The Effect of Delivery System and Light Activation on Tooth Whitening Efficacy and Hydrogen Peroxide Penetration

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ABSTRACT

**Purpose:** To evaluate the whitening efficacy of a new two-layer technology in-office system compared to a conventional gel-type system and determine hydrogen peroxide penetration (HPP) into the pulp cavity.

**Materials and Methods:** Extracted molars (n = 60) were assigned to group NC: glycerol gel; group QPRO: 20% HP varnish (Zoom Quick Pro, Philips Oral Healthcare); group ZOOM.NL: 25% HP gel (Zoom Chairside Whitening); and group ZOOM.WL: 25% HP gel (Zoom Chairside Whitening) with light-activation. HPP levels were estimated with leucocrystal-violet and horseradish-peroxidase. Instrumental color measurements were performed at baseline (T₀), 1-day post first whitening (T₁), 1-day post second whitening (T₂), 1-day post third whitening (T₃), and 1-month post whitening (T₄). One-way analysis of variance followed by post hoc Tukey’s HSD test was performed to detect difference in ΔE* and HP penetration levels (α = 0.05).

**Results:** ΔE* of NC was lower than other groups, whereas ΔE* of ZOOM.WL was greater than the other three groups, at T₃ and T₄. Mean HPP level obtained from ZOOM.WL (1.568 ± 0.753 µg/mL) was significantly greater than those obtained from the other groups, whereas the mean HPP level observed in NC group (−0.131 ± 0.003 µg/mL) was significantly lower than the other groups.

**Conclusions:** Tooth whitening efficacy and HPP levels vary based on whitening systems used.

CLINICAL SIGNIFICANCE

The two-layer technology in-office varnish system may be an alternative whitening option to reduce chair time in the office. (J Esthet Restor Dent 00:00–00, 2016)

INTRODUCTION

Tooth whitening is a conservative and effective method to lighten discolored teeth and has become an integral component of esthetic dentistry. The popularity is reflected by the wide scope of whitening options, ranging from professional in-office procedures and custom fabricated tray-based home whitening to a variety of over-the-counter products.

Among the different whitening modalities, professionally administered in-office whitening has gained popularity due to the fact that it provides instant whitening results and that there is no need to wear a tray at home. The efficacy of a variety of...
systems has been investigated, and many studies have shown that increasing the exposure time and frequency will improve efficacy in terms of tooth color change.\textsuperscript{1–4} Active concentration of the whitening material is another important factor to consider. However, several studies have shown that the higher the concentration the faster the whitening rate,\textsuperscript{5–7} other studies did not find significant differences between various concentrations of carbamide peroxide.\textsuperscript{8–10} Furthermore, activating light sources such as plasma arc lamps, laser systems with a variety of wavelengths, and light emitting diodes (LED) have been used to enhance whitening efficacy. However, study results on the use of light activation in tooth whitening have been equivocal due to the variability of study designs and the use of different whitening materials as well as different activating lights.\textsuperscript{11}

Whitening efficacy has also been related to the ability of hydrogen peroxide (HP) to diffuse into the tooth structure.\textsuperscript{12} HP diffusion into the pulp cavity was enhanced by higher HP concentrations,\textsuperscript{13} prolonged whitening time,\textsuperscript{14} heat,\textsuperscript{14} and large open dentinal tubules of young teeth.\textsuperscript{15} LED light and laser activation demonstrated increased HP penetration levels into the pulp cavity,\textsuperscript{16} but HP levels seemed not to be correlated with whitening efficacy.\textsuperscript{12}

With the continuous introduction of new in-office whitening systems varying in delivery method and concentration, there is lack of information on the relative efficacy of these systems. Henceforth, the purpose of this study was to evaluate the whitening efficacy of a new two-layer technology in-office system compared to a conventional gel-type system and determine HP penetration into the pulp cavity. Additionally, degree of tooth color change was correlated with HP penetration levels. The null hypotheses to be tested was that first, there would be no difference in tooth whitening efficacy in terms of tooth color change in $\Delta E^*$, $\Delta L^*$, and $\Delta b^*$ among the three different systems tested. Second, there would be no difference in HP penetration levels into the pulp cavity among the different groups. Third, there would be no correlation between HP penetration levels and degree of post whitening overall tooth color change.

MATERIALS AND METHODS

Sample Selection and Preparation

Sixty extracted human molars were collected prior to the study and stored in 0.1% Thymol at 4°C. The University of Iowa Institutional Review Board approved the use of extracted human teeth with no identifiers in this study (IRB ID# 201504753). Teeth were cleaned and observed for the absence of anomalies, caries, existing restorations, and deep crack lines. The roots were marked 2 mm apical to the cementoenamel junction and trimmed off with a sectioning machine (TechCut 4, Allied High Tech Products, Inc., Compton, CA, USA). A cavity was prepared by enlarging the pulp chamber with pointed tapered diamond burs (NeoDiamond, Microcopy, Kennesaw, GA, USA) toward the lingual in order to maintain intact labial tooth structure of 2 mm thickness and encompass 50 $\mu$L of acetate buffer. A circular adhesive label 6 mm in diameter was adhered at the center of the labial surface to establish a standardized color reading and whitening area. The remaining tooth was painted with gray nail varnish (Sally Hansen, New York, NY, USA), and the adhesive label removed after drying, leaving a standardized window.

Whitening Protocol

The specimens were randomly assigned into four groups as follows, group NC: glycerin gel (Sigma–Aldrich, St. Louis, MO, USA) acting as the negative control; group QPRO: 20% HP varnish (Zoom QuickPro, Philips Oral Healthcare); group ZOOM_NL: 25% HP gel (Zoom Chairside Whitening, Philips Oral Healthcare); without light activation and group ZOOM_WL: 25% HP gel (Zoom Chairside Whitening, Philips Oral Healthcare) with LED light activation (Zoom WhiteSpeed, Philips Oral Healthcare, peak wavelength: 466 nm) set at high intensity (190 mw/cm²). The varnish was a one-time application for 30 minutes whereas the HP gel was removed and replenished with a new gel after 15 minutes as per manufacturer recommendation. A jig was fabricated...
for each specimen by gently placing the lingual surface of each tooth into a polyvinyl siloxane impression material (Aquasil Ultra Heavy, Dentsply Caulk, Milford, DE, USA) at a 30° angle from the base. Teeth were kept at room temperature (25°C) in a closed humid chamber with 100% relative humidity (General Glassblowing Co. Lab Apparatus, Richmond, CA, USA) throughout the 30-minute treatment procedure. All teeth were subjected to three whitening sessions with an interval of 3–5 days.

**Color Measurements**

Instrumental color measurements were performed with a contact-type intraoral spectrophotometer (Vita Easyshade Compact Advance, Vita Zahnfabrik, Bad Säckingen, Germany) with a 5 mm diameter probe. The Easyshade Compact was calibrated and placed perpendicular and flush to the exposed tooth surface according to manufacturer’s instruction. Color measurements were taken at baseline \((T_0)\), 1-day post first whitening \((T_1)\), 1-day post second whitening \((T_2)\), 1-day post third whitening \((T_3)\), and 1-month post whitening \((T_4)\). Measurements were performed under a color controlled lightening box (MM 4e GTI Mini Matcher, GTI Graphic Technology, Inc., Newburgh, NY, USA) at CIE D65, a color temperature of 6,500 K and light intensity of \(\approx 1,200\) lux. Color difference was calculated as \(\Delta E_{ab}^*\) from the Commission Internationale de l’Eclairage.\(^{17}\) It was calculated from the following equation:

\[
\Delta E_{ab}^* = \left[\left(L_2^* - L_1^*\right)^2 + \left(a_2^* - a_1^*\right)^2 + \left(b_2^* - b_1^*\right)^2\right]^{1/2}
\]

**Measurement of HP Penetration Levels**

HP penetration (HPP) levels were measured after the first 30-minute whitening treatment and estimated according to the method of Mottola and colleagues.\(^{18}\) Acetate buffer (40 \(\mu\)L) retrieved from the pulp cavity was mixed with 1 mL of leucocrystal violet solution (0.5 mg/mL), 0.5 mL of horseradish peroxidase solution (1 mg/mL), and 1 mL of acetate buffer. The final color intensity was measured in an UV/Visible Spectrophotometer (Perkin Elmer, Model Lambda 20, USA, Waltham, MA, USA) at a wavelength of 596 nm. A standard calibration curve with known amounts of HP was used to determine the amount in microgram equivalents in the samples.

**Statistical Analysis**

Descriptive statistics were conducted to profile all variables in the study. One-way analysis of variance (ANOVA), followed by the post hoc Tukey’s HSD (honestly significant difference) test, was performed to detect both the difference in color parameter values as well as the change in color parameters among the four groups at each post whitening time point. The change in each color parameter is defined as the difference between baseline and each post whitening time point. Moreover, one-way ANOVA with repeated measures, followed by the post hoc contrast test, was used to determine whether there was a significant difference in the color change in each color parameter within each experimental group.

One-way ANOVA, followed by the post hoc Tukey’s HSD test, was performed to assess the difference in the HPP levels among the four experimental groups. Additionally, Pearson correlation test was used to evaluate the correlation between HPP level and color change parameter \(\Delta E^*\) at 1-month post whitening. All tests utilized a significance level of 0.05. SAS for Windows (v9.4, SAS Institute Inc., Cary, NC, USA) was used for the data analysis.

**RESULTS**

**Analysis of Color Change Parameters**

Baseline color parameters showed that there was no significant effect of the type of experimental groups for \(L^*, a^*,\) and \(b^*\) \((p > 0.05\) in all instances).
TABLE 1. Comparison of color change (ΔL*, Δb*, and ΔE*) from baseline to each post whitening time point by group and within each group

<table>
<thead>
<tr>
<th>Group</th>
<th>I-day post first whitening</th>
<th>I-day post second whitening</th>
<th>I-day post third whitening</th>
<th>I-month post whitening</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ΔL* (Mean (SD))</td>
<td>Δb* (Mean (SD))</td>
<td>ΔE* (Mean (SD))</td>
<td>ΔL* (Mean (SD))</td>
</tr>
<tr>
<td>NC</td>
<td>0.01 (0.03)</td>
<td>0.29 (0.18)</td>
<td>1.46 (0.86)</td>
<td>-0.06 (0.07)</td>
</tr>
<tr>
<td></td>
<td>0.20 (0.13)</td>
<td>1.60 (0.71)</td>
<td>-0.41 (0.67)</td>
<td>0.35 (1.20)</td>
</tr>
<tr>
<td></td>
<td>1.38 (0.68)</td>
<td>-0.40 (1.02)</td>
<td>0.11 (1.02)</td>
<td>1.38 (0.69)</td>
</tr>
<tr>
<td>QPRO</td>
<td>1.95 (3.24)</td>
<td>-0.85 (2.22)</td>
<td>3.77 (2.47)</td>
<td>4.67 (2.33)</td>
</tr>
<tr>
<td></td>
<td>3.20 (3.13)</td>
<td>-4.67 (2.64)</td>
<td>6.63 (2.31)</td>
<td>-6.57 (2.51)</td>
</tr>
<tr>
<td></td>
<td>2.14 (3.32)</td>
<td>-5.85 (2.36)</td>
<td>7.70 (2.54)</td>
<td>7.23 (2.54)</td>
</tr>
<tr>
<td></td>
<td>4.85 (2.59)</td>
<td>-8.28 (3.06)</td>
<td>10.0 (2.82)</td>
<td>-</td>
</tr>
<tr>
<td>ZOOM_NL</td>
<td>3.65 (2.04)</td>
<td>-1.73 (2.06)</td>
<td>4.51 (2.14)</td>
<td>3.36 (2.51)</td>
</tr>
<tr>
<td></td>
<td>3.87 (1.58)</td>
<td>-4.39 (2.24)</td>
<td>6.17 (2.01)</td>
<td>-5.85 (2.36)</td>
</tr>
<tr>
<td></td>
<td>3.36 (2.51)</td>
<td>-5.85 (2.36)</td>
<td>7.23 (2.54)</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>4.85 (2.59)</td>
<td>-8.28 (3.06)</td>
<td>10.0 (2.82)</td>
<td>-</td>
</tr>
<tr>
<td>ZOOM_WL</td>
<td>2.21 (2.27)</td>
<td>-3.16 (1.57)</td>
<td>4.40 (1.81)</td>
<td>4.27 (1.89)</td>
</tr>
<tr>
<td></td>
<td>-8.25 (2.03)</td>
<td>9.56 (1.82)</td>
<td>9.56 (1.82)</td>
<td>-10.96 (1.81)</td>
</tr>
<tr>
<td></td>
<td>12.34 (2.06)</td>
<td>5.87 (2.37)</td>
<td>-13.66 (1.99)</td>
<td>15.15 (2.20)</td>
</tr>
</tbody>
</table>

Within each column under each variable measured, means with the same upper letter are not significantly different using the post hoc Tukey’s HSD test (p > 0.05). Within each row under the same color parameter, means with the same lower letter are not significantly different using the post hoc contrast tests (p > 0.05).

The post hoc Tukey’s HSD test revealed that mean HP penetration levels obtained from ZOOM_WL were significantly lower than those observed in the other three experimental groups. No significant difference was observed in NC (0.13 ± 0.03 g/mL) was significantly lower than those observed in the other three experimental groups. Results of the one-way ANOVA showed that there was a significant effect of the type of whitening systems on the HP penetration levels (F(3,56) = 33.15, p < 0.0001). Descriptive statistics are presented in Table 2, and Figure 1 illustrates the HP penetration levels among the four experimental groups.

Analysis of HP Penetration Levels

Table 1 summarizes the comparison of color change parameters from baseline to each post whitening time point by group and within each group. Assessing the within group effects revealed that the negative control group NC showed no significant difference in ΔL*, Δb*, and ΔE* values in 1-day post first whitening, 1-day post second whitening, 1-day post third whitening, and 1-month post whitening, respectively. It is noteworthy to point out that ZOOM_NL showed no significant difference in Δb* and ΔE* among the four different time points. When evaluating the difference among the groups at 1-day post first whitening, ΔE* of NC was lower than other groups although no difference was found among other groups, whereas ΔE* of ZOOM_WL was greater than the other three groups. No difference was found between QPRO and ZOOM_NL.
was found between QPRO (0.558 ± 0.485 µg/mL) and ZOOM_NL (0.615 ± 0.282 µg/mL).

**Correlation of HP Penetration Levels and Overall Color Change at 1-Month Post Whitening**

No significant difference was found between HP penetration levels and ΔE* within NC, QPRO, ZOOM_NL, and ZOOM_WL using the Pearson correlation test ($p > 0.05$ in all instances). However, there was a significant difference when combining all groups together ($p < 0.0001$). Thus, the Pearson correlation coefficient of 0.68 indicated a moderate positive relationship between HP penetration levels and overall color change Figure 2.

**DISCUSSION**

The ADA Council on Scientific Affairs advised patients to consult with their dentist to determine the most appropriate whitening treatment and emphasized that based on data accumulated over the last 20 years there are no significant, long-term oral or systemic health risks associated with professional at-home tooth whitening materials containing 10% carbamide peroxide (3.5% HP). This may be the most important reason that clinicians use professional at-home tooth whitening as their first treatment option to treat discolored teeth. Other benefits include relatively low cost for the whitening treatment, reduced chair-time, allocation of most of the task to the dental staff and lower incidence and severity of tooth whitening induced sensitivity when compared to in-office whitening using highly concentrated whitening material. However, it is vital to realize our patients’ needs and embrace alternatives to accommodate most everybody desiring whiter and brighter teeth. In-office whitening offers the benefit of faster whitening results that can motivate the patient to continue treatment. It is also highly appreciated by patients that cannot comply with wearing trays. However, the prolonged chair time, advocated use of light activating units and higher incidence of tooth sensitivity were significant drawbacks that limited its use to patients that could afford the higher cost and would tolerate the tooth whitening induced sensitivity.

**TABLE 2.** Comparison of hydrogen peroxide penetration levels (µg/mL) among the four groups

<table>
<thead>
<tr>
<th>Group</th>
<th>N</th>
<th>Mean (SD)</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Median</th>
</tr>
</thead>
<tbody>
<tr>
<td>NC</td>
<td>15</td>
<td>−0.131 (0.003)A</td>
<td>−0.136</td>
<td>−0.124</td>
<td>−0.131</td>
</tr>
<tr>
<td>QPRO</td>
<td>15</td>
<td>0.558 (0.485)B</td>
<td>0.037</td>
<td>1.741</td>
<td>0.445</td>
</tr>
<tr>
<td>ZOOM_NL</td>
<td>15</td>
<td>0.615 (0.282)B</td>
<td>0.126</td>
<td>1.044</td>
<td>0.574</td>
</tr>
<tr>
<td>ZOOM_WL</td>
<td>15</td>
<td>1.568 (0.753)C</td>
<td>0.643</td>
<td>3.278</td>
<td>1.751</td>
</tr>
</tbody>
</table>

Means with the same letter are not significantly different using the post hoc Tukey’s HSD test ($p > 0.05$).
Manufacturers have put efforts in overcoming these drawbacks and realized that they could make a more user-friendly system by changing their delivery method from the conventional gel type to a two-step varnish system. Philips Zoom Quick Pro that was used in this study is comprised of a first layer of 20% HP varnish followed by a second sealant layer that locks the HP layer into place and prevents inadvertent exposure to the surrounding tissue. The system was released in the USA in 2014. Upon application of the two layers, which takes about 5 minutes in the office, the patient can leave the office and peel off the adhesives at home after 30 minutes. This reduces the in-office chair time significantly and there is also no need for the use of light activation. However the efficacy and the incidence of sensitivity associated with this two-layer technology have not been well documented. This study evaluated the efficacy of this new varnish system compared to a 25% HP gel used with and without light activation.

Based on the results, our first null hypothesis was rejected. There was a significant difference in overall color change based on the whitening system used. At 1-month post whitening the light activated group (ZOOM_WL) demonstrated the highest overall color change supporting the efficacy of light activation used in conjunction with 25% HP. This is in concordance with other studies that showed that light activation enhances the degree of lightening and reduction of chroma. The use of light activation is still a topic of dispute and there are also in vitro and clinical trials indicating that light activation does not affect the change in color produced by whitening. However, it important to point out that the 20% HP varnish (QPRO) was as effective as the 25% HP gel without light activation (ZOOM_NL). To the best of our knowledge there are no studies on the efficacy of this in-office varnish system compared to the gel system and although there was a difference in formulation and concentration, it was as effective in terms of overall color change. Our study used the widely adopted color difference formula within dental research, derived from the CIE-L*a*b* system. The 50:50% perceptibility threshold of $\Delta E^* = 1$ which is used in a controlled environment and a 50:50% acceptability threshold of $\Delta E^* = 2.7$ was used to interpret tooth color change results. Notably, regardless of whitening system there was a significant increase with continuous whitening sessions.

The tooth is permeable and Bowles and Ugwuneri were the first to spectrophotometrically detect the amount of HP penetration into the pulp cavity. An in vitro model proved useful for further studies investigating various factors that might influence HPP into the pulp cavity and was also used for this study. Our second null hypothesis was rejected, since there was a significant difference in HPP levels among the four groups with the light activated group (ZOOM_WL) demonstrating the highest HPP level and no difference between the varnish system (QPRO) and the nonlight activated (ZOOM_NL) group which interestingly followed the same pattern as for overall color change. The increase in HPP levels with light activation is very consistent with another study that has shown increased levels with laser and LED lights. Increased concentration is also associated with higher penetration levels. Although there was a difference in concentration between QPRO and ZOOM_NL, the concentrations of HP found in the buffer in the pulp cavities were not significantly different. The ability to penetrate into dental hard tissue has also been related to the rheologic properties of the material. On the basis of viscosity difference, it would have been expected to observe more HPP with the 25% HP gel compared to the 20% HP varnish. This suggests that other proprietary factors may have been exerting a modulating influence. This may have to be investigated further to determine the effect of formulation on HP penetration.

Our third hypothesis was rejected. Despite the fact that within each group there was no correlation between overall color change and HPP levels, when combining all the groups together there was a modest correlation ($r = 0.68$) indicating that the higher the HPP level the higher the overall color change. Studies that evaluated the correlation between HPP level to overall color change are scarce. One study compared 40% HP using a sealed technique without
replenishment of the whitening material versus the conventional technique where the material is replenished every 20 minutes. However, another study compared the use of 40% HP with and without light activation. Both studies found that there was no correlation between HPP levels and overall color change. This may seem contradictory to our results; however, it is important to notice that even in our study we did not find any correlation within the four groups. It was only when we combined all groups together that we found a modest correlation. It is noteworthy to emphasize that penetration levels cannot be measured clinically. It is also important to point out that penetration studies are limited by the absence of the dynamics in the oral environment including the absence of positive pulpal pressure. Additionally, the pulp cavity has to be enlarged to enable quantification using acetate buffer, leucocrystal violet, and horseradish peroxidase that compromises the resemblance to in vivo. Despite these limitations higher HPP levels may be also associated with higher incidence and severity of tooth whitening induced sensitivity. As such, it would be beneficial to develop or aim for a whitening system with minimal HPP without compromising tooth whitening efficacy.

Within the limitations of this study it can be concluded that the two-layer technology in-office varnish system was as effective as the conventional gel-type system without light activation and that light activation enhanced the whitening efficacy. The hydrogen peroxide penetration levels were positively correlated with overall tooth color change. Future studies addressing the clinical efficacy, incidence and severity of tooth sensitivity, patient satisfaction, and acceptance toward this new in-office varnish system are needed to support the evidence behind this new technology.

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REFERENCES


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