Original Article

Phytochemicals in vital tooth bleaching: Spectrophotometric evaluation of efficacy with papaya, pineapple, or kiwi extracts and 30% hydrogen peroxide

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Abstract

Context: Dental bleaching, a common cosmetic treatment, typically uses hydrogen peroxide (H_2O_2). Yet, the search for natural options has prompted an investigation into fruit extract efficacies such as papain, bromelain, and actinidin for tooth whitening.

Aim: The aim of this study was to evaluate the efficacy of 30% hydrogen peroxide and fruit extracts (papaya, pineapple, and kiwi) on human enamel using a spectrophotometer at different time intervals.

Study Design: Eighty maxillary anterior teeth were stained with tea solution and evaluated for baseline color. They were then divided into four groups: hydrogen peroxide alone and hydrogen peroxide combined with papaya, pineapple, or kiwi extracts. Each group was further divided based on bleaching duration: 10 or 20 min.

Materials and Methods: The color value of the bleached teeth was measured using a reflectance spectrophotometer. In order to analyze the data, one-way ANOVA, *post hoc* Tukey, and paired *t*-tests were used. The significance level was established at $\alpha = 0.05$.

Results: Combining hydrogen peroxide with pineapple extract showed the highest efficacy, followed by papaya and kiwi extracts. Hydrogen peroxide alone also demonstrated significant bleaching efficacy, albeit lower than the combinations with fruit extracts.

Conclusion: Combining hydrogen peroxide with pineapple and papaya extracts notably improves dental bleaching efficacy, as shown by reduced color difference (ΔE) values. This underscores the potential of natural enzymes in tooth whitening.

Keywords: Bromelain enzyme; hydrogen peroxide; papain enzyme; reflectance spectrophotometer; tooth bleaching

INTRODUCTION

In modern dentistry, bleaching is favored for minimal invasiveness and superior esthetics over veneers and crowns.[1] Tooth discoloration, caused by extrinsic stains on the enamel, is treatable with prophylactic procedures and bleaching.[2] Dental bleaching, using hydrogen peroxide, has been popular since its introduction for at-home whitening in 1989.[3] Today, carbamide peroxide and H_2O_2 are common, with 10% carbamide gel yielding 3.5% H_2O_2. H_2O_2, up to 40% in concentration, releases oxygen species that whiten teeth by oxidizing chromophores in enamel and dentin.[4] Tooth sensitivity, affecting 15%–78% of individuals undergoing tooth bleaching,

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is often attributed to minor enamel damage, enabling reactive oxygen species to access dental nerves.[5] Hydrogen peroxide, while effective in tooth bleaching, induces unavoidable clinical side effects, altering enamel surface texture, composition, and microhardness. Integrating vegetative enzymes with hydrogen peroxide lowers its concentration while accelerating and enhancing the bleaching process.[6] Combining it with plant enzymes such as papain and bromelain aims to enhance bleaching efficacy while minimizing side effects.[7,8] Kiwifruit enzymes show promising potential due to their broader pH range and enhanced hydrolysis capability.[9,10]

MATERIALS AND METHODS

The Institutional Ethical Committee granted approval for the current study under reference (IEC/2022/39).

The sample size of 80 was determined to achieve a statistical power of approximately 0.80, with a significance level (alpha) set at 0.05, based on a medium effect size ($d = 0.50$) between groups. This calculation was performed using SPSS Version 20.0. (Armonk, NY: IBM Corp.). The null hypothesis (H_0) suggested that there would be no variance in the tooth whitening effectiveness among hydrogen peroxide and its combination with papaya, pineapple, and kiwi extracts during vital bleaching.

Specimen preparation

Eighty permanent maxillary anterior teeth were obtained, following extraction due to periodontal disease. Teeth showing noticeable cracks, caries defects, decalcifications, cervical lesions unrelated to caries, resorption, or formative abnormalities were avoided [Figure 1a].

The remaining teeth were cleaned to remove calculus and tissue debris using an ultrasonic scaler. Once that was done, samples were preserved in a 0.2% thymol solution and refrigeration was maintained at 4°C until needed.

Group distribution

- Group 1: 30% hydrogen peroxide
- Group 2: 30% hydrogen peroxide + papaya extract
- Group 3: 30% hydrogen peroxide + pineapple extract
- Group 4: 30% hydrogen peroxide + kiwi extract.

Then, each group was further subdivided into two subgroups of 10 teeth each:

- Subgroup ($n = 10$) based on time duration
  - Subgroup A: 10 min
  - Subgroup B: 20 min.

Preparation of papaya extract

The unripe papaya was cleaned with distilled water, stripped, and deseeded. Its tissue was mixed and stressed to separate the unripe pulp juice (UPJ). After centrifuging the UPJ at 2000 rotation per minute (rpm) for 15 min at 4°C, it was filtered using a 0.22 µm syringe filter to obtain sterilized UPJ, ready for immediate use.

Pineapple extract preparation

Two hundred grams of pineapple were stripped, diced, and squashed before being mixed with 25 ml of deionized water. The subsequent combination was then exposed to centrifugation at 2000 rpm for 2 min at 4°C. Following this, the clarified liquid was filtered and stored in a refrigerator at 4°C until required.

Preparation of kiwi extract

A mixture comprising 50 g of kiwi fruit pulp and 175 ml of a solution containing 6% sodium chloride and 2% boric acid at a ratio of 1:4 (weight:volume) was prepared. Following this, the cooled centrifuge was meticulously agitated at a speed of 6000 rpm and subsequently filtered. The resulting crude extract was then collected and stored until required for utilization.

Staining of specimen

A tea bag was steeped in 100 ml of boiling deionized water, which was then cooled to room temperature and filtered after a 5-min infusion. The teeth were immersed in this solution for a full day for staining, cleaned afterward, and maintained at 100% humidity and 37°C.

Baseline color value

Twenty stained specimens were chosen at random to

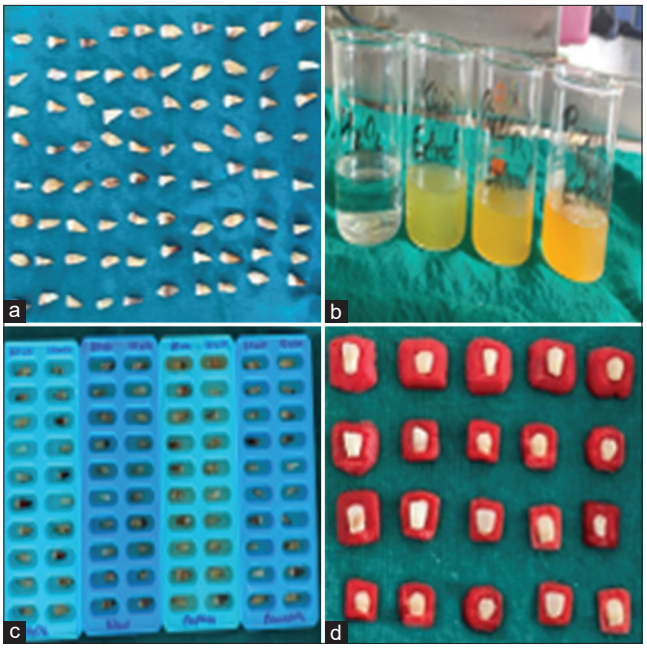

Figure 1: (a) Extracted permanent maxillary anterior, (b) Prepared bleaching solution containing hydrogen peroxide with extracts, (c) Teeth immersed in prepared bleaching solutions, (d) Teeth mounted in acrylic resin exposing labial surface

establish baseline color values. The random allocation sequence was done using a manual method where numbered slips of paper corresponding to each tooth specimen were randomly drawn from a container. These slips were then placed into sequentially numbered containers representing each treatment group and sealed for concealment until assignment. Randomization procedures were carried out by the study team, and teeth specimens were enrolled accordingly. Blinding after assignment to intervention was not feasible in this study.

The specimens' reflectance was measured using a spectrophotometer against a white backdrop. Each tooth was then dissected at the cementoenamel junction, and the labial surface was exposed by mounting with acrylic resin [Figure 1d].

Whitening protocol

The bleaching mixture consisted of a combination of 1 ml of either pineapple, papaya, or kiwi extract with 1 ml of phosphate-buffered solution and 28 ml of hydrogen peroxide [Figure 1b]. Group 1 was treated exclusively with hydrogen peroxide, while Groups 2, 3, and 4 were bleached in a mix with papaya, pineapple, and kiwi separately, respectively. The specimens underwent immersion for either 10 min (Subgroup A) or 20 min (Subgroup B) [Figure 1c]. After the bleaching process and rinsing, the teeth were placed in synthetic saliva at 37°C for a duration of 24 hours.

Spectrophotometric analysis for color assessment

The analysis used a reflectance spectrophotometer against a white backdrop. Measuring color parameters L^*, a^*, b^* was based on the Commission Internationale de l'Eclairage (CIE) Lab color system, which captures color variations. The formula utilized in this system to calculate color differences is: $\Delta E = ([\Delta L^*] 2+ [\Delta a^*] 2+ [\Delta b^*] 2)^{1/2}$.

Statistical analysis

Data analysis employed SPSS Version 20.0. (Armonk, NY: IBM Corp.), utilizing a significance level of $P \leq 0.05$, and statistical tests included paired *t*-tests, one-way ANOVA, and *Post hoc* Tukey's tests. Paired *t*-tests compared bleaching efficacy within each group over 10 and 20 min. One-way ANOVA assessed differences in efficacy among groups at each time. *Post hoc* Tukey's tests identified groups with significant variations in efficacy.

RESULTS

Table 1 depicts significant differences in bleaching efficacy observed at 10 and 20 min among groups ($P < 0.001$). Hydrogen peroxide + pineapple extract showed the highest efficacy. At 10 min, hydrogen peroxide + papaya extract improved efficacy ($P = 0.013$), while hydrogen peroxide + kiwi extract did not ($P = 0.996$). No notable distinction was seen between hydrogen peroxide and hydrogen peroxide + papaya extract at 20 min ($P = 0.841$). Significant differences were found between hydrogen peroxide + kiwi extract and other combinations at 20 min ($P = 0.023$). Overall, hydrogen peroxide + pineapple extract had the highest efficacy, followed by hydrogen peroxide + papaya extract, while hydrogen peroxide alone or with kiwi extract showed lower efficacy.

DISCUSSION

Growing awareness drives demand for esthetic dental treatments. Dental appearance, including tooth color, shape, and position, significantly influences facial attractiveness and social interactions. Dental bleaching is popular in cosmetic dentistry due to increasing esthetic concerns. While hydrogen peroxide remains a key agent, alternative methods offer quicker and more affordable solutions.[11] Over-the-counter products such as toothpaste, flushes, gums, paint-on stains, and strips fluctuate in viability, often targeting extrinsic stains rather than true bleaching, raising concerns about potential damage to teeth and gums.[12] Professional tooth bleaching effectively addresses extrinsic and intrinsic stains, considering factors such as diet, tobacco, medication, and trauma. Dental experts tailor techniques to individual requirements and oral health for optimal results.[12]

Hydrogen peroxide, a common dental bleaching agent, reacts vigorously due to its hydrogen and oxygen content. It oxidizes chromogens in enamel, reducing pigmentation for whitening. Its effectiveness is boosted with heat or light, activating the peroxide for a stronger whitening effect.[13] Improper use may cause sensitivity and root

Table 1: Bleaching efficacy (ΔE), standard deviation, and *P* values at 10–20 min

Time period (min)	Group	Mean bleaching efficacy (ΔE)±SD	*P* (overall)	*P* (group comparison)
10	H_2O_2	21.99±1.80	<0.01	-
	H_2O_2 + pineapple extracts	26.08±1.47	-	<0.001*
	H_2O_2 + papaya extracts	24.04±1.26	-	0.013
	H_2O_2 + kiwi extracts	22.14±1.18	-	0.996**
20	H_2O_2	25.78±2.47	<0.001	-
	H_2O_2 + pineapple extracts	32.63±2.64	-	<0.001*
	H_2O_2 + papaya extracts	26.48±1.09	-	0.841**
	H_2O_2 + kiwi extracts	23.20±0.83	-	0.023

Level of Significance $P \leq 0.05$, *Significant, **Non Significant. H_2O_2: Hydrogen peroxide, SD: Standard deviation

resorption, reaching the pulp and triggering inflammation. In high concentrations, it can irritate gums. Caution is advised, especially considering its potential adverse effects on dental tissues.[14]

Consumer preference for animal-free and environmentally sustainable products drives the demand for natural options in teeth whitening. Studies explore plant-based enzymes such as papain and bromelain in oral care products, aiming to degrade pellicles and enhance stain removal as alternatives to peroxide bleaching agents.[15] The study utilized bromelain, papain, and actinidin from pineapple, papaya, and kiwi, respectively, extracted following established protocols. The enzymes, along with hydrogen peroxide, were utilized in two separate bleaching sessions: one lasting 10 min and the other 20 min. Results were assessed using a reflectance spectrophotometer, chosen for its accuracy and reproducibility. A spectrophotometer measures how much light is reflected from a surface at different wavelengths. Unlike colorimeters, spectrophotometers can precisely measure and compare tooth color using systems like CIE lab, detecting subtle changes crucial for evaluating teeth whitening efficacy.[16]

Bromelain, extracted from pineapples, shows promise as a tooth-bleaching agent. Studies by Chaurasiya and Hebbar,[17] Corzo *et al*.,[18] Vejai Vekaash *et al*.,[6] Epple *et al*.,[19] Varilla *et al*.,[20] and Sheshadri *et al*.[21] highlight its efficacy with hydrogen peroxide, suggesting its potential for safe and effective dental whitening applications. The study's findings support Bromelain's effectiveness in teeth bleaching. Group 3 (hydrogen peroxide + pineapple extract) showed notable efficacy, with mean ΔE values of 26.08 ± 1.47 at 10 min and 32.63 ± 2.64 at 20 min, surpassing other groups and aligning with existing research.

Group 2 (hydrogen peroxide + papaya extract) in our study, featuring papain, demonstrated significant bleaching efficacy with mean ΔE values of 24.04 ± 1.26 at 10 min and 26.48 ± 1.09 at 20 min. These results align with studies by Kalyana *et al.* (2011)[14] and Chakravarthy and Acharya[22] on papain-containing dentifrices, highlighting their potential in dental bleaching. Gunde and Amnerkar[23] and Yang *et al*.[24] emphasize papain's medicinal properties, while Choudhary[25] and Tadikonda *et al*.[8] confirm its efficacy in enhancing hydrogen peroxide bleaching, suggesting its promising role in revolutionizing teeth whitening methodologies.

Group 4 (hydrogen peroxide + kiwi extract) in the present study, featuring actinidin from kiwifruit, showed significant bleaching efficacy with mean ΔE values of 22.14 ± 1.18 at 10 min and 23.20 ± 0.83 at 20 min. These results align with Richardson *et al*.[9] and Satpal *et al*.[10] on kiwi fruit's antioxidant properties, indicating its potential in dental bleaching.

The present study demonstrates that groups using hydrogen peroxide combined with natural extracts from pineapple, papaya, or kiwi showed significantly higher ΔE values after both 10 and 20 minutes of treatment, compared to groups using hydrogen peroxide alone. This approach not only boosts effectiveness but also potentially reduces tooth sensitivity and gum irritation, appealing to consumers preferring natural dental treatments.

Limitations

Conducted *in vitro*, it may not fully reflect real-world conditions impacted by factors such as saliva and daily activities. Limited ranges of hydrogen peroxide concentrations and extract combinations suggest that further investigation is needed for optimal results. Safety concerns, including tooth sensitivity and gum health, warrant thorough exploration. Results may not universally apply due to enamel variations and oral health differences, highlighting the need for broader research and clinical trials.

Future scope

To address challenges in preparing fresh extracts, innovative preservation methods or standardized formulations could be explored. Collaboration with pharmaceutical or biochemical experts could optimize extract stability and bioavailability for dental use. Future research should conduct diverse, long-term clinical trials to assess treatment effectiveness and safety, exploring varied concentrations and combinations of hydrogen peroxide and natural extracts.

CONCLUSION

The present study pioneers dental research by exploring hydrogen peroxide's efficacy with natural fruit extracts such as papaya, pineapple, and kiwi on human enamel, departing from traditional teeth-whitening methods. Addressing the lack of literature on natural enzymes with bleaching agents inspires further clinical studies. Results reveal significant differences among groups at 10 and 20 min ($P < 0.001$), with hydrogen peroxide combined with pineapple extract showing the highest efficacy, followed by papaya. This suggests the potential of natural extracts, especially pineapple and papaya, in enhancing dental bleaching. The findings propel future research on optimal dosages, long-term effects, and personalized treatments, advancing cosmetic dentistry sustainably.

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Conflicts of interest

There are no conflicts of interest.

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