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EFFECTS OF LOW TEMPERATURE ATMOSPHERIC PRESSURE PLASMA ON

TOOTH WHITENING

by

Denise Michelle Claiborne B.S.D.H. May 2010, Old Dominion University

A Thesis Submitted to the Faculty of Old Dominion University in Partial Fulfillment of the Requirements for the Degree of

MASTER OF SCIENCE

DENTAL HYGIENE

OLD DOMINION UNIVERSITY December 2011

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ABSTRACT

EFFECTS OF LOW TEMPERATURE ATMOSPHERIC PRESSURE PLASMA ON TOOTHWHITENING

Denise Claiborne Old Dominion University, 2011 Director: Prof. Gayle McCombs

Low temperature atmospheric pressure plasma (LTAPP) is a novel science being studied as an alternative light source to enhance tooth whitening. The safety and effectiveness of LTAPP has not been established therefore; the purpose of this study was to determine if LTAPP along with H_2O_2 gel would safely and effectively accelerate the tooth whitening process, in terms of lightness and temperature. Two treatment groups were utilized: 36% H₂O₂ gel only and 36% H₂O₂ gel plus LTAPP. Control group received no treatment. Experimental teeth were exposed to LTAPP at various time intervals (10, 15, and 20 minutes). Temperature was measured throughout the treatment. Pre and post photographs were taken to compare color using the CIE $L^* a^* b^*$ system. Only L^* (lightness) values were measured. Data were analyzed using descriptive statistics and ttest at the .05 level. There was a statistically significant difference in mean CIE L^* values after exposing teeth to LTAPP plus H₂O₂ gel versus H₂O₂ gel only, in the 10 minute group (p-value of .0003) and 20 minute group (p-value of .0103). There was no statistically significant difference in mean CIE L^* values among the 15 minute group (pvalue of .3815). The temperature in both groups remained under 80°F throughout the study, which is below the thermal threat for vital tooth bleaching. Results indicate that LTAPP + H_2O_2 mean CIE L* values in the 10 and 20 minute groups were significantly greater than the H_2O_2 only groups. However, the mean CIE L* values for 15 minute group were not significant.

ACKNOWLEDGEMENTS

I give God all the honor and praise for successful completion of this thesis. Without Him, none of this would have been possible. I am truly blessed for the strong relationship that I have with God, he keeps my head leveled and my heart strong.

To my extraordinary committee, it was a pleasure to be amongst so many scholars. I want to thank all of you for believing in me. I have shared a moment with each of you throughout this process that will stay with me forever. Thank you for your guidance and direction in conducting the research and with the development of this thesis. To Gayle McCombs, thank you for providing direction and tremendous support. You saw something in me while I was in the undergraduate dental hygiene program, and you have watched me grow and given me expertise advice throughout this process. To Margaret Lemaster thank you for your wisdom and sharing your experiences with me. You ladies have helped me get through this process. You were resourceful, dependable, and always had a listening ear. To Dr. Laroussi, thank you for allowing me to use the plasma lab and providing me with excellent graduate students who shared their knowledge about plasma science. Your department has been a tremendous help throughout this process. A special thanks to Arda Akman who worked with me one-onone throughout this process and helped with carrying out the experiment; whenever, I needed something you were there and very efficient in getting things completed.

To Clair Richardson, I greatly appreciate your help with the statistical analysis and taking time to explain things and meeting with me whenever needed. I want to next give thanks to my wonderful parents, Dennis and Priscilla Claiborne. They have raised me into a positive, goal-driven, and kind-hearted woman. My parents have always supported my decisions and stood by me throughout my academic years. They never had to force me to obtain an education; education was one thing that I have always been passionate about since my grade-school years. I remember the day I was dropped off at college freshman year of undergraduate school; I knew it was just the beginning of wonderful things that were yet to come.

To my big brother Dennis Jr., it has always been just the two of us, which I am glad. Thank you for believing in me and for all of your support throughout my academic career.

To my best friend and sister Amanda Bradley, we have stuck by each other throughout our academic careers, and I want to thank you for your support. To my significant other J'von McKinney, thank you for your support and encouragement throughout this process.

There are so many people who have mentored me along the way. Some of you probably do not even realize the positive impact you have made. Thank you for being such magnificent leaders and sharing your experiences with me. I would also like to thank Nakia Howard. We first met at Gresham Main residence hall and I told you that my goal was to be a dental hygienist and the rest was history. We connected the first time meeting. I was your resident, patient, student, and now your colleague. Thank you so much for being there every step of the way. To Kelly Williams, thank you for taking me under your wing and opening my eyes to endless possibilities in the profession of dental

hygiene. I cannot believe I went from being your student to now your colleague. Lastly but not least, I would like to thank the director of the dental hygiene department, Michele Darby. You are an amazing woman and one whom I admire. I have enjoyed working with you throughout my academic career, and I have learned an abundance of information from you. Thank you for believing in me. In summary, thank you to everyone who played a part in helping with the completion of this thesis.

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CHAPTER I

INTRODUCTION

Over the past decade tooth whitening has become the most sought-after cosmetic dental procedure. Aesthetic appearance of teeth is of great importance to individuals. In the United Kingdom it has been reported that 28% of adults are dissatisfied with their current tooth color appearance, and in the United States, 34% of adults are dissatisfied with the appearance of their teeth.¹ Often patients report to the dental office with the goal of having their teeth whitened because of dissatisfaction with their current aesthetic appearance. Therefore, it is the role of the oral health professional to education patients on the benefits and risks of whitening procedures, and options for both professional and over the counter (OTC) products. Patients should be informed that not all stains or discolorations can be lightened by tooth whitening procedures and results may vary among individuals.² For example, intrinsic stains such as fluorosis or tetracycline staining may not respond well to tooth whitening. The intent of tooth whitening is to remove extrinsic staining and lighten tooth surfaces. Many individuals overuse tooth whitening products because they want whiter teeth within an unreasonable time frame. Often patients buy OTC whitening products and do not follow manufacturer guidelines or use multiple whitening products simultaneously. Although, American Dental Association approved whitening products are safe as long as consumers follow manufacturer's instructions; however, patients should be advised that tooth sensitivity and oral irritation are common side effects.

Cold plasma, also known as low temperature atmospheric pressure plasma (LTAPP) is a novel science consisting of neutral and charged particles, including free radicals, which can be used to inactivate microorganisms.³ Researchers have investigated

plasma technology for use in biomedical and commercial applications including: decontamination of food, military equipment, sterilization of medical/dental instruments, and the killing of airborne and surface pathogens.⁴ LTAPP has potential uses related to the oral health profession such as inactivating microorganisms associated with oral diseases and wound healing, as well as enhance tooth whitening.⁵

Currently, there are several options available for tooth whitening which include: professional in-office, professionally dispensed, OTC and at-home application. Often professional tooth whitening procedures are augmented with an accelerator light. The role LTAPP plays in tooth whitening has become a new area of interest. The purpose of this study was to determine if LTAPP plus 36% hydrogen peroxide will safely accelerate and enhance the tooth whitening process.

Problem Statement

Can LTAPP along with 36% hydrogen peroxide gel accelerate the tooth whitening process? The intent of this research project seeks to answer the following research questions:

- 1. Does LTAPP plus 36% H_2O_2 gel lighten teeth better than H_2O_2 alone?
- 2. Does LTAPP increase tooth surface temperature?

Definition of Variables/ Terms

- **Tooth Whitening:** Whitening (lightening or bleaching) of a single tooth or multiple teeth via the application of an external agent or agents.⁶
- International Commission on Illumination (CIE) L*a*b Color Value System: Tooth whitening will be measured by the CIE Color Value System which is a mathematical scheme to describe the three dimensions of color within a color space

of equally perceived gradations. The L^* axis describes the lightness value, with values ranging from 0 (black) – 100 (pure white). A* axis measures red-green, and B* axis measures blue-yellow.

- Low Temperature Atmospheric Pressure Plasma (LTAPP): also known as cold or non-thermal plasma is a medium consisting of neutral and charged particles, including free radicals, which can be used to destroy or inactivate microorganisms.³
- **Plasma Pencil:** The plasma pencil is a plasma generator that can emit a plasma plume/jet several centimeters long.⁷
- Hydrogen Peroxide: Hydrogen Peroxide (H₂O₂) is a naturally occurring byproduct of oxygen, made from two hydrogen atoms and two oxygen atoms. H₂O₂ is a weak acid that is used for bleaching, disinfecting, and as an antiseptic.
- Temperature Measuring Device: Extech 42350 model wide range non-contact thermometer measuring ranges from 58°F 1000°F or (50°C 538°C); this was used to measure any changes in tooth surface temperature before, during, and after (Extech Instruments).

Research Hypotheses

Hypotheses were tested at the .05 level of significance

HO₁: There is no difference in the lightness of teeth exposed to LTAPP plus $36\% H_2O_2$ gel compared to those with $36\% H_2O_2$ gel alone.

HO₂: There is no difference in temperature of teeth exposed to LTAPP plus 36% H₂O₂ gel compared to 36% H₂O₂ gel alone.

Assumptions and Limitations

The plasma equipment was reliable. The equipment used to determine temperature and measure lightness was reliable. Non-vital teeth were used versus vital teeth; therefore, generalization can only be applied to non-vital teeth. The teeth obtained for the study came from various sources; therefore, tooth color varied at baseline. In addition, the teeth in the study were different in texture and thickness.

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CHAPTER II

REVIEW OF THE LITERATURE

History of Tooth Whitening

Tooth whitening also known as bleaching is the most requested cosmetic dental procedure performed; therefore, it is important that the oral health professional and patient collaborate together to determine the most appropriate whitening treatment. Fehernbach suggests that two criteria must be met when determining the need for tooth whitening: noticeable tooth discoloration is present, and teeth to be treated are healthy and free of any restorations.⁸ When discussing tooth whitening with a patient the goal is to help them understand that whitening must be used in moderation, and to maintain the desired color shade, certain lifestyle changes may have to be made. For example, tobacco, coffee, red wine, and certain foods can darken teeth over time. A heavy coffee/tea drinker may have difficulty achieving desired results, or once results are achieved, discoloration may revert back to the original darker shade. In general, before whitening procedures begin, whether in-office or at-home, the patient must have realistic expectations.

Hydrogen peroxide (H₂O₂) is a widely used tooth bleaching agent that is effective and safe. The exact mechanism of H₂O₂ tooth bleaching is not completely understood; however, the most common theory is that H₂O₂ breaks down to produce oxygen radicals, which attack organic pigment molecules, thus causing enamel lightening.⁹ Traditional vital tooth bleaching may be performed at home and in-office. Professional in office bleaching, generally consists of a high concentration of H₂O₂ (35 - 50%) or carbamide peroxide (CP) (35 - 40%) accompanied by an accelerant light.² Typically whitening procedures performed at home consists of 3 - 10% H₂O₂ or 10 - 30% CP.¹⁰ At-home whitening procedures may be used as a maintenance treatment after in-office bleaching or as standalone applications.

In office vital tooth bleaching is usually achieved using a custom fabricated tray that fits over the patient's teeth. H_2O_2 gel or CP gel is dispensed into the tray and placed on the teeth for 30 minutes to two hours, with or without a light source, while the patient remains in the dental office. Researchers suggest the efficacy of tooth bleaching is determined by the duration of the bleaching.² In a study conducted by Haywood and colleagues, subjects used at-home night-guard vital bleaching for six weeks with 10% CP. The purpose of the study was to gather information on the efficacy and esthetics of at-home tooth whitening procedures. The results revealed that 92% of the 38 individuals enrolled experienced lightening of the treated teeth.¹¹ Follow-up questionnaires mailed to the participants, periodically over a 3 year period, revealed that 23 of the respondents experienced little or no reversal in color.

Non-vital bleaching of discolored pulpless teeth was first described by Truman in the mid 1960's.¹² A variety of agents such as chloride, sodium hypochlorite, sodium perborate, and hydrogen peroxide have been used to lighten teeth, alone in combination, with and without heat activation.² The "walking bleach" technique, introduced in the 1960's, involves a mixture of sodium perborate and water placed into the pulp chamber and sealed off between the patient's dental visits.¹³ This method was later modified and the water was replaced by 30-35% H_2O_2 . Hydrogen peroxide acts as a strong oxidizing agent, which produces free radicals, reactive molecules and hydrogen peroxide anions. The reactive molecules react with long chains of dark-colored chromophore molecules, and split them into smaller more diffusible molecules. Overall, the outcome of bleaching

depends on the concentration of the bleaching agent, the ability of the agent to react with chromophore molecules, and the number of times the agent reacts with chromophore molecules

Adverse Effects

Hypersensitivity, mucosal irritation, enamel erosion, and reduced bonding strength of restorations are common side effects of vital external bleaching. Even though hypersensitivity and oral mucosal irritations are usually transient, the effects range from slight to severe depending on treatment used. Data from various studies of 10% CP revealed that 15-65% of the patients reported tooth sensitivity with the use of 10 % CP athome whitening treatment.^{11, 14, 15} In regards to in-office bleaching, approximately 68% of patients reported tooth sensitivity after in-office bleaching with 30-35% H₂O₂ gel and a heat source.² Risk factors for sensitivity include: the percentage of hydrogen peroxide or carbamide, length of exposure and frequency of bleaching. Oral tissue irritation is a common result of H₂O₂ or CP exposure on the palate, gingival or throat. In animal experiments using 1% H₂O₂, tissue exposure for 6 to 48 hours resulted in epithelial damage and acute inflammation.² Bleaching also can affect amalgam restorations by reducing the bonding strength, thus causing an increased risk of mercury to be released.² Lastly, enamel erosion may result if bleaching agents are used too frequently or for long periods of time.²

Acidity (pH)

Acidity, exposure time, and frequency are important factors to be considered when performing whitening procedures. Manufactures suggest leaving the whitening products on the teeth anywhere from 1 hour - 8 hours. The pH of the oral cavity is between 7.0 and 7.4; whereas, stable H_2O_2 gel has a pH of approximately 4.5 - 6. The pH of the tooth whitening product will determine how long the gel should remain on the tooth surface before demineralization occurs. For that reason, it is important to follow manufacturer's recommendations when performing whitening procedures.

Price, Sedarous, and Hiltz, evaluated the pH of 26 commercially available tooth whitening products. ¹⁶ The products were divided into four categories: over-the-counter (OTC); in-office bleaching (IOB); dentist supervised home-bleaching (DSHB); and whitening tooth paste. The pH levels of Colgate Total toothpaste and two carbonated drinks (Pepsi and Coca-Cola) were also evaluated. The pH was measured using a HANNA pH meter and an ORION semi-micro pH electrode (Orion Research Incorporated, Boston, MA). Results demonstrated that for all whitening products the mean pH ranged from 3.67 - 11.13; pH of (DSHB) ranged from 5.66 - 7.35; IOB products ranged from 4.22 - 8.35, and the carbonated drinks ranged from 2.45 - 2.49.¹⁶ DSHB and OTC products to the carbonated drinks the pH was significantly lower ranging from (2.45 - 2.49).¹⁶

Oehmigen and colleagues evaluated the role of LTAPP acidification in reducing antimicrobials.¹⁷ The researchers studied water disinfection by indirect plasma treatment using a surface dielectric barrier discharge (DBD). The results suggest that indirect DBD treatment of different liquids resulted in a decrease of pH from 7 to less than 4 within minutes. Similarly, other research support the theory that plasma influences the pH of liquids.^{18, 19} Tang et al., studied the effects of plasma exposure on pH in algal cultures and

culture media.²⁰ Researchers observed a time-dependent pH decrease in algal cultures, culture media, and deionized water after plasma treatment. The range of pH was dependent on the length of plasma exposure. For example, after 640 seconds of plasma exposure, the pH in deionized water dropped to 5.27, and the cultural media dropped to 6.66. When exposure time was 320 seconds or longer the pH in all cultures dropped below 3.0.²⁰

In conclusion, when comparing the pH of LTAPP to H_2O_2 , acidity ranges are similar. One element that is consistent in all the published research is the correlation between exposure time and pH; as time increases, pH decreases.

Lightness/Temperature

In-office bleaching has some advantages over at-home bleaching such as: reduction in material ingestion, limited soft tissue exposure, reduced total treatment time, and greater potential for immediate results. In office "power bleaching" suggests reduced treatment time, by accelerating the whitening procedures with a light source such as lasers. The light source enhances bleaching by heating the H₂O₂, thus increasing the rate of decomposition and oxygen to form oxygen free radicals, which aids in the release of stain-containing molecules.²¹ Luk and colleagues evaluated the effect of light energy on peroxide tooth bleaching.²¹ Researchers randomly selected 125 extracted human teeth that were longitudinally divided into buccal and lingual halves and randomly assigned into 25 groups of ten. Baseline shades of the teeth were assessed using the Vita Lumin Shade Guide (Vita Zahnfabrik H. Rauter GmbH & Co., Bäd Sackingen, Germany). A prototype electronic dental color analyzer assessed the tooth color by calculating numerical values from 1-16 (1= lightest to 16=darkest). Teeth were mounted in ethylene

vinyl acetate molds. Whitening agents tested included: Opalescence Xtra, Quick White, StarBrite Power Pack, Nupro Gold Teeth Whitening Gel, and a placebo gel. The light sources tested were: Spectrum Halogen Curing Light, Prototype Infrared Light, Argon Laser, or Carbon Dioxide Laser, no light was used as a control. The researchers applied various light sources according to manufacturer's recommendations. Bleaching gels were placed on the middle third of the tooth, approximately 2-mm in thickness. The light sources were then directed onto the bleaching gel from a distance of 1-2 mm, for 30 seconds. Researchers allowed the bleaching gel to remain on the tooth for 180 seconds before rinsing. For each tooth, the bleach and light application were repeated six times, sequentially, for a total exposure of 21 minutes for each bleaching application and a total of 3 minutes for each light exposure. Temperature was measured on the outer and inner enamel and dentin, before and after the 30 second light exposure, using a thermocouple thermometer.²¹ Pre and post-treatment color was measured. Researchers stored the teeth in distilled water and incubated them at 98.6°F after approximately one week; the teeth were re-evaluated for color changes.

The results revealed that the treatment groups exposed to Infrared light and carbon dioxide laser showed the highest mean increase in outer or inner surface temperature. The Infrared light's outer enamel surface mean temperature increased to 73.9°F, and inner dentin mean temperature increased to 70.7°F. The carbon dioxide laser outer enamel surface mean temperature increased to 58.7°F, and the inner dentin mean temperature increased to 58.7°F, and the inner dentin mean temperature increased to 50.4°F. The degree of temperature increase in these groups was affected by the type of bleach and light used²¹ but did not reach the threshold beyond which pulpal inflammation occurs.²² Limited conclusions can be made with regards to

temperature in this *in-vitro* study due to varied results; perhaps in a *vivo* study the pulpal blood flow would help stabilize increases in pulpal temperature.²¹

Buschalla and Attin studied external bleaching procedures utilizing the activation of heat, light, or laser.²³ Factors that influence temperature changes are individual tooth properties such as microhardness, thickness of applied bleaching material, and the length of exposure of the bleaching agent to the tooth surface.²³ Researchers found that activating bleaching agents by any form of heat may have adverse effects on pulpal tissue due to the increase of intra-pulpal temperature. The researchers suggested ~118°F is the critical value that can cause pulpal necrosis in vital teeth, which leads to irreversible damage.²³

Tooth Whitening Applications

Machot, Noack, and Hoffman, studied the effects of an at-home paint-on whitening product using two methods of color analysis: Digital Adobe Photoshop and Shade Vision System (X-Rite).²⁴ Oualifying teeth were free of restorations or caries, and had a minimum Vita shade of A3 or darker. To standard measurements and stabilize the teeth during color measurements, they were intruded into blocks of Flextime Easy Putty. Ninety extracted maxillary teeth were divided into two groups: prophylaxis and nonprophylaxis. The prophylaxis group received debridement with ultrasonic instrumentation, followed by polishing with a soft rubber cup using prophy paste. The prophylaxis groups were further divided into three whitening subgroups (X, Y, and Z) consisting of 15 teeth each. Group X (positive control) received Colgate Simply White gel and Colgate Palmolive; Group Y (test group) received Easy White gel; Group Z (placebo group) received methylhydroxyethylcellulose gel, anhydrous glycerol,

propylene glycol, and water.²⁴ The test gel contained 7.0% hydrogen peroxide; the positive control gel contained 5.9% H₂O₂. Treatment lasted for two weeks consisting of applying one of the three gels, twice daily with 8 hours between applications. At the end of the 15 minute whitening procedure the teeth were rinsed with water and then placed in fresh artificial saliva at 98.6°F to mimic remineralizing conditions. Tooth color assessment was performed using digital imaging analysis (Adobe Photoshop) and tooth shade was evaluated by a colorimeter (Shade Vision).

Results determined that whitening is effective with or without prophylaxis; however, prior prophylaxis contributed to improved post-treatment outcomes. Both test gel and positive control gel resulted in greater tooth color improvements compared to placebo gel. Data concluded that there was no difference in the teeth that receive a prophylaxis verses those that did not, yet researchers suggest that pre-whitening prophylaxis is beneficial.²⁴

Foundational Plasma Research

Low temperature Atmospheric Pressure Plasma (LTAPP), also known as cold or non-thermal plasma, is a medium consisting of neutral and charged particles, including free radicals, which can be used to destroy or inactivate microorganisms.³ Plasma is the fourth state of matter that is a partly ionized gas comprised of molecules, atoms, electrons, and ions. The remaining 1% of the visible universe consists of three other states of matter: solids, liquids and gases. Non-thermal plasma can be thought of as cold combustion producing highly reactive free radicals via electron-neutral collisions instead of using heat.³ Researchers have utilized LTAPP in a number of biomedical and commercial applications such as wound healing, decontamination of food and equipment, and inactivation of airborne and surface pathogens.^{4,7} One of the characteristics of nonequlibrium plasmas is its enhanced chemistry without the need for elevated gas temperatures.⁷ The plasma pencil plume can be focused into the direction of the object or objects to be treated. The plasma plume measures 5-10 cm; however the length can be controlled by the gas flow rate and by the magnitude of applied voltage pulses and/or their width. The plasma plume remains stable at low temperature and can be touched or directed at human tissues without harm. The plasma pencil developed by Laroussi, is used for direct plasma exposure to a target.⁷ The plasma pencil is a hollow tube measuring about 2.5 cm in diameter that contains two copper electrodes. A gas such as helium or a mixture of gases such as helium and oxygen is ignited by applying submicrosecond square high voltage pulses between two electrodes which spark the plasma plume. The gas mixture content can be adjusted throughout procedures to allow stabilization for several hours.

LTAPP Enhanced Tooth Whitening

The intent of the current study is to evaluate the effectiveness of LTAPP to enhance tooth whitening. Researchers have been studying the effects of LTAPP on tooth whitening for several years.^{9, 25, 26, 27} Published research has been performed using human extracted teeth. Some researchers have stained the teeth prior to performing LTAPP enhanced whitening procedures, while others compared the whiteness of the teeth without staining.

Lee and colleagues conducted a study to determine if LTAPP had potential for tooth whitenin.⁹ Researchers used 28 extracted human teeth that were cut longitudinally and placed into either experimental or control group. In order to evaluate the whitening of external tooth surface and dentin, internal and external bleaching was performed. The experimental group received 28% H_2O_2 every 30 seconds plus plasma for 10 minutes; the control was treated using H_2O_2 alone for the same duration. Researchers analyzed bleaching results by comparing the overall color changes with before and after photos using a Canon EOS Digital with Macro lens and ring flash (Tokyo, Japan). The International Commission on Illumination was used to evaluate the difference in CIE L* values via Adobe Photoshop CS2 series. Photographs revealed an increased brightness of the experimental teeth, but not the control teeth. In the experimental group tooth surface temperature increased from room temperature (77°F) and stabilized near 100.4°F after 1.5 minutes of operation. The bleaching efficacy in the experimental group was three times better than the control group. Results suggest that plasma has a positive effect on time in the tooth whitening process; not only does it enhance whitening, but accelerates the process.⁹

Lee and colleagues studied the effects of H_2O_2 using a plasma microjet (PMJ).²⁵ Thirty extracted human teeth were cut longitudinally, one half of each tooth was randomly assigned to either the experimental or control group. Fifteen of the sectioned teeth were placed into coffee, and fifteen were placed in red wine, for seven days. Before treatment the teeth were rinsed in distilled water to remove loose debris. Teeth in the experimental group were exposed to plasma plus H_2O_2 for 20 minutes; teeth in the control group were treated using only H_2O_2 for 20 minutes. Photographs were taken before and during treatment at 5 minute intervals, using a digital camera. To assess color change the CIE color scale was used. Results showed a color change occurred over time in the experimental group, whereas no significant color change occurred in the control group. The difference between brightness and color tone in the experimental group was

significant after 20 minutes of treatment. The combination of plasma and H_2O_2 improved the bleaching efficacy by a factor of 3.1 for coffee and 3.7 for wine compared with using H_2O_2 alone.²⁵

Sun and colleagues conducted a study to evaluate the effects of a plasma microjet (PMJ) along with 35% H₂O₂, compared to H₂O₂ alone.²⁶ Researchers used sixty caries free premolars extracted for orthodontic reasons. The teeth were divided into three groups: Group A received 35% H₂O₂ gel only and was kept at room temperature for 20 minutes; Group B received 35% H₂O₂ gel PMJ for 20 minutes; and Group C received 35% H₂O₂ gel and was placed in an incubator for 20 minutes. The bleaching results were analyzed with a Crystaleye Spectrophometer (Olympus Corporation, Tokyo, Japan).²⁶ In order to quantify the results the images of the tooth specimens were transformed into color classification scheme using CIE. The results revealed that PMJ plus H₂O₂ gel (Group B) showed greater increase in lightness than those in Group A and C. Throughout the bleaching treatment temperature did not exceed 104°F. Researchers concluded PMJ treatment does not pose safety issues related to thermal effects, and that direct-current PMJ in conjunction with 35% H₂O₂ gel is effective in accelerating the tooth whitening process.²⁶

Pan and colleagues conducted a study evaluating the effectiveness of LTAPP in the tooth whitening process.²⁷ Sixty extracted premolars were kept whole and divided into three groups: Group A: exposed to an air blow and saline solution; Group B: exposed to LTAPP and saline solution; and Group C: exposed to 35% hydrogen peroxide gel. Exposure time was 20 minutes for all treatment groups. Researchers measured the following: temperature, microhardness, lightness, morphology, Reactive oxygen species (ROS), and Optical Emission Spectra (OES). Temperature was measured throughout treatment using a thermal couple temperature remained at 104°F throughout the experiment. Microhardness was determined with a microhardness tester (Shimadzu Corporation, Kyoto, Japan). Lightness was evaluated before and after treatment using a Crystaleye Spectrophotometer then quantified in the CIE.²⁷ OES of the air PMJ were recorded in the UV-visible-near infrared range along the axial direction. The light was collected by a fiber optics cable and imaged onto the entrance slit of a 0.75-m spectrometer (Princeton Instrument/Action Spectra Pro 2750) equipped with an 1800-groove/mm blazed holographic grating. ROS generated at the plasma liquid interface are believed to be essential for tooth whitening processes Three ROS were evaluated: hydroxyl radical, superoxide anion radical, and singlet oxygen.

Results revealed the whiteness of the teeth in the LTAPP plus saline group improved significantly after treatment (Group B).²⁷ Researchers believe that the improved whitening efficacy of the PMJ with the saline solution is attributed to the ROS from plasma and water interacting directly or indirectly with stain molecules on the tooth surface thus breaking the bonds of the long carbon chains. ROS generated at the plasmaliquid interface are probably more easily accessible to the tooth than those produced by hydrogen peroxide and localized acid pickling on the surface of the teeth may also contribute to the increased whitening efficacy. Even though ROS is a possible contributor to the whitening process, it can be harmful to the oral tissues and respiratory system if not properly controlled. Researchers concluded that PMJ has no adverse effect on enamel microhardness. This study suggest that the use of LTAPP and saline will enhance the tooth whitening process over the use of H_2O_2 alone, or air blow and saline.²⁷ LTAPP and its role in dentistry is fairly new, yet the future of LTAPP in whitening applications is promising. Many questions have been raised over the past years about LTAPPs effectiveness and its safety. The literature published to date suggests no thermal injury. Whether plasma removes stain or is actually whitening the tooth surface remains an area of debate. The ultimate goal is for LTAPP is to be effective and safe not only in whitening applications, but in other areas of dentistry as well.

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CHAPTER III

METHODS AND MATERIALS

Sample Description

Thirty extracted human teeth (21 premolars and 9 anteriors) free of caries lesions and stain were used to evaluate the effect of LTAPP on tooth whitening. Prior to study initiation extrinsic stains were removed with an ultrasonic scale. Study teeth were stabilized in dispensing cups using dental plaster. To localize the plasma target area and gel placement, paper discs were placed over each tooth so that only the center was exposed (Figure 1). To standardize the lightness measurements, three points were chosen, in a horizontal pattern, across the middle section of the tooth. Mean CIE L^* value measurements were obtained.



Figure 1. Target Area with Three Standardized Measurement Points

Procedures

Thirty anterior (A) and posterior (P) teeth were divided into three groups:

LTAPP + 36% H_2O_2 (n=12); 36% H_2O_2 alone (n=12); and control (n=6). Treatment teeth were subdivided according to time (10, 15, and 20 minutes) (Table 1). The 36% H_2O_2 only group received gel for all three time intervals. The LTAPP plus 36% H_2O_2 group received gel followed by plasma exposure, for the same time intervals. The control group received no treatment. The placement of H_2O_2 gel was approximately 1-mm thick on each tooth. The plasma generator remained 1 inch from the surface of the tooth, while the plume was in contact with the tooth the entire duration of exposures (Figure 2). The plasma settings consisted of the following parameters: 5 KHz frequency, 500 nanoseconds (ns) pulse width, and 7 kV amplitude; and the gases were helium and oxygen.



Figure 2. Plasma Plume Affixed at Target Area

N=30 $P = posterior A = anterior$						
n=6 teeth		n=12 teeth	n=12 teeth			
1P, 1A		3P,1A	3P,1A	20		
1P, 1A		3P,1A	3P,1A	15		
1P, 1A		3P,1A	3P,1A	10		
treatment						
No		H_2O_2		minutes		
Control		LTAPP +	H ₂ O ₂ Only	Time –		
Table 1. Treatment Groups						

Table 1. Treatment Groups

Temperature was measured periodically throughout the experiment using Extech 42350 wide range non-contact thermometer (Extech Instruments, South Burlington, VT), which calculates temperature ranging from $58^{\circ}F - 1000^{\circ}F$ or $(50^{\circ}C - 538^{\circ}C)$ (Figure 3). The researchers monitored the surface temperature to ensure that it remained below the critical value for vital tooth whitening (106.7°F). Photos were taken pre and post exposure to compare CIE *L** values using the Canon EOS macro lens with ring flash and 140 magnification digital camera (Cannon USA, Inc., Lake Success, NY) (Figure 4). To ensure standardization photos were taken using the same camera, photographer, room and lighting. Lightness was obtained using the International Commission on Illumination Color System. Color was measured using CIE *L** values ranging from 0 (black) to 100 (pure white). Photographs were taken before and after each treatment step, then transferred from the camera to Adobe Photoshop CS5 series. Three standardized points were calculated on each tooth photographic image and the mean *L** values were averaged (Figure 1). The mean *L** values were entered into Microsoft Excel and analyzed.



Figure 3. Extech Thermometer for External Temperature Measurement



Figure 4. Canon Digital Camera for Pre and Post Photos

Statistical Analysis Section

Hypotheses were tested at the .05 level of significance. Descriptive statistics were ran for each group and time interval. The descriptive statistics included: mean, mean difference, standard deviation, and standard error. Data analysis consisted of the following: descriptive statistics, F-test, and t-test Assuming Equal or Unequal variances were used in analyzing the data. The F-test was used in determining the appropriate t-test with each data set.

CHAPTER IV

RESULTS

HO₁: There is no difference in the lightness of teeth exposed to LTAPP plus 36% H_2O_2 gel compared to those with 36% H_2O_2 gel alone, when tested at alpha = .05.

Results demonstrated a statistically significant difference in lightness after using LTAPP plus H_2O_2 gel versus H_2O_2 gel alone, in the 10 minute group (p-value of .0003) (Table 2). The LTAPP plus H_2O_2 gel group revealed mean CIE L^* values differences between pre and post photos of 8.48; whereas, the H_2O_2 only group reported a mean CIE L^* value difference of 1.85 (Table 3) (Figure 5).



Figure 5. Mean difference in CIE L^* values between Plasma + H₂O₂ and H₂O₂ only for 10 minutes

	Variable 1	Variable 2
Mean	1.85	8.475
Variance	1.203333333	3.115833333
Observations	4	4
Pooled Variance	2.159583333	
Hypothesized Mean		
Difference	0	
Df	6	
	-	
t Stat	6.375524175	
P(T<=t) one-tail	0.0003499*	
t Critical one-tail	1.943180274	
P(T<=t) two-tail	0.0006998	
t Critical two-tail	2.446911846	

Table 2. Mean difference in CIE L^* values between Plasma + H₂O₂ and H₂O₂ only for 10 minutes

Table 3. Mean difference in CIE L^* values between Plasma+ H₂O₂ and H₂O₂ only for 10 Minutes

LTAPP+H₂O₂ 10 Min Group				H ₂ O ₂ Only 10 Min Group			
Teeth	Pre	Post	Difference	Teeth	Pre	Post	Difference
10A1	52	60.3	8.3	10B1	55.6	58.3	2.7
10A2	67.3	78.3	11	10B2	59.3	62	2.7
10A3	58	65	7	10B3	63.6	64	0.4
10 A 4	57	64.6	7.6	10B4	54	55.6	1.6
Mean	58.575	67.05	8.475 [*]	Mean	58.125	59.975	1.85*
Std Error	3.1907092	3.8979696	0.8825862	Std Error	2.1359522	1.8763329	0.5484828
Std Dev	6.3814183	7.7959391	1.7651723	Std Dev	4.2719043	3.7526657	1.0969655

Results for the 15 minute group showed no statistically significant difference in lightness between LTAPP plus H_2O_2 gel vs. H_2O_2 gel only (p-value of .3815) (Table 4). The LTAPP plus H_2O_2 gel group revealed mean CIE L^* values differences between pre and post photos of 4.25; whereas, the H_2O_2 only group reported a mean CIE L^* value difference of 3.75 (Table 5 and Figure 6).



Figure 6. Mean difference in CIE L^* values between Plasma + H₂O₂ and H₂O₂ only for 15 minutes

Table 4. Mean	difference in CIE L* values between Plasma + H ₂ O ₂
and H ₂ O ₂ only	for 15 minutes

	Variable 1	Variable 2
Mean	3.75	4.25
Variance	8.843333	0.756667
Observations Hypothesized	4	4
Mean Difference	0	
df	4	
t Stat	-0.32275	
P(T<=t) one-tail	0.381526	
t Critical one-tail	2.131847	
P(T<=t) two-tail	0.763052	
t Critical two-tail	2.776445	

Table 5. Mean	difference in	CIE L*	values	between	Plasma	+	H ₂ O ₂ and	H_2O_2	only
for 15 minutes									

LTAPP+H ₂ O ₂ 15 Min Group				H ₂ O ₂ Only 15 Min Group			
Teeth	Pre	Post	Difference	Teeth	Pre	Post	Difference
15A1	68	72.6	4.6	15B1	53	55	2
15A2	64.6	69	4.4	15B2	65.3	69.6	4.3
15A3	66.6	71.6	5	15B3	52.6	60.3	7.7
15 A 4	60.3	63.3	3	15B4	50.3	51.3	1
Mean	64.875	69.125	4.25	Mean	55.3	59.05	3.75
				Std			
Std Error	1.6769889	2.0846163	0.4349329	Error	3.3860006	3.9720901	1.4868871
Std Dev	3.3539777	4.1692325	0.8698659	Std Dev	6.7720012	7.9441803	2.9737743

Results for the 20 minute group demonstrated that there was a statistically significant difference in lightness before and after using LTAPP plus H_2O_2 gel versus H_2O_2 gel alone (p-value of .0103) (Table 6). The LTAPP plus H_2O_2 gel group showed a mean difference in the CIE *L** values between pre and post photos of 8.675; whereas, the H_2O_2 only group reported a mean difference of 3.675 (Table 7, Figure 7).



Figure 7. Mean difference in CIE L^* values between Plasma + H₂O₂ and H₂O₂ only for 20 minutes.

	Variable 1	Variable 2	
Mean	3.675	8.675	
Variance	3.469167	6.8225	
Observations	4	4	
Pooled Variance	5.145833		
Hypothesized Mean Difference	0		
Df	6		
t Stat	-3.11715		
P(T<=t) one-tail	0.01033*		
t Critical one-tail	1.94318		
P(T<=t) two-tail	0.02066		
t Critical two-tail	2.446912		

Table 6. Mean difference in CIE L^* values between Plasma+ H_2O_2 and H_2O_2 only for 20 minutes

Table 7. Mean difference in CIE L^* values between Plasma + H₂O₂ and H₂O₂ only for 20 minutes

LTAPP+H ₂ O ₂ 20 Min Group				H ₂ O ₂ Only 20 Min Group			
Teeth	Pre	Post	Difference	Teeth	Pre	Post	Difference
20A1	60.6	68.6	8	20B1	63.6	66.3	2.7
20A2	53.6	65.6	12	20B2	54.6	60.3	5.7
20A3	56.3	62	5.7	20B3	63.3	68	4.7
20A4	55	64	9	20B4	65	66.6	1.6
Mean	56.375	65.05	8.675*	Mean	61.625	65.3	3.675*
Std Error	1.5123795	1.393736	1.3059958	Std Error	2.3707857	1.7073371	0.931285
Std Dev	3.0247589	2.787472	2.6119916	Std Dev	4.7415715	3.4146742	1.8625699

Considering there was statistically significant difference found in the 10 minute and 20 minute LTAPP plus H_2O_2 groups, a *t*-test was run to evaluate what impact time presented in the whitening process. The results demonstrate that it cannot be assumed a greater difference in lightness using LTAPP plus H_2O_2 gel for 10 minutes, as opposed to 20 minutes (p-value of .4515) (Figure 8, Table 8, and 9).





	Variable 1	Variable 2
Mean	8.475	8.675
Variance	3.1158333	6.8225
Observations	4	4
Pooled Variance	4.9691667	
Hypothesized Mean Difference	0	
df	6	
t Stat	-0.1268829	
P(T<=t) one-tail	0.4515892	
t Critical one-tail	1.9431803	
P(T<=t) two-tail	0.9031784	
t Critical two-tail	2.4469118	

Table 8. Mea	n difference in	CIE L* value	s Plasma + H_2O_2
at 10 and 20	minutes after I	LTAPP exposu	re

Table 9. Mean difference in CIE L^* values Plasma + H₂O₂ at 10 and 20 minutes after LTAPP Exposure

H ₂ O ₂ +LTAPP 20 Min Group				H ₂ O ₂ +LTAPP 20 Min Group			
Teeth	Pre	Post	Difference	Teeth	Pre	Post	Difference
10A1	52	60.3	8.3	20A1	60.6	68.6	8
10A2	67.3	78.3	11	20A2	53.6	65.6	12
10A3	58	65	7	20A3	56.3	62	5.7
10A4	57	64.6	7.6	20A4	55	64	9
Mean			8.475	Mean			8.675

HO₂: There is no difference in temperature of teeth exposed to LTAPP plus 36% H₂O₂ gel compared to 36% H₂O₂ gel alone, when tested at the .05 level.

Temperature was evaluated periodically throughout the experiment; using the Extech non-contact thermometer directed to the center of the tooth within the target area. During LTAPP exposure temperatures remained below 80°F for all groups.

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CHAPTER V

DISCUSSION

The present study explored the effectiveness of LTAPP as a potential method for tooth whitening. The primary purpose of this study was to test the difference in lightness utilizing LTAPP plus H₂O₂, as opposed to H₂O₂ alone. Secondly, the study measured temperature. Previous studies suggest that LTAPP causes no thermal harm to tooth surfaces and that tooth surface temperatures stabilized at 98.6°F – 104°F.^{25, 26, 27} Researchers suggest temperatures between 114.8°F - 123.8°F are deemed safe for bleaching non-vital teeth.²¹ Additional literature suggests that during vital tooth whitening temperatures above 106.7°F may lead to irreversible pulpal damage.^{22, 23} In the current study, temperatures remained below 80°F for all groups.

Results from the present study revealed that the most significant difference in lightness (CIE L^* values) was revealed in the 10 and 20 minute LTAPP plus H₂O₂ groups. The 15 minute group revealed no statistically significant difference in lightness between groups. In all treatment groups, the lightness shade at baseline varied, which may have been a contributing factor in lightness differences between 10 and 20 minute group, versus the 15 minute group. In previous studies, researchers did not see a significant difference in lightness until the 20 minute interval whereas, in the current study, differences were seen as early as 10 minutes.^{9, 25, 26, 27} Results from the present study suggest that the use of LTAPP with the plasma pencil plus H₂O₂ safely enhances and accelerates the tooth whitening process.

Limitations

Due to utilizing non-vital teeth from various sources, results cannot be generalized to vital teeth. Several attempts were made to maintain the validity and reliability throughout the study, such as: stabilizing teeth, standardizing exposure area, and using one person to operate the plasma and photograph equipment. Posterior and anterior teeth CIE L* values were calculated together.

Recommendations for Future Studies

Plasma and its role in dentistry is fairly novel. The future of plasma enhanced whitening is very promising. Researchers have evaluated tooth whitening using LTAPP and determined that whitening is safely enhanced and accelerated with the use of LTAPP.^{9, 25, 26, 27} To date, studies have been conducted on non-vital teeth with varying results. The following are recommendations for future studies:

- Focus on lightness or (CIE *L** values).
- Use vital teeth
- Store teeth in a saline solution or in 0.1% thymol solution to avoid dehydration.
- Evaluating pH on teeth that are exposed to LTAPP and H_2O_2 .
- Evaluate more anterior teeth for lightness.
- Measure internal and external temperature during specific time intervals

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CHAPTER VI

SUMMARY and CONCLUSIONS

The present study examined the effects of LTAPP in tooth whitening. Results demonstrate that there is a statistically significant difference in lightness when using LTAPP plus H_2O_2 gel in the 10 and 20 minute groups; however, the 15 minute group revealed no statistically significant difference. Temperature for both groups (H_2O_2 gel plus LTAPP and H_2O_2 gel only) remained under 80°F, which is below the thermal threat for vital bleaching (106.7 °F). Therefore the following assumptions are put forth:

- HO₁: There is no difference in the lightness of teeth exposed to LTAPP plus 36% H₂O₂ gel compared to those with 36% H₂O₂ gel alone, when tested at the .05 level. Hypothesis rejected.
- 2. HO₂: There is no difference in temperature of teeth exposed to LTAPP plus 36% H_2O_2 gel compared to 36% H_2O_2 gel alone, when tested at the .05 level. Hypothesis accepted.

LTAPP has a vital role in tooth whitening. However, more research is needed to demonstrate that LTAPP is safe and an effective method to enhance tooth whitening.

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APPENDIX A

RAW DATA

	10A1	10A2	10A3	10A4 Ant teeth	Controls	
Before	52	67.3	58	57	Premolar 57.3	
After	60.3	78.3	65	64.6	Anterior 57	
	10B1	10B2	10B3	10B4 Ant teeth		
Before	55.6	59.3	63.6	54		
After	58.3	62	64	55.6		
	15A1	15A2	15A3	15A4 Ant teeth	Controls	
Before	68	64.6	66.6	60.3	Premolar 60	
After	72.6	69	71.6	63.3	Anterior 55	
	15B1	15B2	15B3	15B4 Ant teeth		
Before	53	65.3	52.6	50.3		
After	55	69.6	60.3	51.3		
	20A1	20A2	20A3	20A4 Ant teeth	Controls	
Before	60.6	53.6	56.3	55	Premolar 63.6	
After	68.3	65.6	62	64	Anterior 53	
	20B1	20B2	20B3	20B4 Ant teeth		
Before	63.6	54.6	63.3	65		
After	66.3	60.3	68	66.6		

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