Influence of gel thickness on tooth bleaching efficacy

Influência da espessura do gel na eficácia do clareamento dental

Influencia del espesor del gel en la eficacia del blanqueamiento dental

Abstract
The aim of the study is to evaluate the influence of different thicknesses of bleaching gel on the efficacy of tooth bleaching. Seventy specimens in the form of standardized bovine enamel-dentin blocks were prepared, sequentially polished, and randomly divided into three groups, according to the bleaching agent used: CT (control-no bleaching; n=10) – Artificial saliva; HP (n=30) – 38% Hydrogen Peroxide and CP (n=30) – 10% Carbamide Peroxide. The HP and CP groups were divided into 3 subgroups according to the bleaching gel thickness: A – 0.5mm; B – 1.0mm and C – 2.0mm. The color was measured before and after (24 hours) of the bleaching treatment. Color difference (ΔE) and translucency (TP) were calculated. ΔE variations were statistically analyzed with two-way ANOVA. Translucency values were analyzed with the Student’s T-test (p≤0.05). Regarding, the ΔE and TP values, there were no significant differences between groups (p≥0.05) for different thicknesses of bleaching gel. The thickness of the bleaching gel did not influence the effectiveness of bleaching, regardless of the bleaching agent used.

Keywords: Tooth bleaching; Bleaching agents; Color.

Resumo
O objetivo do estudo é avaliar a influência de diferentes espessuras de gel clareador na eficácia do clareamento dental. Foram confeccionados 70 corpos de prova na forma de blocos padronizados de esmalte-dentina bovina, polidos sequencialmente e divididos aleatoriamente em três grupos, de acordo com o agente clareador utilizado: CT (controle-não clareados; n=10) – Saliva artificial; HP (n=30) – 38% de peróxido de hidrogênio e CP (n=30) – 10% de peróxido de carbamida. Os grupos HP e CP foram divididos em 3 subgrupos de acordo com a espessura do gel clareador: A – 0.5mm; B – 1.0 mm e C – 2.0 mm. A cor foi medida antes e após (24 horas) o tratamento clareador. A diferença de cor (ΔE) e a translucidez (TP) foram calculadas. As variações de ΔE foram analisadas estatisticamente com ANOVA de duas vias. Os valores de translucidez foram analisados com o teste T de Student (p≤0.05). Em relação aos valores de ΔE e TP, não houve diferenças significativas entre os grupos (p≥0.05) para diferentes espessuras de gel clareador. A espessura do gel clareador não influenciou na eficácia do clareamento, independente do agente clareador utilizado.

Palavras-chave: Clareamento dental; Clareadores; Cor.

Resumen
El objetivo del estudio es evaluar la influencia de diferentes espesores de gel blanqueador sobre la eficacia del blanqueamiento dental. Se prepararon 70 especímenes en forma de bloques estandarizados de esmalte-dentina bovina, pulidos secuencialmente y divididos aleatoriamente en tres grupos, de acuerdo con el blanqueador utilizado: CT (control-sin blanqueamiento; n=10) – Saliva artificial; HP (n=30) – 38% Peróxido de hidrógeno y CP (n=30) – 10% Peróxido de carbamida. Los grupos HP y CP se dividieron en 3 subgrupos según el espesor del gel blanqueador: A – 0.5 mm; B – 1.0 mm y C – 2.0 mm. El color se midió antes y después (24 horas) del tratamiento de decoloración. Se calcularon la diferencia de color (ΔE) y la translucidez (TP). Las variaciones de ΔE se analizaron estadísticamente con ANOVA de dos vías. Los valores de translucidez se analizaron con la prueba T de Student (p≤0.05). En cuanto a los valores de ΔE y TP, no hubo diferencias significativas entre grupos (p≥0.05) para diferentes espesores de gel blanqueador. El espesor del gel blanqueador no influyó en la efectividad del blanqueamiento, independientemente del agente blanqueador utilizado.

Palabras clave: Blanqueamiento de dientes; Blanqueadores; Color.
1. Introduction

Tooth bleaching has been described in the literature for over a century (Alqahtani, 2014; Kihn, 2007; Li, 1996). Initially, all the methods described were intended for the treatment of non-vital teeth (Alqahtani, 2014). Latimer et al., presented the first report of bleaching of vital teeth, using oxalic acid only in 1868, during the debate of the society of dental surgeons. Since then, several products have been introduced to the market and several studies have been carried out to prove the efficacy and safety of these products for tooth bleaching (Dietschi et al., 2010; Hannig et al., 2007; Dahl & Pallesen, 2003; Sulieman, 2008).

The bleaching agents used for bleaching are hydrogen peroxide, carbamide peroxide, and sodium perborate (Sulieman, 2008; Kwon & Wertz, 2015). In all three bleaching agents, the active bleaching agent is hydrogen peroxide, which breaks down into reactive free radicals that degrade the pigments present in the tooth structure through an oxidation reaction. (Dahl & Pallesen, 2003; Kwon & Wertz, 2015; Plotino et al., 2008). The mechanism of action of bleaching is not completely elucidated, but it is practically based on three steps: diffusion of free radicals through the tooth structure; breakdown of macromolecules that pigment the tooth structure, known as chromophores; and alteration of the absorption and reflection of light on the dental surface (Kwon & Wertz, 2015).

Bleaching regimens for vital teeth can be classified according to technique: at-home bleaching, in-office bleaching, and combined bleaching (Hannig et al., 2007; Dahl & Pallesen, 2003; Sulieman, 2008; Kwon & Wertz, 2015; Matis 2003; Tredwin, 2006; de Souza et al., 2020). At-home bleaching is performed by the patient at home, under the guidance of the dentist. Since the patient will be responsible for the application of the bleaching gel, bleaching agents with low concentrations are used: 3% to 9.5% Hydrogen peroxide or 5% to 22% Carbamide peroxide (Matis, 2003; Tredwin et al, 2006). The bleaching gel is applied in an individual tray, for 2 to 8 hours daily, during a period of 2 to 6 weeks (Haywood, 2000). In-office bleaching is performed under the direct supervision of the dentist. This is responsible for the application of the bleaching gel, which has high concentrations: 35% to 50% Hydrogen peroxide or 35% to 40% Carbamide peroxide (Tredwin et al., 2006).

The number of applications of the bleaching gel may vary depending on the concentration of the bleaching agent (Sulieman et al., 2004; Bernardon et al., 2010; Moghadam et al., 2013). Leonard et al., (2001) observed that concentrations of 10% and 16% of carbamide peroxide bleached more quickly in two weeks than concentrations of 5%. However, after three weeks of treatment, the 5% concentration achieved a bleaching effect similar to that of higher concentrations.

However, there is no consensus in the literature and among manufacturers about the ideal thickness of bleaching gel, nor about its influence on the efficacy of tooth bleaching and sensitivity. Most studies employ an amount of gel ranging from 1 to 2 mm for both carbamide and hydrogen peroxide (Al-Harbi et al., 2013; Batista et al., 2013; Borges et al., 2015; Caneppele et al., 2013; D’Arce et al., 2013; Torres et al., 2010; Travassos et al., 2010; Wiegand et al., 2005; Wiegand et al., 2008). It is known that the action of peroxides is dependent on reaction conditions, including temperature, pH, light, and the presence of transition metals (Joiner, 2006).

The present study aimed to evaluate the influence of different thicknesses of bleaching gel on the efficacy of tooth bleaching. The null hypotheses tested were: 1) The thickness of the bleaching gel does not influence the bleaching effectiveness 2) The thickness of the bleaching gel does not influence the translucency.

2. Methodology

Specimen preparation

Specimens were prepared according to the method described by Wiegand et al., (2005) with modifications. Seventy healthy, freshly extracted bovine incisors were cleaned and stored in 0.1% thymol solution at room temperature (~22 °C) until preparation - Figure 1a.
Each crown was sectioned with a diamond disc (Buehler), in a serial cutting machine (IsoMet 1000 Precision Cutter, Buehler) with constant water cooling, obtaining blocks with an area of 36mm² (8mm wide and 8mm long) – Figure 1b. To standardize the enamel-dentin thickness to 2mm (1mm enamel and 1mm dentin), the buccal and pulp surfaces of the blocks were polished. They flattened in a polisher (DP-10, Panambra), with sequential silicon carbide discs (grit sizes, #600, #800, and #1200), under water irrigation. Between each disc application, the specimens were rinsed in an ultrasonic bath with distilled water for 10min (Ultrasonic Washer 1440D, Odontobras). The thickness of the specimens was checked with a digital caliper (520.105BL, King Tools) and those with non-compatible dimensions, surfaces with cracks, and/or imperfections were discarded. A total of 70 specimens were obtained in block form.

The specimens were then accommodated in the center of a PVC ring (Tigre) 12 mm in diameter and 2 mm in thickness. The ring and specimen space were filled with acrylic resin (Jet, Lapa). The set was taken to a vice adapted to a microhardness tester (Sematic, Alpha) to allow the buccal and pulpal surface of the block to remain parallel with the external surface of the PVC ring – Figure 1c. After polymerization of the acrylic resin, the specimens were rinsed in an ultrasonic bath to remove the excess acrylic resin present on the enamel and dentin surface.

The specimens (n=70) were randomly divided into 3 groups according to the bleaching agent used: CT (control-no bleaching; n=10) – Artificial saliva; HP (n=30) – 38% hydrogen Peroxide (Opalescence Boost, Ultradent), and CP (n=30) – 10% Carbamide Peroxide (Opalescence PF 10%, Ultradent). The HP and CP groups were divided into 3 subgroups according to the bleaching gel thickness: A – 0.5mm; B – 1.0mm, and C – 2.0mm.

To standardize the gel thicknesses in the HP and CP subgroups, a PVC matrix 12 mm in diameter and with the same thickness as in each subgroup (0.5mm, 1mm, and 2mm) was fixed to the specimens with plastic adhesive for PVC (Tigre, Joinville, SC, Brazil), Figure 1d. Thickness standardization of the matrix was performed with a caliper.

Figure 1. Specimen preparation. a - Marking of the central area of the bovine tooth crown. b - Preparation of specimens with 8 mm side and 2 mm thickness (1 mm enamel and 1 mm dentin). c - Insertion of specimens into 2 mm thick PVC discs with acrylic resin. d - Tint with a thickness of (0.5 mm, 1 mm, and 2 mm) glued on the ring. e - Application of the bleaching gel. f - Leveling the bleaching gel.

Source: Authors.
Bleaching

After storage in artificial saliva for 24 hours, the bleaching procedures were performed according to the manufacturers' instructions. In the HP group, the 38% hydrogen peroxide gel was applied once a week, with three applications of 15 min (3 x 15 min) per appointment, for 4 weeks. Between applications, the gel was removed with a disposable surgical suction device, and the gel was renewed. The bleaching gel was applied in slight excess, which was then removed and leveled to the height of the matrix with the aid of a plastic spatula – Figure 1e-f. In the CP group, the 10% carbamide peroxide gel was left undisturbed on the surface for 8 hours for 28 days (8 h / 28 days). The bleaching gel was applied with a slight excess to the enamel surface, which was then pressed against a glass plate interposed by a transparent PVC film. In this way, the gel was leveled to the thickness determined by the matrix. To simulate the clinical circumstances of high humidity, a moistened gauze was placed on the pulp surface of the specimens and a new glass plate was placed on top.

After the bleaching procedure, the specimens were rinsed with distilled water for 60 seconds to remove the bleaching gel. They were then stored in artificial saliva at 37±1°C until the next treatment. The specimens of the control group were stored in artificial saliva for 24 hours.

Color measurement

The color was measured using the coordinates L*, a*, and b* established by the Commission Internationale de l’Eclairage (CIE), which locates the color of an object in three-dimensional space. The L* axis represents the degree of luminosity and varies from 0 (black) to 100 (white), the a* plane represents the degree of green/red color and the b* plane represents the degree of blue/yellow. The L*, a*, and b* coordinates of each specimen were evaluated according to the environmental conditions standardized by the CIE in 1976, with a spectrophotometer (EasyShade, VITA). Six consecutive measurements were performed on each specimen, three on a white background and three on a black background. To measure the color, the specimens were carefully dried, without dehydrating, then placed on the background. The sensor on the device was firmly positioned in the center of the specimens at a right angle to the enamel surface, with the aid of a silicone addition guide (Express XT, 3M ESPE), with a 6mm central hole, the same diameter as the measuring tip of the device. Color measurement was performed before (initial) and 24 hours after the bleaching treatment. The color difference after bleaching was observed by calculating the variation in the L* (ΔL), a* (Δa), and b* (Δb), measured on a white background. The total color difference (ΔE*ab) was calculated using the following formula:

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

Translucency was calculated according to the formula below, using the coordinates of the CIE-Lab system, where the subscript P refers to the color coordinates of the specimen on the black background and subscript B to the measurements on the white background.

$$TP = \sqrt{(L_P^* - L_B^*)^2 + (a_P^* - a_B^*)^2 + (b_P^* - b_B^*)^2}$$

Statistical analysis

The number of specimens per group was based on previous studies (Schilke et al., 2000; ten Cate et al., 1986; Tagami et al., 1989). The normality of the data was verified by the Kolmogorov-Smirnov and Levene test. ΔE variations were statistically analyzed with two-way ANOVA. Variations in translucency values, L*, a* and b* were analyzed with the Student’s T-test. The analyses were performed at a significance level of $\alpha = 5\%$. 
3. Results

The mean values and standard deviation (SD) of ΔE in HP and CP groups according to the bleaching gel thicknesses are shown in Table 1. Two-way ANOVA showed that there was no statistical difference between the evaluated groups (p>0.05).

### Table 1: ΔE values (mean± SD) and two-way ANOVA results.

| Groups | ΔE     |
|--------|--------|
|        | A      | B      | C      |
| HP     | 16 ± 3.00<sup>AA</sup> | 17 ± 2.83<sup>AA</sup> | 16 ± 3.18<sup>AA</sup> |
| CP     | 16 ± 3.03<sup>AA</sup> | 15 ± 2.84<sup>AA</sup> | 14 ± 2.74<sup>AA</sup> |

Different lowercase letters within the column and uppercase letters within each row indicate significant differences (p <0.05).

Source: Authors.

The initial and final mean values and SD of translucency, L*, a*, and b* in the CT group, and HP and CP groups according to the bleaching gel thicknesses are shown in table 2. The student's T-test showed that there was no statistically significant difference between the initial and final translucency in all groups (p>0.05). However, the L* parameter significantly increased in all groups (p<0.05), except the CT group, and the a* and b* parameters significantly decreased in all groups (p<0.05), except the CT group.

### Table 2: Initial and final values (mean± SD) of translucency, L*, a*, and b*

| Evaluation period | CT     | HP     | CP     |
|-------------------|--------|--------|--------|
|                   | A      | B      | C      | A      | B      | C      |
| Translucency      |        |        |        |        |        |        |
| Initial           | 10 ± 2.54<sup>a</sup> | 10 ± 3.09<sup>a</sup> | 8 ± 2.51<sup>a</sup> | 10 ± 2.83<sup>a</sup> | 10 ± 1.89<sup>a</sup> | 8 ± 2.56<sup>a</sup> | 9 ± 2.18<sup>a</sup> |
| Final             | 10 ± 2.29<sup>a</sup> | 10 ± 3.08<sup>a</sup> | 8 ± 2.04<sup>a</sup> | 10 ± 2.48<sup>a</sup> | 10 ± 2.37<sup>a</sup> | 8 ± 3.19<sup>a</sup> | 9 ± 2.16<sup>a</sup> |
| L*                |        |        |        |        |        |        |
| Initial           | 88 ± 2.87<sup>a</sup> | 88 ± 2.87<sup>a</sup> | 89 ± 2.24<sup>a</sup> | 87 ± 2.65<sup>a</sup> | 87 ± 3.79<sup>a</sup> | 87 ± 3.28<sup>a</sup> | 87 ± 1.50<sup>a</sup> |
| Final             | 90 ± 3.22<sup>a</sup> | 95 ± 3.42<sup>a</sup> | 96 ± 3.24<sup>a</sup> | 94 ± 3.54<sup>a</sup> | 94 ± 3.57<sup>a</sup> | 94 ± 2.89<sup>a</sup> | 94 ± 1.94<sup>a</sup> |
| a*                |        |        |        |        |        |        |
| Initial           | 2 ± 1.55<sup>a</sup> | 3 ± 1.04<sup>a</sup> | 2 ± 0.94<sup>a</sup> | 3 ± 1.07<sup>a</sup> | 3 ± 1.20<sup>a</sup> | 3 ± 1.00<sup>a</sup> | 3 ± 0.97<sup>a</sup> |
| Final             | 2 ± 1.51<sup>a</sup> | -0.4 ± 0.69<sup>a</sup> | -0.7 ± 0.69<sup>a</sup> | -0.4 ± 0.73<sup>a</sup> | -0.1 ± 0.85<sup>a</sup> | -0.05 ± 0.48<sup>a</sup> | 0.1 ± 0.97<sup>a</sup> |
| b*                |        |        |        |        |        |        |
| Initial           | 40 ± 4.79<sup>a</sup> | 40 ± 5.52<sup>a</sup> | 40 ± 4.85<sup>a</sup> | 41 ± 5.14<sup>a</sup> | 40 ± 4.26<sup>a</sup> | 39 ± 4.70<sup>a</sup> | 41 ± 2.51<sup>a</sup> |
| Final             | 39 ± 5.8<sup>a</sup> | 21 ± 3.73<sup>b</sup> | 21 ± 2.61<sup>b</sup> | 22 ± 2.61<sup>b</sup> | 25 ± 1.86<sup>b</sup> | 24 ± 3.06<sup>b</sup> | 25 ± 1.71<sup>b</sup> |

Different lowercase letters within the column indicate significant differences (p <0.05).

Source: Authors.

4. Discussion

The current study analyzed two established techniques for tooth bleaching; supervised at-home tooth bleaching with 10% carbamide peroxide and in-office tooth bleaching with 38% hydrogen peroxide. Both bleaching agents were applied
according to the manufacturer's instructions.

In the present study, specimens were obtained from bovine incisors. Bovine teeth have anatomy and physicochemical structure similar to human teeth (Schilke et al., 2000; ten Cate et al., 1986; Tagami et al., 1989). In addition, it has been used in several studies that evaluated color during bleaching and represents a viable model for the evaluation of tooth bleaching (Dietschi, Benbachir & Krejci, 2010; Al-Harbi et al., 2013; Batista et al., 2013; Caneppele et al., 2013; D’Arce et al., 2013; Torres et al., 2010; Travassos et al., 2010).

Tooth bleaching is considered satisfactory when there is a decrease in \(b^*\) values (color change from yellow to blue) and an increase in \(L^*\) values (quantification of black and white) (Dietschi, Benbachir & Krejci, 2010; Borges et al., 2015; Gerlach, Barker & Sagel, 2002; Monteiro, Monteiro & Caldeira de Andrade, 2018). The \(\Delta E\) values are also frequently used in tooth bleaching studies (Dietschi, Benbachir & Krejci, 2010; Bernardon et al., 2010; Al-Harbi et al., 2013; Batista et al., 2013; Borges et al., 2015; Caneppele et al., 2013; D’Arce et al., 2013; Monteiro, Monteiro & Caldeira de Andrade, 2018). From a clinical point of view, \(\Delta E\) values equal to or higher than 3.7 promote clinically perceptible color changes (Khashayar et al., 2014).

In this present study, regardless of the bleaching agent or thickness used, the \(\Delta E\) values were higher than 3.7, and the \(L^*\) increased and the \(b^*\) decreased after bleaching treatment. Therefore, all groups showed bleaching efficacy. This leads to the acceptance of the first null hypothesis. These findings may be related to different factors. First, due to the gel consistency of the bleaching tested, only the portion of the product that is closest to the tooth may be more effective in bleaching since the more distant portions are unlikely to be able to mix with the product that is closer to the tooth structure. Second, tooth permeability allows bleaching to be effective, regardless of the thickness of the applied gel (Kwon & Wertz, 2015; Bertacci et al., 2021).

In this study, the at-home bleaching technique with 10% carbamide peroxide and the in-office technique with 38% hydrogen peroxide had the same bleaching efficacy. Similar results were observed in a clinical study, which evaluated and compared the clinical performance and durability of three bleaching methods: supervised at-home bleaching (10% Carbamide peroxide – CP10%), in-office bleaching (35% Peroxide Hydrogen – HP35%) with and without light, and a combined at-home/in-office technique. The authors observed that the degree of bleaching obtained by the at-home technique was similar to the in-office technique, regardless of irradiation with light or the combination of techniques (Bernardon et al., 2010). This is also in agreement with the findings of Zekonis et al., (2003) who clinically compared the at-home (CP10%) bleaching technique with the in-office (35% HP) bleaching technique, in terms of color change and stability. In general, the authors did not observe differences between the techniques.

It is assumed that the creation of reservoirs in the individual impression tray increases the amount of gel, the availability of peroxide, and, consequently, the bleaching effect. However, clinical studies have observed that bleaching efficacy is the same regardless of the presence or absence of reservoirs (Matis, Hamdan, et al., 2002; Matis, Yousef et al., 2002; Strassler, 1997).

Carbamide peroxide 10% contains the equivalent of 3.6% hydrogen peroxide and is considered safe when applied to individual impression trays overnight (Haywood, 1994; Marshall, Cancro & Fischman, 1995). Carbamide peroxide has a slower reaction rate than hydrogen peroxide, especially at room temperature. Hydrogen peroxide is released in the first few minutes after contact with the tooth surface, while carbamide peroxide remains active after contact, prolonging the release of peroxide (Dhillon et al., 2011; Marson, Sensi & Rodrigo Reis, 2008; Mokhlis et al., 2000; Kawamoto & Tsujimoto, 2004).

Although renewal of bleaching gel during the in-office appointment is indicated by some manufacturers to maintain the reactivity of the bleaching agent (hydrogen peroxide) during the appointment, a recent systematic review and meta-analysis concluded that color change was not impacted by the bleaching gel application regimen (Kury et al., 2022). Know et al., (2013)
also observed that a single application of 38% hydrogen peroxide for one hour was as effective as exchanges every 20 minutes. However, a controversial study exists, Reis et al., (2011), observed that three 15-minute applications of 35% hydrogen peroxide were more effective than a single 45-minute application.

In this present study, the translucency assessment showed that there was no difference between the initial and final values for all groups. Thus, the second null hypothesis was also accepted. Similar results were observed by Caneppele et al., (2013) when evaluating the color and translucency of bovine enamel and dentin (separated and joined) submitted to the bleaching technique with CP10% and HP35%. Regarding initial and final translucency, the authors found no significant differences for the enamel-dentin group (2mm) between the control, CP10%, and HP35% subgroups. Therefore, differences were observed when the substrates were evaluated individually (1mm thick each). According to the authors, this is because enamel-dentin specimens are 2 mm thick, which is twice the thickness of the separate substrates. As described by Yu et al., (2009) the translucency of enamel and dentin increases in inverse proportion to thickness.

The experimental design of the present study was intended to simulate a clinical situation to assess the effect of different thicknesses of bleaching gel on the efficacy of tooth bleaching and change in translucency. The results obtained in this study may be different from those observed clinically since several intrinsic and extrinsic factors are related to the efficacy of tooth bleaching. Due to this alternative scenario, future clinical research is necessary to support the results found in the present study.

5. Conclusion

Within the limitations of this laboratory study, it can be concluded that the thickness of the bleaching gel did not influence the effectiveness of bleaching, regardless of the bleaching agent used.

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