Evaluating the Effectiveness of Two Whitening Formulations and a Barrier Gel Pen: An Ex vivo Study

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Introduction

The deposition of extrinsic stains on the tooth surface typically results from the consumption of discoloring substances such as chromatogenic food and drinks, and tobacco use. These stains are localized mainly on the enamel surface and are either generated by the reaction between sugars and amino acids or acquired from the retention of exogenous chromophores in the pellicle [1].

In-home tooth whitening treatments were first launched in the United States in the 1990s. For many years, the active ingredients in these formulations were usually hydrogen peroxide or carbamide peroxide compounds [2]. Unfortunately, the side-effects of in-home bleaching may include soft-tissue irritation, tooth sensitivity and dental erosion [3]. For this reason, new non-peroxide alternatives are being sought.

Overall goal of this prospective, blinded, ex vivo study was to evaluate the effects of two non-peroxide formulations on the color of dental enamel in extracted teeth. First, the ability of a protective gel pen to prevent tooth discoloration by coffee and red wine was investigated. Next, the time-resolved effects on tea-stained enamel color of a non-peroxide whitening strip formulation vs a peroxide-based strip were evaluated.
Materials and Methods

Samples and protocol
Evaluation of protective gel pen
Twenty-four extracted teeth that appeared healthy to the naked eye and to inspection under the light microscope (x10) were bisected. After standardized photography and colorimetry, one half of each tooth was painted with two coats of the test whitening pen, allowing for 30 seconds of drying time after each application; the other half remained untreated. Twelve of the bisected sample pairs were immersed in individual 20mL aliquots of freshly brewed Via coffee (Starbucks) according to the instructions on the packet: 1 packet per cup of coffee. The remaining 12 sample pairs were immersed in individual 20mL aliquots of red wine (Charles Chaw Cabernet Sauvignon, Trader Joes). Standardized colorimetry was used to determine tooth color every 10 minutes for a total of 50 minutes. The endpoint was selected once no further color changes were recorded over the preceding 10-minute interval.

Test formulation
Protective pen: Lumineux Bright2 PenR, Lumineux Oral Essentials, Beverly Hills, CA 90210

Evaluation of whitening strips
A commonly-use protocol for staining teeth by means of immersing teeth overnight in a concentrated tea solution was employed [4–7]. All samples were stained by overnight immersion in a concentrated black tea solution produced by steeping 2 teabags (Lipton’sR yellow label black tea) in ½ cup of boiling water for 30 minutes. Each sample pair was stained in a separate vial. After baseline colorimetry measurements in 24 extracted teeth, 12 samples each were randomly allocated to receive standardized bleaching treatment with 3D Crest White Strips or Oral Essentials Whitening Strips. L* and b* color indices were measured at hourly intervals over 5 hours for all whitening strips by a blinded, pre-standardized, experienced investigator under standard lighting and moisture conditions. Standardized photographs were also recorded at each time point.

Test formulations
Positive control whitening strips: Crest 3D Whitestrips, Professional EffectsR, Procter and Gamble, Cincinnati, OH 45201

Test whitening strips: LUMINEUX Whitening StripsR, Lumineux Oral Essentials, Beverly Hills, CA 90210

Color measurements
Tooth color was measured midline in the mid-cervical third of each of the tooth samples at each designated timepoint. L* and b* color measurements were collected 3 times for each location and timepoint by 1 pre-standardized, experienced clinician under standardized lighting, distance, and ambient conditions, using the VITA Easyshade V (VITA North America, Yorba Linda, CA 92887) digital spectrophotometer. L* represents lightness from black to white on a scale of zero to 100, while b* represent chromaticity with no specific numeric limits. Negative b* corresponds with a shift towards the blue and a positive b* corresponds with a shift towards the yellow.
Data analysis
Changes in tooth color were calculated by comparing each L* and b* color measurement at each timepoint with its baseline value. Mean change in color was calculated as percent change from baseline averaged across all teeth and analyzed using repeated measures analysis of variance models. Significance was set at p<0.05.

Results
Protective gel pen
Coffee
The teeth began to stain significantly after 30 minutes exposure to coffee when pre-coated with the test pen, whereas the untreated teeth began to stain significantly after 20 minutes exposure to coffee. Thus, using the pen significantly prevented enamel staining for an additional 10 minutes of coffee exposure vs the uncoated teeth.

Coffee exposure of non-coated and OE coated teeth caused a significant decrease in the L* value (darkening) over time starting at 20 mins when the protective pen was not used, vs. 30 minutes when the pen was used. Before these time points, the changes in color were not significant. In non-coated teeth, L* values became significantly darker vs. baseline at 20 minutes (p< 0.01), 30 minutes (p< 0.01), 40 minutes (p< 0.001), and 50 minutes (p< 0.001) respectively. When the protective pen was used, teeth became significantly darker vs. baseline after 30 minutes (p< 0.05), 40 minutes (p< 0.01), and 50 minutes (p< 0.01) minutes respectively. The changes in L* values (darkening) were minimal and not significant at the 10- and 20-minute time points (p>0.01) using the protective pen and these changes continued to be less than in the uncoated teeth at 30-, 40- and 50-minute time points (Figure 1).

![Figure 1](image)

**Figure 1:** Change in enamel lightness (L* value) (S.D.) over time from concentrated coffee exposure.

B* values did not change significantly for either treatment group throughout the duration of the study (p>0.05 for all time points, both groups) (Figure 2).
Red wine exposure of non-coated teeth caused a significant decrease of L* (darkening) in a time-dependent manner after the first 10-minute exposure interval, with significant progressive darkening at each subsequent time point. The following significance levels were calculated for each timepoint: 10 minutes: p < 0.01, 20 minutes: p < 0.01, 30 minutes: p < 0.01, 40 minutes: p < 0.001, and 50 minutes: p < 0.001. OE-coated teeth began to darken significantly after 30 minutes of exposure to red wine, with progressive darkening at each subsequent time point. The following significance levels were calculated for each timepoint: 10 minutes: p > 0.05, 20 minutes: p > 0.05, 30 minutes: p < 0.01, 40 minutes: p < 0.001, and 50 minutes: p < 0.001 (Figure 3).

Red wine exposure of non-coated and OE-coated teeth did not cause a significant change in b* value at any time point (treatment and time effects: p > 0.05 for all time points) (Figure 4).
Fig. 4: Change in b* value (S.D.) over time from red wine exposure.

Whitening strips
Baseline L* values for the 2 groups did not differ significantly (p>0.5). There was no significant difference between the whitening effect of the two strips over the entire test duration (p>0.5) (Fig. 5).

Fig. 5: Change in enamel lightness (L* value) (S.D.) over time after treatment with test and control whitening strip.

Baseline b* values for the 2 groups did not differ significantly (p>0.5). The effects of the 2 strip formulations did not differ statistically over the entire test period (p>0.5). For all samples, the b* values increased to a small extent over time, indicating a mild shift towards the yellow and away from the blue.
Figure 6: Change in $b^*$ value (S.D.) over time after treatment with test and control whitening strip.

Discussion

Whitening strips have been available as OTC products for many years. Traditionally, their formulations have been peroxide-based, providing quick and effective whitening [8-11]. Disadvantages of these formulations include a tendency to cause transient dental sensitivity and a reduction tooth luster or gloss immediately after use [11-13]. For this reason, alternatives to peroxide-based formulations that can provide similar whitening effects without causing sensitivity and loss of tooth luster are under investigation [13-18].

In this ex vivo study, the whitening effects of a non-peroxide formulation based on Dead Sea salt, coconut oil and lemon peel were compared with those of a widely used peroxide-based formulation. While clinical studies can evaluate the efficacy of a formulation, in vivo studies by their nature encompass many variables. It is for this reason that this ex vivo study was performed, wherein the pertinent variables can be much better identified, standardized, and controlled.

Typically, individuals use the whitening strips tested in this study daily for 30 minutes at a time, over a period of 7-10 days, equating to a total of 3.5-5 hours of use. Therefore, an endpoint of 5 hours was set in the whitening strip component of this study, to parallel the application duration of in vivo users of the strips. Hourly color measurements using standard colorimetry techniques showed that the time-based whitening effects of the test and the positive control formulations were very similar. These results are in agreement with the findings of a previous clinical study,13 confirming the equivalence of the whitening effect between the non-peroxide and the peroxide formulations.

While the topic of tooth whitening has been researched and written about extensively, the concept of using a barrier product to prevent tooth staining by agents such as red wine, tea, coffee and tobacco is far more recent, and it has been addressed to a far lesser extent. The goal of using a barrier agent is to limit access of the staining agent to the tooth surface, thus hindering the development of a surface stain on the dental enamel, so that effect of whitening treatments can be maintained consistently and over a longer period of time. This is especially important when whitening treatments that may cause side effects such as sensitivity and structural alterations on the dental enamel surface are used. This study

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demonstrates the feasibility and effectiveness of using a barrier agent, confirming that enamel staining by red wine and coffee can indeed be delayed and reduced by the use of a protectant gel.

In conclusion, this preliminary study confirmed clinical findings of equivalence in the time-based whitening effects of a non-peroxide and a peroxide gel and established the feasibility of delaying and reducing stain development on the tooth surface by means of a protective barrier layer.

References