Study Protocol

Efficacy of Tooth Bleaching with 35% and 6% Hydrogen Peroxide in Primary Dentition: Study Protocol for a Randomized Controlled Clinical Trial

ABSTRACT

Background: The aesthetics of dental elements generates significant psychological and social impacts, both in primary and permanent dentition. One of the most employed techniques to visually improve the appearance of teeth is bleaching. In in-office whitening, the technique used consists of applying whitening gel to dental surfaces in concentrations ranging from 25% to 50% of hydrogen peroxide. However, recent studies showed that low and medium concentration bleaching agents based on hydrogen peroxide have been effective and present a lower risk of sensitivity to the patient. **Aims**: To carry out a controlled and randomized clinical trial to compare the effectiveness of dental bleaching in primary teeth with hydrogen peroxide at concentrations of 35% and 6%.**Methods:** In this study, 38 patients aged 3 to 6 years-old will be selected and allocated in 2 groups (G1 - Hydrogen Peroxide 35%, n=19 and G2- Hydrogen Peroxide 6%, n=19). Bleaching will be carried out in up to three sessions, with an interval of 7 days between each session. Evaluation of color and tooth sensitivity will be carried out 48 hours after each bleaching session. The color assessment will be measured with the aid of a digital spectrometer while the tooth sensitivity will be assessed by using the Pain Level Scale (Wong-Baker Faces®).**Discussion:** It is expected that there will be no significant difference between the groups in terms of color variation and that the low concentration bleaching group will have the lowest sensitivity index.

Keywords: Tooth Bleaching,Tooth Bleaching Agents, Hydrogen Peroxide, Primary Dentition, Tooth Deciduous.

1. INTRODUCTION

The aesthetic appearance of upper incisors, both in primary and permanent dentitions, may influence the smile presentation. In the literature, studies report the social and psychological influence of the smile and its impact in the quality of life[1]. In primary teeth, aesthetic impairment is associated with trauma, pulpal disease, carious lesions, developmental disorders, fluorosis, opacity and endodontic treatment performed with iodoform-based filling materials [1,2].

Dental stains are classified as extrinsic and intrinsic [3–5]. Intrinsic stains represent a major challenge in Pediatric Dentistry[1] since the penetration of the chromogenic agent through the dentinal tubules usually occurs in pre-eruptive phases and affects the majority of erupted primary teeth. This type of staining may result from systemic diseases (for instance, bilirubinemia) or ingestion of certain substances, such as antibiotics (tetracycline) or excess fluoridation[6]. In clinical practice, such pigmentations are difficult to manage since their elimination or masking may not be as aesthetically effective. The management of intrinsic dental stains must be performed according to its pulpal condition: internal bleaching for non-vital teeth and external bleaching for vital teeth, or enamel microabrasion and aesthetic rehabilitation through contact lenses [1,5].

Extrinsic staining, on the other hand, can be efficiently handled in the dental practice, with good aesthetic results [1,3,5,7]. Professional dental cleaning with abrasive and bioactive pastes and the employment of tooth-whitening agents are widespread, with good results and aesthetic acceptance.

There are few protocols and randomized clinical trials that have performed whitening in primary teeth; these were either in vitro studies [3–5] or case reports[8–10]. The authors reported that the most efficient whitening technique in primary teeth, considering the cost-benefit and minimum intervention, is internal bleaching. Its main disadvantage is that the tooth must be devitalized.

Tooth whitening is a simple, conservative, and low-cost treatment. Although this is not a recent procedure (it has been carried out for about 150 years), the current technique was introduced in 1984, with the aim to improve of the aesthetics of natural teeth[11]. Whitening methods include take-home kits and professional whitening in the dental practice. Both techniques are effective; however, side effects have been reported, thus reinforcing the importance of supervision by oral health professionals in order to achieve a safe and successful tooth whitening result [5,12].

According to the American Academy of Pediatric Dentistry, there has been an increase in children and adolescents that are interested in bleaching their teeth. Therefore, they have developed a policy on the use of dental bleaching for this public. According to this policy, tooth whitening procedures have demonstrated to be safe and can be beneficial for children and adolescents. It was verified that the use of bleaching agents can improve dental aesthetics and increase self-esteem and the correct indication and planning are essential before starting any bleaching protocol. The use of bleaching agents should always follow safety and efficacy standards defined by clinical research and bleaching on young patients should be supervised by an adult and under the guidance of a dentist[13].

A systematic review inferred that products with low and medium concentration of hydrogen peroxide for in-office bleaching have a lower risk for the patient and a lower amount of sensitivity after bleaching than high concentration hydrogen peroxide products, with similar effectiveness in terms of whitening. Concluding that the use of low-concentration hydrogen peroxide products can produce the same effectiveness in color change, with the advantage of the lower risk and intensity of sensitivity arising from whitening[14].

The in-office dental bleaching technique consists of applying a bleaching gel in concentrations ranging from 25% to 50% of hydrogen peroxide and carbamide peroxide, with or without a light source, controlled by the dentist. There are different lights that are used by the professional in this procedure, in an attempt to reduce the application time during the treatment; studies report that the use of lights does not change the final result of the treatment[15].

Based on these assertions, this project proposes to carry out a clinical study to evaluate the effectiveness of dental bleaching treatment in primary teeth by using in-office bleaching technique and assess two different concentrations (high and low) of hydrogen peroxide to suggest the most appropriate whitening protocol for deciduous teeth.

2. MATERIAL AND METHODS

2.1 Study design

This study protocol was designed as a randomized, controlled, blind, clinical trial according to the 2013 Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) Statement (Table 1) and the SPIRIT checklist can be found as an additional file.It will be conducted at the dental clinic of UniversidadeMetropolitana de Santos (UNIMES).

Table 1. Schedule of enrolment, interventions, and assessments of the study

	STUDY PERIOD											
	Enrolment	**Allocation**	**Post-allocation**									**Close-out**
TIMEPOINT*	***-t1***	**0**	***T1***	***T2***	***T3***	***T4***	***T5***	***T6***	***T7***	***T8***	***T9***	***T10***
ENROLMENT:												
Eligibility screen	X											

Informed consent	X											
Allocation		X										
INTERVENTIONS:												
[Bleaching 35% hydrogen peroxide gel]			X			X						
[Bleaching 6% hydrogen peroxide gel]			X			X						
ASSESSMENTS:												
[Clinical evaluation]		X	X	X	X	X	X	X	X	X	X	X
[First application]			X									
[Second application]												
[Third Application]									X			
[Wong-Baker FACES® Scale]		X		X	X		X	X		X	X	X
[Assessment of color]		X		X	X		X	X		X	X	X

* -t1 = before the Baseline, 0 = Baseline, T1 = 1ª application, T2 = immediately after 1ª application, T3 = 48 hours after 1ª application, T4 = 2ª application, T5 = immediately after 2ª application, T6 = 48 hours after 2ª application, T7 = 3ª application, T8 = immediately after 1ª application, T9= 48 hours after 1ª application, T10 = 4 weeks after the 1ª application.

2.2 Ethical Considerations

The study will be conducted in accordance with the ethical precepts determined in the Declaration of Helsinki (World Medical Association Declaration of Helsinki, 2008). The protocol for this study was approved by the Human Research Ethics Committee of UniversidadeMetropolitana de Santos - UNIMES (certificate number: 68431023.5.0000.5509/ 6.019.284 - Approval date: April 24, 2023). All information will be detailed and specified in the statement of informed consent, in accordance with Resolution 196 of the National Board of Health (Health Ministry, Federal District, Brazil, March 10, 1996). This is the first version of a protocol that was registered in ClinicalTrials.gov, under the registration number: NCT05789004, first posted on March 29, 2023, and last updated on October 31, 2024.

The legal guardians of the children will agree to participate by signing a statement of informed consent; two copies will be signed – one for the legal guardian of the child and one for the researchers. The participants will be informed that they may withdraw from the study at any time for any reason if they so desire. The researchers may also remove participants from the study, if deemed necessary.

2.3 Study Population

Thirty-eight teeth will be selected from healthy male and female children ranging from three to six years-old (no distinction in terms of ethnicity) enrolled for treatment at the pediatric dental clinic of UniversidadeMetropolitana de Santos (UNIMES). At the first appointment, a form addressing the medical history of the patient will be completed. Next, the volunteers will be submitted to a clinical examination to assess their oral health status. Based on the information collected during this first appointment, the inclusion and exclusion criteria will be applied. Patients who meet the inclusion criteria and whose parents or guardians sign the informed consent form will be scheduled for a second appointment for treatment according to the allocation group.

2.3.1 Inclusion criteria:

- Healthy children with no adverse systemic conditions;
- Age range: from 3 to 6 years-old;
- Vital teeth with severe to moderate discoloration;
- No active carious lesions;
- No lesions in the oral cavity;
- No report of previous tooth sensitivity.

2.3.2 Exclusion criteria:
Children with systemic diseases, Children who, during anamnesis, report allergies to dyes or latex, who withdraw from participating in the study, as well as those who do not attend the callbacks, will be excluded.

2.4 Sample size

To carry out the sample calculation, we assumed the study as non-inferiority design, with delta of 8.24 +-2.45 for color change in the control group [16]. The limit of non-inferiority was considered as -2.5 inferiority, power of 90% and significance level of 5%, which resulted in 17 teeth per group. 10% was added to this number due to possible sample losses, resulting in a sample of 19 primary anterior teeth per group, totaling 38 teeth.

2.5 Randomization and allocation

The type of treatment will be randomly determined for each tooth, through a draw before the intervention. The draw will follow the order electronically generated by the randomizer.org randomization website for a balanced distribution of all teeth between the groups. Thirty-eight vital primary teeth with severe to moderate discoloration will be selected; the teeth will be divided into two groups (Figure 1).

GROUP	Concentration of hydrogen peroxide	Chemical Composition	Material	Comercial Brand
G1 (N=19)	High Concentration	35% Hydrogen peroxide	Whiteness HP Automixx 35%	FGM Dental Group – Joinville-SC – Brazil
G2 (N=19)	Low Concentration	6% Hydrogen peroxide	Whiteness HP Automixx 6%	FGM Dental Group – Joinville-SC – Brazil

Figure 1. Distribution of Experimental Groups.

2.6 Interventions

Before starting the procedures, the color of the darkened tooth will be confirmed using the Vita Easyshade® digital spectrometer (Vita Easyshade®; VITA Zahnfabrik H. Rauter GmbH & Co., KG, Germany). The upper left canine will be used as a color reference because it is the most saturated tooth in the arch (greater dentin mass and greater amount of intrinsic pigments).

Group 1 - Hydrogen Peroxide 35%

In group 1, the following bleaching protocol will be performed:

1. Clinical examination and initial shade color assessment.

2. Sensitivity/pain recording using the Pain Level Scale (Wong-Baker Faces®)

3. Prophylaxis with a Robson brush (Color-Brush – Ultra-Soft; American Burns, Palhoça, SC, Brazil) and prophylactic paste (CleanJoy; Voco GmbH, Cuxhaven, Germany) to remove extrinsic pigments and bacterial plaque.

4. Application of lip balm (F&A, SP, Brazil) on the patient's lips;

5. Placement of the labial retractor (ArchFlex; FGM Dental Group, Joinville, SC, Brazil);

6. Application of the gingival barrier (Top Dam; FGM, Joinville, SC, Brazil) on the tissue free marginal gingival margin and the papillae between the upper right canine teeth and the upper left canine teeth. The gingival barrier material will be photopolymerized from 20 to 30 seconds using high power LEDs (Radii-Xpert; SDI Limited, Victoria, Australia);

7. Application of Whiteness HP Automixx 35% whitening gel (Whiteness HP Automixx 35%; FGM Dental Group, Joinville, SC, Brazil)

8. In the session, a single application of the gel will be carried out. It will remain in contact with the tooth for 15 min. With the aid of a micro-applicator (Cavibrush; FGM Dental Group, Joinville, SC, Brazil), the gel will be moved over the teeth frequently (every 5 minutes) to release oxygen bubbles and improve the contact between the gel and the teeth.

9. The gel must be removed from the teeth with a dental suction device and the teeth cleaned with gauze.

10. Sensitivity/pain recording using the Pain Level Scale (Wong-Baker Faces®)

11. The second bleaching session will be carried out 7 days after the first one and, if necessary, a third session can be carried out 7 days after the second one.

Group 2 - Hydrogen Peroxide 6%

In group 1, the following bleaching protocol will be performed:

1. Clinical examination and initial shade color assessment;

2. Sensitivity/pain recording using the Pain Level Scale (Wong-Baker Faces®)17

3. Prophylaxis with a Robson brush (Color-Brush – Ultra-Soft; American Burns, Palhoça, SC, Brazil) and prophylactic paste (CleanJoy; Voco GmbH, Cuxhaven, Germany) to remove extrinsic pigments and bacterial plaque.

4. Application of lip balm (F&A, SP, Brazil) on the patient's lips;

5. Placement of the labial retractor (ArchFlex; FGM Dental Group, Joinville, Brazil);

6. Application of the gingival barrier (Top Dam; FGM, Joinville, SC, Brazil) on the tissue free marginal gingival margin and the papillae between the upper right canine teeth and the upper left canine teeth. The gingival barrier material will be photopolymerized from 20 to 30 seconds using high power LEDs (Radii-Xpert; SDI Limited, Victoria, Australia);

7. Application of Whiteness HP Automixx 6% whitening gel (Whiteness HP Automixx 6%; FGM Dental Group, Joinville, SC, Brazil)

8. In the session, a single application of the gel will be carried out. It will remain in contact with the tooth for 15 min. With the aid of a micro-applicator (Cavibrush; FGM Dental Group, Joinville, SC, Brazil), the gel will be moved over the teeth frequently (every 5 minutes) to release oxygen bubbles and improve the contact between the gel and the teeth.

9. The gel must be removed from the teeth with a dental suction device and the teeth cleaned with gauze.

10. Sensitivity/pain recording using the Pain Level Scale (Wong-Baker Faces®)

11. The second bleaching session will be carried out 7 days after the first one and, if necessary, a third session can be carried out 7 days after the second one.

2.7Outcome assessment

Clinical evaluations will assess tooth color and sensitivity.

2.7.1Assessment of color

The initial color will be determined by using Vita Easyshade® digital spectrometer (Vita Easyshade®; VITA Zahnfabrik H. Rauter GmbH & Co., KG, Germany) before the rubber cup polishing at the first bleaching session. The spectophotomer measurement will be performed again at 48 hours after the first bleaching section, one week after the end of bleaching and 4 weeks after the beginning of the bleaching treatment.

The tooth shade will be determined using the parameters of the spectrophotometer device, according to the study of Pierote et al 2020[12]. This study assessed the following values: L*, a*, and b*. L* parameter represents the tooth value on a scale from 0 (black) to 100 (white), a* is the measure along the red (a* positive) and green (a* negative) axes and b* is the measure along the yellow (b* positive) and blue (b* negative) axes. The color will be estimated by the equation ΔE = ((ΔL*)2+(Δa*)2+(Δb*)2)1/2. This assessment will be performed to verify the effectiveness of the bleaching treatment.

2.7.2Assessment of of tooth sensitivity

Tooth sensitivity assessment will be performed with the aid of Wong-Baker FACES® Pain Rating Scale (Figure 2). This scale was created to help children communicate about their pain. Nowadays, the scale is used for 3-year-old children and above. The assessment will be performed according to the instructions provided by the Wong-Baker FACES Foundation (www.WongBakerFACES.org), as follows. The professional will explain that each face represents a person with no pain, or some pain, or a lot of pain. Face 0 doesn't hurt at all. Face 2 hurts just a little bit. Face 4 hurts a little bit more. Face 6 hurts even more. Face 8 hurt a whole lot. Face 10 hurts as much as you can imagine, although you don't have to be crying to have this worst pain. After the explanation, the patient will be asked to choose the face that best depicts the pain that they are experiencing [17].

Figure 2 Wong-Baker FACES® Pain Rating Scale

In this study, tooth sensitivity will be assessed before the first bleaching session, immediately after the bleaching section, 48 hours after the end of each bleaching section, one week after the end of the bleaching treatment and 4 weeks after the beginning of the bleaching treatment.

2.8Statistical Analysis

Descriptive analyses will consist of calculations of mean and standard deviation. According to previous studies[12], pain sensitivity data can be subjected to multivariate analysis of variance (MANOVA) with repeated measures and Wilks' Lambda test ($p<0.05$). For shade variation analysis, a completely random analysis of variance will be applied ($p<0.05$).

3. DISCUSSION

There are still no bleaching protocols for primary teeth described in the literature. We believe that there will be no difference in the final whitening result when comparing the two groups. We expect that lower levels of sensitivity will be observed in the group where the whitener with a low concentration of hydrogen peroxide was used, as the product has an

alkaline and stable pH. This whitening agent also has excellent thixotropy and allows the optional use of a gingival barrier.

These characteristics suggest that this product can be a good choice for primary teeth bleaching.

CONSENT AND ETHICAL APPROVAL

Human Research Ethics Committee of UniversidadeMetropolitana de Santos - UNIMES(certificate number: 68431023.5.0000.5509/ 6.019.284 and was registered with the Clinical Trials (https://clinicaltrials.gov/) under number NCT05789004. The guardians of the children agreed to participate by signing a statement of informed consent; two copies of which were signed: one for the guardian and one for the researchers.

Disclaimer (Artificial intelligence)

The authors hereby declare that NO generative AI technologies, such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators, were used during the writing or editing of this manuscript.

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