

Spectrophotometric Analysis of Plateau Effect in Carbamide Peroxide at 10% and 16% Concentrations

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ABSTRACT

Objectives: Aesthetics, particularly a bright smile, hold significant value in today's society. Tooth whitening is the most common, minimally invasive treatment to achieve this. This study aims to identify the plateau phase of tooth whitening, which is crucial for optimizing treatment and minimizing side effects due to overuse.

Materials & Methods: Thirty extracted human incisors and canines were treated with 10% and 16% carbamide peroxide gel for 28 days. Colour parameters (ΔE , Δa , ΔL , Δb) were measured daily using photospectrometry until day 21. Data were analyzed for statistical significance.

Results: Analysis revealed that the colour difference (ΔE) plateaued by day 20 for both tooth types. A significant difference ($P < 0.0001$) between canines and incisors was found at 10% concentration but not at 16%. Brightness (ΔL) also plateaued by day 20, with significant differences ($P < 0.0001$ and $P < 0.01$) for 10% and 16% concentrations, respectively. The red-green axis (Δa) showed significant differences ($P < 0.0001$) for both concentrations, while the yellow-blue axis (Δb) showed a significant difference ($P < 0.001$) at 10% but none at 16%.

Conclusion: The findings indicate that incisors and canines reach a colour change plateau by day 20. At 16%, whitening effects were more consistent between tooth types, suggesting higher concentrations promote uniform results.

Keywords: Spectrophotometry, Carbamide Peroxide, Dose-Response Relationship

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INTRODUCTION

Today's society places great importance on appearance, specifically the smile. Tooth whitening is the most common and least invasive treatment for achieving this goal.¹ Knowing when the plateau phase is reached helps optimize treatment efficacy. In other words, further application of the whitening product will no longer significantly improve tooth shade.

Tooth shade depends on several factors, including extrinsic and intrinsic stains, also known as chromophores. Extrinsic stains are due to the deposition of pigments on the tooth surface and result from poor oral hygiene, certain foods, or smoking. They are superficial and can be easily removed by tooth brushing or dental prophylaxis.²⁻⁴ Intrinsic stains are more challenging to eliminate as they are incorporated into the tooth structure. They may appear due to systemic conditions, such as a genetic disease affecting the tooth's hard tissues (amelogenesis imperfecta, dentinogenesis imperfecta), or be caused by pre- or post-eruptive factors, including age or the use of certain medications. A product, such as a tooth-whitening agent that can penetrate enamel and dentin, is needed to remove these discolourations.⁵⁻⁷ Tooth-whitening products can be applied in a dental office or at home. They differ in their active agent, concentration, and application time. The most recurrent oxidizing active agent is hydrogen peroxide (H_2O_2). Hydrogen peroxide (H_2O_2) is used directly or is generated through a chemical reaction from carbamide peroxide ($CH_6N_2O_3$). In the second case, carbamide peroxide decomposes into hydrogen peroxide (H_2O_2) and urea (NH_2CONH_2). Hydrogen peroxide then breaks down, producing water (H_2O) and oxygen (O_2). These oxygen-free radicals play a vital role in the oxidation of the chromophore molecules that are responsible for colouration.⁸

Recently, interest in whiter teeth has grown considerably, fueled by beauty ideals promoted in the media and social networks. However, patients and dental practitioners must be well-informed about the scientific and practical aspects of these treatments. Knowledge of the plateau phase in tooth whitening is necessary to ensure practical, safe, and economical application while maximizing patient satisfaction.

The main objective of this study is to identify the plateau phase of tooth whitening with the use of two

concentrations of Carbamide peroxide. The determination of the precise moment is essential to optimizing treatment results while avoiding product overuse, which can lead to undesirable side effects. By determining this plateau phase, we can provide more precise, scientifically based guidelines for practitioners and patients, ensuring safe and effective treatments.

MATERIALS & METHODS

Ethics

The Ethics Committee of the Children's Hospital of Queen Fabiola, Free University of Brussels, approved the study under C.E.H. number 62/15.

Informed consent was obtained from all individuals who provided extracted teeth for the study. The teeth were collected from patients who underwent periodontal treatments and required dental extractions as part of their clinical care. The donors consented to using their extracted teeth for research purposes. The study was conducted by the ethical principles outlined in the Declaration of Helsinki.

Selection of teeth

30 extracted human maxillary and mandibular incisors and 30 canines were selected according to the following exclusion criteria

- Presence of caries
- Presence of erosion
- Presence of restorations
- Presence of fissures/cracks

Teeth were scaled and brushed, placed on a polystyrene tray, and numbered. The two groups of teeth (incisors and canines) were equally distributed for the 10% and 16% concentrations.

All specimens were stored in 0.9% physiological saline solution at 37°C in an incubator (Eldelstahl Rostfrei), both before the start of the study and throughout the entire experimental period. Storing the teeth under these conditions-maintained hydration and a constant temperature, simulating intraoral conditions. After each bleaching session, the specimens were rinsed with distilled water and returned to the saline solution in the incubator.

Selection and application of whitening gel

Opalescence™ PF (Ultradent, UT, USA) whitening gel with formulations containing 10% and 16% carbamide peroxide, respectively, was selected. The gels were applied to the vestibular surfaces of the specimens in a 1 mm layer daily for 28 days. According to the manufacturer's recommendations, the gel should be applied between 4 and 6 hours.

Therefore, we left it on for 5 hours. After each application, all teeth were rinsed and dried and returned to the saline solution in the incubator until the next application. The saline solution was changed daily throughout the 28 days.

Determination of tooth shade

To determine the shade of the teeth, we used a VITA Easyshade V® spectrophotometer. This device determines tooth color using three parameters:

- The L* parameter, representing luminosity, ranging from 0 (black) to 100 (white)
- The a* parameter, representing the red (a>0) and green (a<0) components
- The b* parameter represents the yellow (b>0) and blue (b<0) components.

At each measurement, the tip of the spectrophotometer was positioned at the center of the tooth, and the device was calibrated for every shade reading.

Data processing

Our study adopted a methodical approach to color measurements to determine tooth shade and identify the plateau phase of tooth whitening products. These measurements were taken at four key points: before starting treatment, after each 5-hour application, before the next gel application, and one week after the end of treatment to allow for stabilization of tooth color. This procedure made it possible to monitor changes in tooth color throughout the whitening process.

All shade values were recorded in an Excel table and analyzed using the CIE 1976 ΔE formula (CIELAB color difference metric), where,⁹

$$\Delta E^* = \sqrt{(L_2^* - L_1^*)^2 + (a_2^* - a_1^*)^2 + (b_2^* - b_1^*)^2}$$

- ΔE represents the color difference between 2 colors of the same object.

- If ΔE is 0, there is no perceptible difference between the two colors.
- If ΔE is positive and its value exceeds 2, there is a perceptible difference between the two colors to the human eye.

L₁, a₁, and b₁ correspond to the color parameters before treatment, while L₂, a₂, and b₂ correspond to the color parameters after treatment.

Each measurement was taken in the exact location and under standardized lighting conditions. The tip of the spectrophotometer was placed perpendicular to the central area of each tooth's buccal surface.

Four points on each surface were measured and averaged for each tooth every time. The device was calibrated before each measurement session.

All measurements were performed by a single calibrated operator trained using the spectrophotometer.

The measurements were recorded in an Excel file and analyzed using descriptive statistics to determine the average color change for each experimental group. Data normality was verified using the Shapiro–Wilk test. Comparisons between two groups were conducted using the Student's t-test, while one-way ANOVA followed by Tukey and Dunn post hoc tests was applied for multiple group comparisons. Four experimental groups were based on the combination of tooth type (incisors or canines) and bleaching agent concentration (10% or 16% carbamide peroxide). ANOVA was used to compare these groups and evaluate the plateau effect of bleaching according to tooth type and peroxide concentration. A significance level of $p < 0.05$ was considered.

All statistical analyses were performed using GraphPad Prism version 10.4.0 (GraphPad Software Inc., La Jolla, CA, U.S.A)

RESULTS

The descriptive analysis of bleaching outcomes revealed a concentration-dependent effect across both tooth types. For ΔE , both concentrations resulted in greater whitening in incisors than in canines, with mean values of 4.92 (± 1.48) and 5.24 (± 1.66) for incisors and 4.47 (± 1.37) and 4.71 (± 1.30) for canines in the 10% and 16% groups, respectively. While ΔE values in the 10% group did not follow a normal distribution ($p < 0.01$), normality was only observed for incisors at 16% ($p = 0.0910$).

Lightness changes (ΔL^*) followed a similar trend, showing higher increases in incisors (1.60 ± 0.54 and 1.66 ± 0.66) than in canines (1.21 ± 0.11 and 1.16 ± 0.66), although normality was inconsistently distributed. Red-green shifts (Δa^*) were more negative in incisors across both concentrations, but normality was not confirmed for canines at 16% ($p = 0.0115$) and 10% ($p = 0.0139$).

The most significant colour change was observed along the yellow-blue axis (Δb^*), especially in incisors treated with 16% carbamide peroxide (-6.63 ± 1.69) compared to 10% (-4.45 ± 1.68), with both showing non-normal distributions (Table 1 and 2).

Table 1: Descriptive statistics for ΔE , ΔL , Δa , and Δb for incisors and canines treated with 10% carbamide peroxide

Parameter	Tooth Type	Mean	$\pm SD$	SEM	P-value normality
ΔE	Incisor	4.92	1.48	0.31	0.0064
ΔE	Canine	4.47	1.37	0.29	0.0007
ΔL^*	Incisor	1.6	0.54	0.52	0.0059
ΔL^*	Canine	1.21	0.11	0.11	0.1672
Δa^*	Incisor	-0.44	0.13	0.03	0.6266
Δa^*	Canine	-0.16	0.1	0.02	0.0139
Δb^*	Incisor	-4.45	1.68	0.35	0.0351
Δb^*	Canine	-4.15	1.4	0.29	0.0012

Table 2: Descriptive statistics for ΔE , ΔL , Δa , and Δb for incisors and canines treated with 16% carbamide peroxide

Parameter	Tooth Type	Mean	$\pm SD$	SEM	P value normality
ΔE	Incisor	5.24	1.66	0.35	0.091
ΔE	Canine	4.71	1.3	0.27	0.0015
ΔL^*	Incisor	1.66	0.66	0.14	0.0638
ΔL^*	Canine	1.16	0.66	0.14	0.5391
Δa^*	Incisor	-0.3	0.11	0.02	0.1911
Δa^*	Canine	-0.18	0.09	0.02	0.0115
Δb^*	Incisor	-6.63	1.69	0.36	0.0182
Δb^*	Canine	-4.28	1.33	0.28	0.0003

Figure 1 shows the colour difference [ΔE] parameter in the 2 groups with the graphical analysis indicating that the value of ΔE reaches a plateau from day 20th day. Data analysis shows that the plateau phase is similar for incisors and canines. There is a statistically

significant difference ($P < 0.0001$) between canines and incisors in terms of the ΔE variable in 10% concentration. However, there is no statistically significant difference between canines and incisors in terms of the ΔE variable in 16% concentration

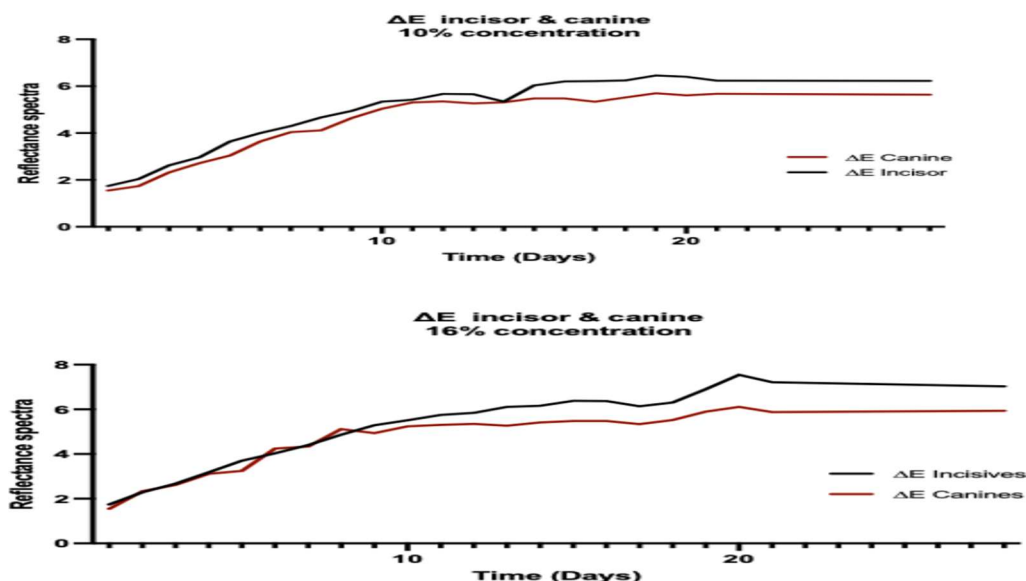


Figure 1: Comparison of ΔE , values between incisors and canines at 10% and 16% concentrations

Figure 2 shows brightness for both concentrations (ΔL), and graphical analyses indicate that the ΔL value reaches a plateau from day 20. Data analysis shows that the plateau phase is similar for incisors

and canines. A statistically significant difference was observed in the ΔL variable between canines and incisors, with p-values of <0.0001 (10%) and <0.01 (16%), respectively

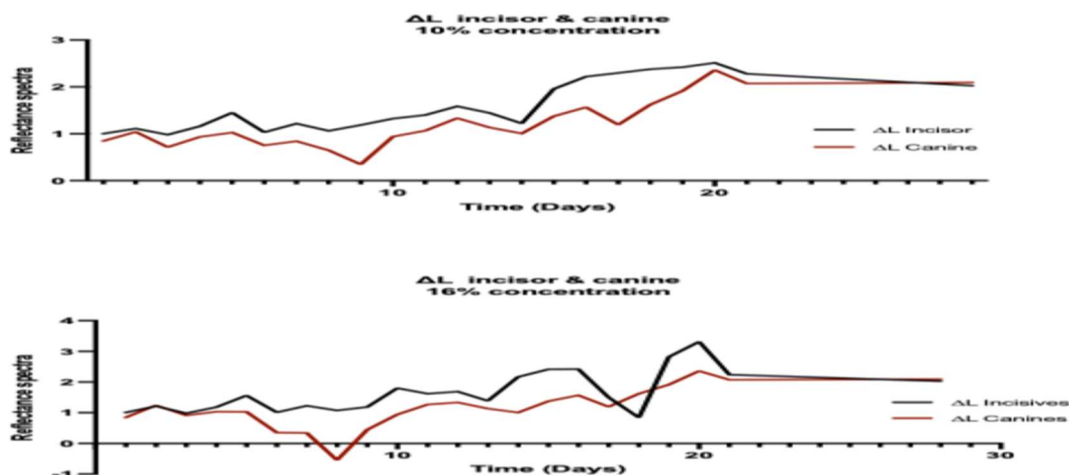


Figure 2: Comparison of ΔL , values between incisors and canines at 10% and 16% concentrations

Figure 3 shows the Red-Green axis parameter (Δa) with Graphical analysis indicating that the value of Δa reaches a plateau from the 20th day. Data analysis shows that the plateau phase is similar for incisors

and canines. There is a statistically significant difference ($P<0.0001$) between canines and incisors in terms of the Δa variable in both concentrations

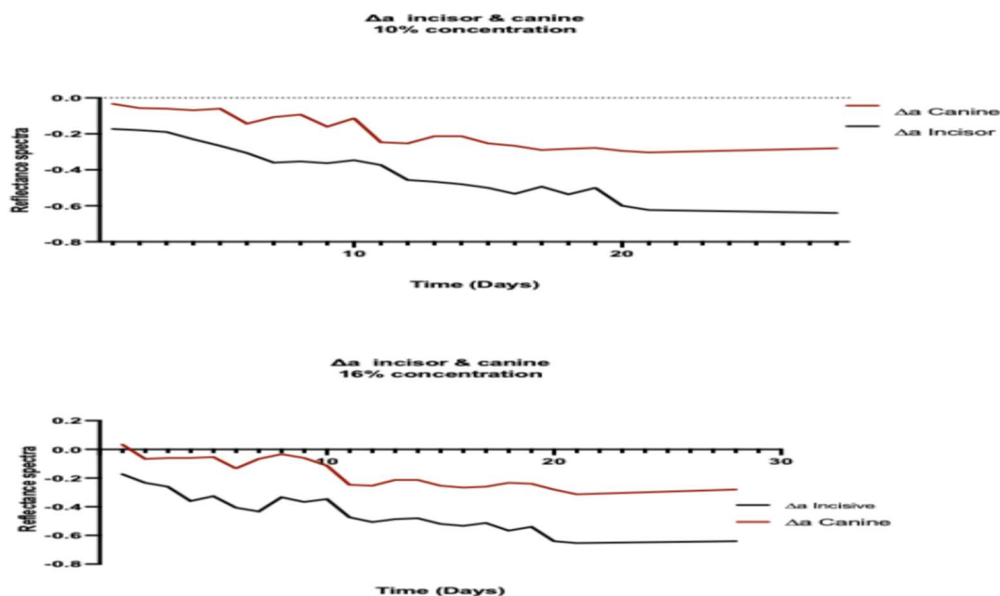


Figure 3: Comparison of Δa , values between incisors and canines at 10% and 16% concentrations

Figure 4 shows the Yellow-Blue axis parameter (Δb), with graphical analysis indicating that the value of Δb reaches a plateau from day 20th. Data analysis shows that the plateau phase is similar for incisors and canines. There is a statistically significant difference

($P < 0.001$) between canines and incisors in terms of the Δb variable in 10% concentration. However, there is no statistically significant difference between canines and incisors in terms of the Δb variable in 16% concentration.

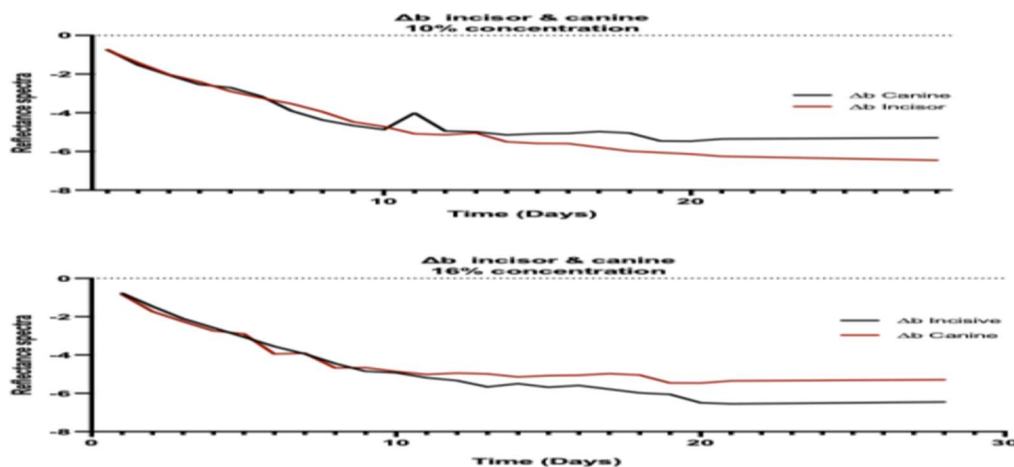


Figure 4: Comparison of Δb values between incisors and canines at 10% and 16% concentrations

DISCUSSION

In our study, we selected carbamide peroxide as the bleaching agent. This choice was based on its prevalence in clinical applications, particularly in home tooth whitening treatments by dentists.10

Carbamide peroxide is commonly used in clinical and at-home whitening due to its stability, ease of application, and sustained action. Upon application, it gradually breaks down to release hydrogen peroxide, ensuring a slower and more controlled

exposure to the tooth structure. This prolonged-release contributes to progressive whitening effects, making it especially suitable for long-term use.^{11,12}

The optimal duration of application of a carbamide peroxide-based tooth whitening product must be carefully determined to maximize treatment efficacy while minimizing potential side effects. The effectiveness and outcome of tooth whitening treatments depend on several factors, such as the concentration of carbamide peroxide, the patient's desired aesthetic results, pre-existing tooth sensitivity, and how the individual responds to the treatment.^{13,14}

For common concentrations of 10% to 16%, clinical protocols generally recommend applications lasting from 30 minutes to several hours a day; these recommendations vary depending on the specific formulation and the supervision of a dental professional. Night-time applications are often suggested for concentrations of 10%, as the slower release of hydrogen peroxide allows for prolonged action without immediate overdosing.^{4,15,16}

Interest in tooth whitening has risen significantly in recent years, driven by increased awareness of dental aesthetics and a growing desire to improve personal appearance. This trend is fueled by technological advances in dental treatments and an ever-increasing presence of bright smiles in the media and advertising, prompting more and more people to seek solutions to achieve a whiter smile.^{12,17} This increase in demand, however, poses new challenges and raises important questions about the safety, efficacy, and long-term implications of these practices, necessitating a thorough exploration of the tooth-whitening products available on the market.

Tooth whitening procedures, although effective in improving the smile's aesthetics, should be viewed with caution because of their potential adverse effects. There is scientific evidence that these treatments can cause tooth sensitivity due to the penetration of bleaching agents into dental microstructures such as dentinal tubules.¹⁸ In addition, they can irritate periodontal tissues, particularly the gingiva, which can exacerbate or initiate inflammatory responses in the periodontium. These phenomena could compromise the teeth' structural integrity and the periodontium's overall health.¹⁹ It is, therefore, essential to balance the

desired aesthetic effects and the potential undesirable effects of tooth whitening.²⁰

The plateau phase during the tooth whitening procedure is defined by a stabilization in the level of tooth shade change, where no substantial improvement is observed despite continued treatment. This phase is critical, indicating the whitening agents' effectiveness limit. The whitening plateau phase occurs because the bleaching agents reach a saturation point where most of the stain molecules or chromogens within the enamel and dentin have been oxidized. At this stage, further applications do not result in a significant additional breakdown of pigments, as the remaining colour-causing molecules are either resistant to bleaching or have already been removed. This saturation limits the effectiveness of prolonged treatment, making continued bleaching less beneficial and potentially harmful.²¹

When the bleaching agent is in contact with dentin, the effects may be more pronounced due to the relatively high organic composition, which makes dentin particularly vulnerable to hydrogen peroxide oxidation. In addition, extending the contact time increases trans-amelodentinal diffusion of the whitening agent. This results in increased penetration of hydrogen peroxide through the enamel and into the underlying dentin, where the intensity and depth of the oxidative action can intensify structural changes and increase the risk of post-treatment sensitivity.^{22,23}

The results of our study indicate that the plateau stage of the whitening process was reached uniformly on the twentieth day of treatment, regardless of the type of tooth considered. A comparison of the conclusions of this study with those of existing research was limited due to a dearth of relevant literature. Currently, there is a need for more specific in vitro data on the plateau effect before it can be translated safely into in vivo clinical applications.

In this study, the statistically significant difference in the ΔE value observed between canines and incisors at the 10% concentration may be due to the different reactivity of enamel and dentin to the bleaching agent. While enamel thickness and dentin composition play a role in the performance of bleaching agents, it is not solely due to overall thickness or density. The statistically significant difference in shade change between incisors and

canines at the 10% concentration may be due to the limited bleaching efficacy of lower carbamide peroxide levels on the more heavily pigmented and thicker dentine of canines. At this concentration, the bleaching agent may effectively lighten the enamel and superficial stains on incisors, which have thinner dentine but may be less able to penetrate or oxidize the deeper, more concentrated intrinsic stains within the thicker dentine of canines.^{24,25} Therefore, the difference in tissue structure and stain depth becomes more apparent at this lower concentration, leading to the observed statistical difference. At a higher concentration of 16%, the bleaching agent's increased oxidative power likely penetrates both incisors and canines more uniformly, reducing the variation in bleaching outcomes and leading to no statistically significant difference.

Our findings showed a similar whitening effect for 10% and 16% carbamide peroxide over the same treatment duration. This aligns with previous studies reporting that lower concentrations can achieve comparable whitening outcomes to higher concentrations when applied over sufficient time.^{14,26} Since higher concentrations are typically associated with increased tooth sensitivity and risk of gingival irritation, a 10% formulation offers a safer and more comfortable alternative—particularly at home bleaching protocols. Therefore, under professional supervision, 10% carbamide peroxide can be recommended as an effective and patient-friendly option that minimizes adverse effects while achieving similar whitening results.²⁷

Multiple clinical factors influence the total duration of a teeth whitening treatment, which should be considered to personalize the protocol, optimize efficacy, and minimize risks or discomfort for the patient.²¹ These factors include the initial tooth shade, the rate of whitening response, and the presence or risk of sensitivity. Patients with higher aesthetic demands may sometimes require extended treatment protocols. However, these must always be balanced against the risk of side effects such as sensitivity or tissue irritation.²⁸ Secondly, the patient's dental sensitivity is a determining factor. Patients with increased sensitivity may require adjustments in the concentration of peroxide or the duration of application to reduce discomfort during whitening. Our results showed that 10% and 16% carbamide peroxide gels reached a whitening plateau at approximately day 20. This finding

suggests that in patients with sensitivity concerns, a less concentrated gel, such as 10% can be effectively used, as it achieves similar whitening outcomes within the same time frame, reducing the risk of sensitivity without compromising results. Thirdly, tooth discolourations, such as those caused by tetracycline or amelogenesis imperfecta, present particular challenges. Tetracycline-induced discolourations, for example, are often deeply embedded in the tooth structure and may require more prolonged or more intensive treatment protocols to achieve satisfactory results. Similarly, amelogenesis imperfecta can significantly affect the response to bleaching, requiring specific treatment adaptations.²⁹

In-home tooth whitening treatments that reach the plateau phase represent, in most cases, an optimal interval for achieving a satisfactory aesthetic result. However, the dental literature needs more data regarding tooth shade anomalies.

CONCLUSION

Dental bleaching is far from a harmless therapeutic procedure. It requires a personalized and meticulous approach, as the process involves chemical changes to the tooth enamel and must be prescribed or supervised by dental professionals. Although popular, such as smile bars, they generally lack the medical expertise to manage such treatments' complexities and potential side effects.

Our findings showed that the bleaching effect plateaued around day 20 regardless of whether 10% or 16% carbamide peroxide was used. Extending treatment beyond this point may offer no additional whitening benefit and could increase the risk of sensitivity. Therefore, we recommend limiting at-home bleaching treatments to a 20-day duration, even with lower concentrations, to achieve optimal results while minimizing adverse effects.

Identifying this phase helps determine the limits of treatment efficacy, which is crucial for guiding clinical practices and ensuring patient safety. Future research should focus on establishing safer and more effective protocols that account for the results obtained at varying concentrations.

Clinical Significance: Tooth whitening has risks and requires an individualized approach. This study highlights the different responses of tooth types, especially at lower concentrations. The consistent plateau by day 20 suggests limited benefit in extending treatment beyond this point.

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Data availability: The data obtained and analyzed during the current study are available from the corresponding author upon reasonable request.

Source of Support

Nil

Conflict of interest

None declared

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