently reported as the main reason why the patient does not undergo this treatment [10,11]. The sensitivity that occurs during dental bleaching has led to patients giving up on the treatment. Thus, the possibility of using violet light with low concentration of bleaching chemical agents or not in patients that present dental hypersensitivity may be an alternative to solve this unwanted effect [12,13]. The use of violet light in dental bleaching may bring a larger number of potential new patients to the dental office.

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Recently published studies show that the application of fractioned violet light may optimize dental bleaching due to its physical effect of breaking up pigments that stain the tooth without causing dental sensitivity [14]. Furthermore, a hypothetical effect of violet light is the repolarization of nervous terminals inside the dentinal tubules, promoting dental "desensitization" during and after dental bleaching. This study investigated the effects of violet light illumination alone or combined with carbamide peroxide and hydrogen peroxide gels on dental sensitivity and tooth-color difference. Our investigation involved dental sensitivity evaluation through an exploratory probe and air jet with a triple syringe for patients of each experimental group (i.e., groups receiving violet-light illumination alone, violet light illumination combined with 10% carbamide peroxide gel, or violet light illumination combined with 35% hydrogen peroxide). Assessment of possible temperature contributions to dental sensitivity was performed via an in vitro experiment of measurement of intrapulpal temperature following the similar protocols used in this study. This in vitro experiment also motivated the use of violet-light illumination instead of blue light illumination. Following violet-light illumination, we also described the tooth-color difference of each tooth type (central incisors, lateral incisors, canines and first premolars) for each of experimental group involved in this study.

Methodology

This randomized clinical study, transversal and single-center was performed in a dental office at the Biophotonics Laboratory, Institute of Physics of Sao Carlos-University of Sao Paulo (USP). All procedures were approved by the Ethics Committee of the Federal University of São Carlos, Sao Carlos, Brazil (approval no: 1.037.128-12/05/2015). All participants handed in a written consent and agreed to take part in the study. This study is registered in the Brazilian Clinical Trials Registry/Registro Brasileiro de Ensaios Clínicos (Re-BEC) under the clinical trial registration number 10859 with the identifiers:

• Órgão emissor do CAAE: Plataforma Brasil (38288114.1.0000.5504)

• WHO International Clinical Trials Registry platform: UTN code U1111-1262-5077

• Approval number of the clinical research ethics committee from Universidade Federal de São Carlos (UFSCAR): 1.037.128

Inclusion and Exclusion of Patients

All the 20 individuals evaluated for eligibility for the clinical

trial met the inclusion criteria. All individuals included in the research had complete dentition, i.e., presence of all anterior teeth plus the permanent superior and inferior pre-molars and first molars. All included volunteers agreed to free-willingly sign a consent form to be part of this clinical experiment. Our study excluded individuals with uncontrolled systemic diseases, pregnant women and smokers. It also excluded volunteers who reported previous reaction to peroxides and presence of staining caused by tetracycline or fluorosis. Volunteers were oriented to avoid the following foods and beverages in the 48 hours following the end of the dental bleaching treatment: black tea, coffee, red wine, sodas, dark green vegetables, and tomato sauces, sauces containing dyes, grape juices, and beetroot. During the intervention phase, 5 subjects dropped out of the study for reasons unrelated to the dental bleaching procedure (e.g., volunteer moved to another city). The steps for volunteer selection, applied interventions and analysis performed are shown in Figure 1.

Light Source

The device used was the Bright Max Whitening (BMW-MMOptics, São Carlos, SP) wavelength of approximately 405 nm, power of 350 mW per LED, containing 4 units of LEDs on the active tip, tip irradiation area of 63 mm \times 15 mm=945 mm2, total estimated power of 1400 mW and irradiance in contact with surface of 165 mW/cm². This device is registered at ANVISA/MS under n° 80051420019.

Bleaching Gel

The study used carbamide peroxide 10% bleaching gel (CP10-Whiteness Perfect-FGM, Joinville, SC, Brazil) registered at ANVISA under n° 80172310020 and hydrogen peroxide 35% bleaching gel (HP35-Whiteness HP Blue Calcium-FGM, Joinville, SC, Brazil) registered at ANVISA under n° 80172310040.

Experimental Groups and Color Measurement

15 patients from the population of São Carlos were randomly selected to receive dental bleaching. A computer program was used for randomization. Volunteers received prophylaxis with a rotating brush and prophylactic paste and after that the color of superior and inferior teeth (from central incisor to first pre-molar) using a colorimeter (Pocket Spec®-PocketSpec Technologies Inc-Denver, CO-USA) to digitally measure tooth color using a coloration RGB-red/green/blue scale in *Figure 2*. The initial color was standardized by using a standard color calibration target indicated by the manufacturer of the colorimeter.



Figure 1: Flowchart of the steps taken throughout this study (CONSORT flow diagram). The flowchart highlights the enrollment of volunteers, their allocation, interventions applied, and analysis during the study. No follow-up was performed in this study. V: Bleaching group with violet light; VCP: Bleaching group with Violet light and carbamide peroxide (10%); VHP: Bleaching group with violet light and hydrogen peroxide (35%).



Figure 2: Showing digital measurement by using a colorimeter (Pocket Spec®-PocketSpec Technologies Inc-Denver, CO-USA).

After measurement of initial tooth color the 15 patients received dental bleaching and were divided into V group (dental bleaching using only violet light), VCP group (dental bleaching using violet light and 10% Carbamide Peroxide (CP) gel) and VHP group (dental bleaching using violet light and 35% Hydrogen Peroxide gel (HP)). Two dental bleaching sessions were carried out, in which the color of superior and inferior teeth was assessed before and after each session. This assessment was performed by using color scale Vitapan Classical (Vita Zahnfabrik, H. Rauter GmbH and Co. KG.D-7880 Säckingen, Germany) and a colorimeter. Details of experimental groups and protocol can be found in *Table 1*.

Sensitivity Evaluation

All patients were submitted to dental sensitivity evaluation through an exploratory probe and jet air through a triple syringe and evaluated by means of visual analysis (VAS) of pain on a scale from 0 to 10. The volunteer scored 0 when they had no pain and the other numbers to quantify pain until maximum score in pain of 10.

Air jet was applied on the vestibular surface of all superior and inferior bleached teeth, except for the molars that did not receive treatment. In addition, dental sensitivity was assessed with exploratory probe horizontally back and forth three times in the buccal cervical of the bleached teeth. Dental sensitivity (pain) scores ranged from 0 to 10 for each tooth. The sum of these scores for all teeth (n=16 per patient) corresponded to the dental sensitivity score of a given patient at each session, i.e., at times T0 (initial), T1 (after 1st session) and T2 (after 2nd session) and compared to evaluate the effects of sensitivity during and after dental bleaching applying violet light with and without gel. Sensitivity tests were performed before and after each session as shown in *Figure 3*.

Group	volunteers	Light	Gel	Time violet light
V	5	LED Violet 408 nm	No gel	30 minutes (20 c, 30 sec A, 1 min P)
VCP	5	LED Violet 408 nm	Carbamide Peroxide 10%	30 minutes (20 c, 30 sec A, 1 min P)
VHP	5	LED Violet 408 nm	Hydrogen Peroxide 35%	30 minutes (20 c, 30 sec A, 1 min P)

Table 1: Protocol details of experimental groups involved in this study. C=cycles, A=active, P=pause.



Figure 3: Methods of evaluation of dental sensitivity with probe and air. The figure in right side illustrates the procedure for pain evaluation upon touching of an exploratory probe, whereas the figure in the left side shows the procedure for jet air through a triple syringe.

In Vitro Assessment of Intrapulpal Heating

When light-activated tooth whitening procedures are performed, there is significant concern about the heat generated by the light source, which can cause pulp irritation or serious damage. The aim of this in vitro study is to quantify the intrapulpal heating during tooth whitening using LED violet (410 nm) and blue LED (450 nm). Three premolars extracted for orthodontic treatment purposes were selected, were sectioned their crowns, and inserted a digital thermometer (Termopar ET-2082C; Minipa, São Paulo, SP, Brazil) into the pulp chamber. Then, the teeth were irradiated with violet LED devices (200 mW/cm²) (Bright Max Whitening-MMOptics, São Carlos, SP, Brazil) for 30 minutes fractionated (20 cycles of violet light illumination of 1 min and 30 seconds pauses/ standby) and Blue LED (80 mW/cm²) (Bright Max-MMOptics, São Carlos, SP Brazil) and 35% hydrogen peroxide gel for 10 continuous minutes. The temperature was monitored for the entire illumination interval.

Colorimetric Analysis

Once RGB (red, green, blue) data was collected, we used MATLAB (MathWorks Inc., Natick, Massachussets) routines to convert the RGB readings to CIE L*a*b* color coordinates, established by the International Commission of I'Eclairage (CIE; International Commission on Lighting). L*a*b* color space consists of a lightness coordinate (L) ranging from 0 to 100 (black to white), as well as the unbounded (no specific range of values) color dimensions a* (varying from green to red) and b* (varying from blue to yellow). L*a*b* values were obtained by considering the reference white point as the CIE standard illuminant D65 [0.9504, 1.0000, 1.0888], which simulates noon daylight with correlated color temperature of 6504 K. Since the L*a*b* is a device independent color space, the L*a*b* coordinates are more reliable when comparing studies using different devices.

We also calculated the color differences in dental bleaching between the situations pre-treatment (start/initial; T0), after the first whitening session (1st session; T1) and post-treatment (after the second whitening session, 2nd session; T2) by using:

$$\Delta color = color^{f} - color^{t}$$

Where colort represents the initial color coordinates and color orf the final color coordinate. In this study we computed the color difference for the blue coordinate of the RGB readings (Δ blue or Δ B) compared to the reading before whitening treatment, according to:

$$\Delta blue = \Delta B = B^f - B^i$$

Where B^{f} is the blue reading after the 1^{st} or the 2^{nd} sessions

 $(B^{1st \text{ session}} \text{ or } B^{2nd \text{ session}})$ and Bt is the blue reading pre-treatment. The $L^*a^*b^*$ coordinates can also be used to calculate the total color change (or color difference) given by:

$$\Delta E = \sqrt{(\Delta L)^2 + (\Delta a^*)^2 + (\Delta b^*)^2}$$

Statistical Analysis

Statistically significant differences between color measurements of the V, VCP and VHP group were assessed by the Kruskal-Wallis test and two-way ANOVA test. Significance level was set at 5% (p<0.05). All pairwise comparison results were calculated by using the Tukey-Kramer Method (Tukey's test) as post-hoc test. We used the MATLAB (MathWorks Inc., Natick, Massachussets) software for all statistical analysis performed in this study.

Results

In this study, dental sensitivity was verified by applying an air jet on the vestibular surface of all superior and inferior bleached teeth as well as performing tests with exploratory probe horizontally back and forth three times in the buccal cervical of the bleached teeth. *Table 2* shows the sum of dental sensitivity scores (varying from 0 to 10 for each tooth) of all the 80 teeth of each group (16 teeth from central incisors to premolars of 5 patients) at T0 (initial), T1 (after 1st session) and T2 (after 2nd session) [15]. The dental sensitivity is illustrated separately each experimental group undergoing dental bleaching/whitening treatment. These results are shown in *Table 2*.

Our results suggest that volunteers in the V group presented much higher initial dental sensitivity compared to the VCP and VHP groups. In all groups, we observed that this sensitivity reduced after dental bleaching. Even though we cannot precisely confirm how much reduction was promoted by the violet light illumination combined with carbamide and hydrogen peroxide gels, we can clearly see a reduction of 100% of the dental sensitivity assessed with the exploratory probe in all groups. The reduction was also 100% for the air-jet pain evaluation of VCP and VHP groups, whereas the V group exhibited a reduction of 95.6% after the 1st session (T1) and 96.7% after the 2nd session (T2).

In addition to the pain/dental sensitivity reduction, the violet light illumination alone or combined with peroxide gels increased parameters associated with tooth whitening. *Table 3* quantifies the whitening effect by showing color differences in ΔE and Δ blue, as well as the relationship with the amplitude of the blue readings. No statistically significant differences were found between V, VCP and VHP groups for significance level of 5% (p<0.05).

Table 2: Dental sensitivity before clinical sessions (T0), after first session (T1) and after second session (T2) of dental bleaching. V:Violet light illumination group, VCP: Violet light illumination+carbamide peroxide gel 10% group, VHP: Violet light illumination+hy-
drogen peroxide gel 35%.

		-	
	V	VCP	VHP
Т0			
Probe	14	0	0
Air	90	2	0
T1			
Probe	0	0	0
Air	4	0	0
T2			
Probe	0	0	0
Air	3	0	0

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Table 3: Whitening effects of dental bleaching protocols for each experimental group. Blue: Blue reading of the RGB colorimeter. Δ blue:Difference in the blue reading compared to the start (before intervention). Δ E: total color change compared to the start. V: Violet lightillumination group, VCP: Violet light illumination+carbamide peroxide gel 10% group, VHP: Violet light illumination+hydrogen per-
oxide gel 35%.

	Blue			Δb	lue	ΔΕ	
	Start (T0)	1 st session (T1)	2 nd session (T2)	1 st session (T1)	2 nd session (T2)	1 st session (T1)	2 nd session (T2)
V	87.2 ± 9.9	97.8 ± 10.6	99.0 ± 6.8	10.5 ± 5.6	11.7 ± 4.3	5.6 ± 4.3	7.3 ± 4.2
VCP	82.3 ± 10.8	90.8 ± 7.9	91.8 ± 7.4	8.5 ± 3.2	9.4 ± 13.2	5.6 ± 4.3	6.6 ± 4.0
VHP	63.8 ± 8.6	68.5 ± 9.5	70.5 ± 8.6	4.7 ± 3.6	6.7 ± 4.1	4.5 ± 3.7	4.6 ± 3.1

The color difference ΔE was higher for the V group, which indicates this group had a more pronounced whitening effect, followed by the VCP and then by the VHP groups. Although the whitening effect was lower when using peroxide gels, it is important to note that ΔE and Δ blue exhibit the same trend as the blue readings of the RGB colorimeter. This result suggests that the whitening effect may be correlated to the initial blue readings.

In order to evaluate whether device-independent color difference was tooth dependent (due to the position of the teeth with respect to the violet light illumination device), we calculated ΔE for central incisors, lateral incisors, canines and first premolars of each group after the 1st session (T1) and 2nd session (T2), as shown in *Table 4*.

For better visualization of results of *Table 4*, we plotted several graphs emphasizing the ΔE differences between experimental groups (V, VCP and VHP), tooth types (central incisors, lateral incisors, canines and first premolars) and whitening/dental bleaching sessions (1st or 2nd sessions). *Figure 4* highlights the ΔE differences among tooth types, which are grouped by dental bleaching session for each experimental group. Our results suggest that canines and premolars had a more pronounced whitening effect (larger ΔE) compared to central and lateral incisors for the V group. A similar trend was observed for the VCP and VHP groups, except by the 2nd session of VCP and canines in the 1st session of the VHP group. In average, an increase in ΔE can be seen between the 1st and 2nd sessions of the V and VCP groups, whereas that of the VHP group is not evident.

Figure 5 highlights the ΔE differences after the 1st or 2nd dental bleaching sessions of each experimental group. Results are displayed separately for each tooth type. ΔE increase between 1st and 2nd sessions was more pronounced for the V and VCP groups among all tooth types, except by the canines of the VHP group and the first premolars of the VHP group.

Figure 6 highlights the ΔE differences after the 1st or 2nd dental bleaching sessions for each tooth type. These differences are shown separately for each experimental group. For the V group, increasing ΔE values were obtained when going from the anterior to the posterior teeth within the same dental bleaching session. ΔE increase between 1st and 2nd sessions was more pronounced for central and lateral incisors of all groups, except by the lateral incisors and canines of the VHP group.

Finally, *Tables 5* and *6* indicate statistical results for the Kruskal-Wallis and two-way ANOVA tests and corresponding Tukey-Kramer post-hoc tests among the experimental groups covered in this study. No statistically significant differences were found for ΔE comparisons between groups.

Clinical Cases

All patients treated in this experiment were documented with initial and final photographs using the Vitapan Classical (Vita Zahnfabrik, H. Rauter GmbH and Co. KG.D-7880 Säckingen, Germany) color scale for later color comparison of results obtained with dental bleaching. *Figure 7* shows photos of 3 clinical cases before and after dental bleaching, one clinical case of each experimental group.

Intrapulpal Heating

Our results indicated an intrapulpal heating 8.5°C after 10 minutes by applying blue LED with hydrogen peroxide gel and 2°C after 30 minutes with violet LED (*Figure 8*).We can conclude that the violet LED used in the protocol of the experiments of illustrated in our clinical study does not heat the inside of the pulp chamber and may not cause damage to dental pulp [16]. In addition, our results agree with the predictions of light transport in biological tissues, especially those with high scattering coefficients as the intrapulpal temperature upon violet light illumination is minimal due to the shallow penetration of violet light in tissues [17-22]. Since the penetration is minimal (about units to tens of microns) the heating due to the light absorption is also superficial, which maximizes the temperature exchange with the surrounding environment.

Table 4: Mean and standard deviation of the color change (ΔE) of each set of teeth at each session, Measurements correspond to colorimetric readings of 4 teeth of each type in the 1st and 2nd whitening sessions. No significant statistical differences (p<0.05) were found between the same type of tooth (e.g., central incisors) of V, VCP, and VHP groups at a given whitening session (e.g., 1st session). V: Violet light illumination group, VCP: Violet light illumination+carbamide peroxide gel 10% group, VHP: Violet light illumination+hydrogen peroxide gel 35%.

	Central incisors (1 st session)	Central incisors (2 nd session)	Lateral incisors (1 st session)	Lateral incisors (2 nd session)	Canines (1 st session)	Canines (2 nd session)	First premolars (1 st session)	First premolars (2 nd session)
V	4.1 ± 3.0	5.7 ± 3.3	5.1 ± 2.9	5.7 ± 3.0	6.0 ± 5.8	8.0 ± 4.7	7.5 ± 4.5	9.9 ± 4.2
VCP	5.2 ± 3.3	7.1 ± 4.1	4.5 ± 3.7	6.3 ± 5.0	6.6 ± 5.7	7.4 ± 3.8	6.1 ± 4.1	5.5 ± 2.8
VHP	4.0 ± 2.2	4.3 ± 3.1	4.7 ± 4.9	3.9 ± 2.0	3.7 ± 2.9	5.2 ± 2.9	5.5 ± 4.2	5.1 ± 4.2

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Figure 4: ∆*E* differences among tooth types grouped by dental bleaching session. Results are shown separately for each experimental group (V: violet light illumination group, VCP: violet light illumination+carbamide peroxide gel 10% group, VHP: violet light illumination+hydrogen peroxide gel 35%). *Note:* () Central incisor; () Canine; () First premolar.



Figure 5: ΔE differences after the 1st or 2nd dental bleaching sessions of each experimental group. Results are displayed separately for each tooth type. *Note:* () 1st session; () 2nd session.



Figure 6: ΔE differences after the 1st or 2nd dental bleaching sessions for each tooth type. These differences are shown separately for each experimental group. CI: central incisors, LI: lateral incisors, C: canines and FP: first premolars. *Note:* () 1st session; () 2nd session.

Tuble 5: P-values of Kruskal- wallis and post-noc lesis for each leeth type at 1° and 2° whilehing sessio	d post-hoc tests for each teeth type at 1 st and 2 nd whitening sessions
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	Central incisors (1 st session)	Central incisors (2 nd session)	Lateral incisors (1 st session)	Lateral incisors (2 nd ses- sion)	Canines (1 st session)	Canines (2 nd session)	First premolars (1 st session)	First pre- molars (2 nd ses- sion)
Kruskal-Wallis p-value	0.68	0.45	0.83	0.57	0.06	0.07	0.4	0.07
Post-hoc test								
V and VCP	0.99	1	0.41	0.76	0.96	0.98	0.99	0.91
V and VHP	0.58	0.49	0.3	0.33	0.58	0.67	0.23	0.14
VCP and VHP	0.49	0.45	0.98	0.76	0.76	0.8	0.18	0.05

Table 6: P-values of two-way A	NOVA and post-hoc test	s for each teeth type at 1^{st}	and 2 nd whitening sessions.
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	Central incisors (1 st session)	Central incisors (2 nd session)	Lateral incisors (1 st session)	Lateral incisors (2 nd session)	Canines (1 st session)	Canines (2 nd session)	First premolars (1 st session)	First premolars (2 nd session)
ANOVA p-value	0.52	0.43	0.92	0.52	0.06	0.19	0.17	0.04
Post-hoc test								
V and VCP	0.95	0.92	0.48	1	0.91	0.98	0.76	0.96
V and VHP	0.89	0.99	0.42	0.78	0.95	0.79	0.61	0.33
VCP and VHP	0.72	0.85	0.99	0.82	0.76	0.88	0.26	0.24



Figure 7: Procedure of evaluation of the whitening effect of dental bleaching after the 1st and 2nd treatment sessions of each experimental group using color scale Vitapan Classical (Vita Zahnfabrik, H. Rauter GmbH and Co. KG.D-7880 Säckingen, Germany). V: violet light illumination group, VCP: violet light illumination+carbamide peroxide gel 10% group, VHP: violet light illumination+hydrogen peroxide gel 35%.



Figure 8: Shows the results show an intrapulpal heating 8.5 °C after 10 minutes by applying blue LED with hydrogen peroxide gel and 2 °C after 30 minutes with violet LED alone [16].

Discussion

In modern odontology as well as in medicine, less invasive treatments, also considered conservative, are preferably indicated as they allow a major preservation of human tissues [23]. In odontology, dental bleaching may be considered a minimally invasive treatment and conservative since its aesthetic results occur without abrasion of dental surfaces. When basic safety assumptions such as time and periodicity of light application and peroxides are respected, in-office dental bleaching does not cause damage to oral soft tissues or structural damage to dental tissues. Dental bleaching can be performed in the office, at home or by combining both methods. Homemade bleaching is less safe due to a higher possibility of contact between chemical agents and living tissues in the buccal cavity and digestive system. The main agents used in dental bleaching are Carbamide Peroxide (CP) and Hydrogen Peroxide (HP) presented in several peroxide concentrations (6%, 10%, 16% or 35% CP or 35 and 38% HP) which, when combined with light, may increase the speed of the dental bleaching process.

In-office dental bleaching is also considered safer because all the process is directly supervised by a professional, thus allowing the control of bleaching agents used on dental surfaces and buccal mucosa. Besides, the procedure is performed regardless of the patient's cooperation and the clinical results may be seen after the first session. In-office dental bleaching delivers fast aesthetic and rejuvenating effects improving the patient's self-esteem.

Peroxides present caustic and carcinogenic effects on soft digestive tissues and should therefore be carefully and safely applied on patient [24]. Another unwanted effect of peroxides is promoting dental sensitivity during and after treatment [25]. Higher dental sensitivity is reported in homemade dental bleaching as exposure time of teeth to chemical agents is longer. Reactive oxygen (O⁻) molecules produced from peroxide gels are responsible for breaking up pigments in teeth whitening and have low molecular weight. This allows reactive oxygen molecules (O⁻) to penetrate the pulp through the dentinal tubules and change the tooth/dental sensitivity leading to pain. The use of hydrogen peroxide in high concentrations between 30% and 38% or carbamide between 35% and 37% for in-office bleaching is the most researched and applied in odontology. Peroxides in these concentrations produce a caustic effect on soft tissues and therefore it is necessary to have use a rubber dam or a gingival barrier for gingival protection. Peroxides also have a demineralizing effect on dental enamel and should be used in a controlled way.

Dental hypersensitivity is the main unwanted effect caused by dental bleaching, resulting in pain and discomfort during and after the procedure [25]. Bleaching gels used in dental bleaching cause dental sensitivity during and after treatment due to the release and diffusion of free oxygen radicals. These radicals with negative charge of oxygen are responsible for the oxidization of pigments that stain teeth. These pigments when oxidized, or fragmented into smaller particles by gels, reflect light in a way that will deliver teeth a lighter color. Oxygen negative radicals (O⁻) present low molecular weight. This oxygen penetrates the enamel's prisms and also microscopic defects in the dental structure until they reach the dentinal tubules causing reversible pulpitis and consequently dental hypersensitivity.

Dental hypersensitivity is experienced by about 50% of adult population and can be increased during and after dental bleaching. It is a bothersome sensation that can affect about 60% of patients submitted to conventional bleaching. Dental hypersensitivity can result from several causes among which is dentinal exposure that allows hydrodynamic flow due to pressure difference in dentinal tubules promoting pain sensitization of the odontoblastic nerve terminals [26]. Another cause could be attributed to a direct stimulus of chemosensitive and thermosensitive nervous terminals. This is based on direct stimulus of dental nerves and their properties of transduction mechanism of the trigeminal primary afferents that innervate the tooth. These nervous fibers are myelinated and responsible for the fast transmission of acute pain through A-delta nervous fibres [27].

This study evaluated both dental bleaching performed with and without peroxides and the results of the effect of violet light regarding dental hypersensitivity developed in treated patients during and after two clinical treatment sessions. Our results of dental hypersensitivity (*Table 2*) indicated that dental sensitivity decreased for all groups after dental bleaching sessions. The observed improvement in dental sensitivity suggests "desensitization" upon application of violet light illumination alone or with peroxide gels. This desensitization causes an effect of pain relief which clearly shown with the reduction of pain scores upon repeated dental bleaching sessions.

Pain relief upon light illumination has been shown by previous studies quantifying this relief upon application of red and infrared light. Light in the red and infrared wavelengths applied in case of dental hypersensitivity through laser or LED has been described as capable of acting on delta A and C fibers causing hyperpolarization of the nervous fiber and promoting pain relief [28]. Literature reports that laser or LED prevent transduction, blocking nervous transmission. In face of this dental sensitivity adverse effect in conventional bleaching, it may be hypothetically stated that application of violet light may have a repolarization effect on nervous terminals inside dentinal tubules, blocking and controlling electric stimuli that provoke algic effect during and after dental bleaching.

Regarding the teeth whitening effect obtained after bleaching by the three experimental groups, the color difference ΔE followed the same trend as Δ blue (*Table 3* and *Figures 4-6*), suggesting the two parameters may be correlated. Another correlation that may be present is the relationship between higher average ΔE and average Δ blue upon higher average initial blue readings of teeth in each group. This relationship may indicate that teeth with higher pigmentation in the blue wavelength range tend to be less affected by dental bleaching using violet light illumination alone or combined with peroxide gels. However, we emphasize that blue readings may also be different for colorimeters compared to cameras and color perception by naked eye, as measurements of this study were collected from a point (the same point was illuminated by the colorimeter with its excitation light and had its reflected light delivered to the detector) probe different tissue depths compared to imaging. Therefore, blue light probes a much more superficial tissue (tens of microns) than green and red light (about hundreds of microns), which makes the blue light more sensitive to whitening effects happening on the surface such as the effects induced by violet light illumination. With this in mind, pigments broken at the tooth surface may be detected in the blue readings of the RGB colorimeter, whereas pigments broken at deeper tissue layers may not be detected by blue light of the colorimeter. Still, the color difference due to less pigmentation in deeper tissue layers will be accessed by green and red light (R and G of the RGB colorimeter), which penetrate deeper into tissue [29-42]. The color perception of a digital spectrophotometer may also be different depending on the geometry and wavelength resolution of the light sent and collected, which may change the color perception in multiple ways. In terms of differences between experimental groups, no groups showed statistically significant differences (p<0.05) for ΔE values.

The concept of deeper light penetration for longer wavelengths of light can be also illustrated by the intrapulpal heating experiment performed in this article. We showed results of an *in vitro* experiment comparing the application of blue and violet light for tooth whitening [16]. In the comparison using 10 minutes continuous blue light and whitening gel on the surface of human teeth treated for dental bleaching in relation to the teeth that only received fractional violet light application in a total of 30 minutes, we can observe that the heating was much smaller (approximately 2°C) when applying violet light. The heating in the treatment with blue light plus the whitening gel was 8.5°C, which is not indicated for tooth whitening, considering the intrapulpal heating studies [43]. In fact, a deeper heating for light at longer wavelengths (wavelength of blue light (450 nm) is longer than violet light (410 nm)) is already expected, as light at longer wavelengths penetrate deeper in biological tissues, especially those with high scattering coefficients such as teeth and bones [18,44-46]. Then, the intrapulpal temperature upon violet light illumination is minimal due to the shallow penetration of violet light (about units to tens of microns) in tissues, whereas blue light illumination leads to a deeper heating due to a slightly deeper penetration (about tens of microns) and absorption at deeper tissue layers [47-59]. The thermal equilibrium is

determined by the depth at which light is absorbed and converted to heat as well as the temperature exchange with the surrounding environment. Since violet light is restricted to the tissue surface, the temperature exchange with the surroundings is much higher than that of blue light. The same is valid for pigment break, which is mainly restricted to the tissue surface.

Previous clinical studies have assessed the total color difference ΔE and dental sensitivity for similar protocols compared to those exploited in this study *(Table 7)*. Based on these studies, our results agree with the ΔE obtained by Gallinari et al. for the VCP group after the 2nd dental bleaching session (ΔE =6.18 ± 2.82 for Gallinari et. al. and ΔE =6.6 ± 4.0 for this study) [60]. However, our results were slightly different compared to those of Kury et al. for the same VCP group (ΔE =7.8 ± 2.0) [61]. For the same study of Kury et al. a discrepancy was also found for the V and VHP groups compared to our study, even though both studies used the same protocol fractionated violet light illumination and similar formulations of hydrogen peroxide gel [61]. With that in mind, the colorimetric evaluation and the patients involved were different, which may be possible causes of such discrepancy. Our results also disagree in terms of dental sensitivity, where a slight sensitivity increase was reported by Kury et al. as opposed to the "desensitization" observed in our study [61]. Because of discrepancies in the results, our main conclusions on ΔE measurements differ from what has been reported in the past (*Table 8*), even when considering *in vitro* studies in bovine teeth [12,62].

Despite controversial studies of dental bleaching showing the need or not to apply light to help increase efficacy of this treatment, our findings as well as those from other authors have shown that violet light stands out as a physical element capable of improving efficacy in dental bleaching treatment [20,63,64].

 Table 7: Comparison of tooth sensitivity and whitening effects of the V, VCP, and VHP groups with respect to the clinical trials conducted in previous studies.

Authors	Light used	Peroxide formulation (bleaching gel) used	Experimen- tal groups	Dental sensitivity technique	Dental sensi- tivity results (comparison with V, VCP, and VHP groups)	Color evaluation	Whiten- ing results (comparison of ΔE of V, VCP, and VHP groups)
This study (Panhóca and Nogueira et. al.)	Violet LED light (V; 405- 410 nm) for 30 minutes (20 cycles of fractioned illumination consisting of 30 seconds of active illumination, 1 minute of pause)	10% Carbam- ide Peroxide (CP) or 35% Hydrogen Peroxide (HP) combined with violet light illumina- tion (V)	V, VCP, and VHP	Visual anal- ysis (VAS) of pain on a scale from 0 to 10 upon application of an air jet on the vestibular surface of all superior and inferior bleached teeth, as well as tests with exploratory probe hori- zontally back and forth three times in the buccal cervical of the bleached teeth.	100% pain reduction for VCP and VHP after 1 st and 2 nd dental bleaching sessions. For the V group, pain reduction of 95.6% after the 1 st session and 96.7% after the 2 nd session.	Subjective color eval- uation with VITA visual shade guide unit and contact-type intraoral spec- trophotometer easy Shade	V after 1 st session: $5.6 \pm$ 4.3, after 2 nd session: $7.3 \pm$ 4.2; VCP after 1 st session: 5.6 ± 4.3 , after 2 nd session: 6.6 ± 4.0 ; VHP after 1 st session: $4.5 \pm$ 3.7, after 2 nd session: 4.6 ± 3.1

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Kury	Violet	37% Carbam-	LED, LED/	Spontaneous	Median	Subjective	LED after
(2020a)61	LED light	ide Peroxide	CP, CP, LED/	indication in	(minimum,	color eval-	2 nd session:
	(LED) for	(CP) or 35%	HP and HP	visual scale	maximum) as	uation with	$\Delta E=3.4 \pm$
	30 minutes	Hydrogen		ranging from	follows: LED	VITA visual	1.3, 14-days
	(20 cycles	Peroxide (HP)		0 to 10 upon	1 st session:	shade guide	follow up:
	of fractioned	along or in		general appli-	0.0 (0.0;4.0),	unit and	$\Delta E=3.7 \pm 1.4;$
	illumination	combinations		cation of the	2 nd session:	contact-type	LED/CP after
	consisting of	with LED		whole dental	0.0 (0.0;4.0),	intraoral spec-	2 nd session:
	30 seconds			bleaching	3 rd session:	trophotometer	$\Delta E=7.8 \pm$
	of active			procedure	0.0 (0.0;2.0);	Easy Shade	2.0, 14-days
	illumination,			-	LED/CP 1st		follow up:
	1 minute of				session: 0.0		$\Delta E = 8.6 \pm 2.1;$
	pause)				$(0.0;3.0), 2^{nd}$		LED/HP after
	· · ·				session: 0.0		2 nd session:
					$(0.0;3.0), 3^{rd}$		$\Delta E=12.9 \pm$
					session: 0.0		2.6, 14-days
					(0.0:7.0):		follow up:
					LED/HP 1 st		$\Delta E=14.4 \pm$
					session: 2.0		2.2
					(0, 0.7, 0) 2 nd		2.2
					session: 1.0		
					(0, 0.9, 0) 3 rd		
					(0.0, 9.0), 9		
					(0.0.7.0)		
Gallinari	Violet I FD	10% Carbam-	CP and VI /	Detection was	No report on	Digital	VI /CP after
(2020)60	light (VI ·	ide Perovide	CP	performed	tooth sensi-	spectropho-	2 nd session:
(2020)00	405 nm-410	(CP)	CI	using the	tivity and/or	tometry for	6.18 ± 2.82
	nm 30 min	(01)		Thermo-Sen-	discomfort	color analysis	0.10 ± 2.02
	twice per			sory Analysis	during the	by using these	
	twice per			IL (TS A II)	blooching	visual analog	
	three weeks)			aquinment	protocol	visual allalog	
	2nd apagier			equipment.	protocol.	scale	
	- 2 ^m session						
	nappened in						
	the first week						

Table 8: The main findings of previous studies on tooth sensitivity and whitening effect on bovine teeth conducted in clinical trials or in vitro experiments.

Authors	Light used	Peroxide formulation (bleaching gel) used	Experimen- tal groups	Dental sensitivity technique	Dental sensitivity findings	Color evaluation	Tooth whitening findings
Kury	Violet	37% Carbam-	LED, LED/	Spontaneous	LED and HP	Subjective	Violet LED
(2020a)61	LED light	ide Peroxide	CP, CP, LED/	indication in	treatments	color eval-	alone (LED
	(LED) for	(CP) or 35%	HP and HP	visual scale	promoted the	uation with	group)
	30 minutes	Hydrogen		ranging from	lowest (16%)	VITA visual	produced the
	(20 cycles	Peroxide (HP)		0 to 10	and highest	shade guide	lowest ΔE
	of fractioned	along or in			(94.4%) of	unit and	$(3.4 \pm 1.3),$
	illumination	combinations			risk of tooth	contact-type	but enhanced
	consisting of	with LED			sensitivity,	intraoral spec-	HP bleach-
	30 seconds				respectively.	trophotometer	ing results
	of active				No significant	Easy Shade	by $\Delta E=4.1$
	illumination,				statistical		after the last
	1 minute of				difference		bleaching ses-
	pause)				groups		sion and by
					(p>0.05) was		$\Delta E=4.4$ after
					found for oth-		14 days. Pa-
					er groups (CP		tients treated
					(44%), LED/		with LED/CP
					CP (61%)		reached the
					and LED/HP		same efficacy
					(88%)).		of HP.

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Kury (2020b)62	Violet LED (VL) light	Non Thermal Atmospher- ic Plasma (NTAP) com- bined with or without 35% Hydro- gen Peroxide (HP) and 37% Carbamide Peroxide (CP)	Bovine teeth: VL, VL/ HP, VL/CP, NTAP, NTAP/ HP, NTAP/ CP, HP, CP, and C (con- trol) groups	not applicable	not applicable	Spectrophoto- metric assess- ment of color and white- ness change (CIELAB ΔEab ,, CIEDE2000 $\Delta E00$, white- ness index ΔWID), color parameters (ΔL , Δa^* , and Δb^*), and intrapulpal concentration ($\mu L/mL$) of HP	VL increased Δ Eab and Δ WID of CP (p<0.05). VL and NTAP alone resulted in perceptible color and whiteness change. This change was lower than those for the gel-treat- ed groups (p<0.05)
Gallinari (2019)12	Violet LED light (VL; 405-410 nm, 30 min, twice per week for three weeks)- 2nd session happened in the first week	Gels contain- ing different concen- trations of Hydrogen Peroxide (HP).	Bovine teeth: GI (placebo without light); GII (35% HP without light); GIII (17.5% HP without light); GIV (placebo with violet LED); GV (35% HP with violet LED); and GVI (17.5% HP with vio- let LEDs)	not applicable	not applicable	Color difference $(\Delta E, \Delta L, \Delta a^*, and \Delta b^*)$ evaluation upon by visi- ble-ultraviolet reflection spectropho- tometer 7 days after each bleach- ing session	GIV differed from the con- trol group in terms of ΔE, GVI exhibit- ed higher ΔE values than GIII
Gallinari (2020)60	Violet LED light (VL; 30 min, twice per week for three weeks)	10% Carbam- ide Peroxide (CP)	CP and VL/ CP	Detection was performed using the Thermo-Sen- sory Analysis II (TSA II) equipment.	No report on tooth sensi- tivity and/or discomfort during the bleaching protocol.	Digital spectropho- tometry for color analysis by using these Visual Analog Scale	Hemiarch ir- radiated with VL presented the highest values of col- or difference compared with the side which did not receive irradiation. VL enhances color alter- ation when combined with 10% CP.

Note: $L^*a^*b^*$ are color space consists of a lightness coordinate (L) ranging from 0 to 100 (black to white), as well as the unbounded (no specific range of values) color dimensions a^* (varying from green to red) and b^* (varying from blue to yellow).

Conclusion

Considering the results and limitations of the present study, we concluded that the application of violet light with or without peroxide gels had a positive effect in obtaining improved in-office dental bleaching. The application of violet light alone (V group) resulted in a more pronounced whitening effect compared to that achieved with combinations with peroxide gels, as larger color differences (ΔE) followed the same trend of blue color difference (Δ blue). It is worth noting color difference findings were obtained with colorimetric point measurements which may probe shallower tissue layers with blue light compared to cameras and naked eye. In addition, we observed that violet light applied on teeth in a fractioned way during dental bleaching did not provoke dental hypersensitivity during or after the dental bleaching procedure. In contrast, violet light promoted "desensitization" by decreasing dental sensitivity in volunteers throughout the study. However, we emphasize that studies with more volunteers should be performed to confirm the results that found in this study.

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Compliance with Ethical Standards

Conflict of Interest

The authors declare no competing interests.

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Ethical Approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed Consent

Informed consent was obtained from all individual participants included in the study.

Author Contributions

VHP Conceptualization, Methodology, Investigation, Resources, Visualization, Writing-Original Draft, Writing-Review and Editing, Project administration; MSN Methodology, Software, Validation, Formal analysis, Resources, Data Curation, Visualization, Writing-Original Draft, Writing-Review and Editing; FAAZ Conceptualization, Writing-Review and Editing, APB Writing-Review and Editing, ABJ Writing-Review and Editing, VSB Resources, Supervision, Funding acquisition.

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