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Minimum Exposure Time of Light-Emitting Diode (Led) Light Curing Machine in Reducing Class I Cavity Preparation Microbial Contaminants

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Abstract: **Cavity medication or disinfection is a common procedure after cavity preparation in order to remove debris and reduce microbial contaminants in the preparation before placement of any restorative material. However, the oral antiseptic solutions commonly used for this procedure potentially cause either irreparable pulpal damage or staining. Light Emitting Diode (LED) light at 405 to 525 mm wavelengths has been shown to exert antimicrobial effect. This study determined the minimum exposure time for Light Emitting Diode (LED) light curing machine to reduce microbial contaminants on Class I cavity preparation. It aimed to compare the number of microbial contaminants before and after twenty, thirty, forty- second exposure to 480 nm wavelength LED light, using plate count; and to compare the relative decrease of microbial counts in the different exposure periods. Thirty teeth with Class I cavity preparations were included in the study, each exposure time having 10 samples. Right after a Class I cavity was prepared, pre-treatment microbial samples were obtained from the disto-pulpal angle after LED light exposure for 20, 30 and 40 seconds. All swabs were inoculated onto agar, incubated for 24 hours and plate counted. Results showed that microbial counts from the cavity preparations were generally lower after LED light treatment in all exposure times. Using one-tailed Student's t-test, the lowest exposure time that showed a significantly lower number of microbial count was at 30 seconds (mean = 51.29, SD = 67.98, p = 0.046), with an average percent reduction equal to 53.5 %. Results suggest that the minimum time required for the LED light curing machine to reduce microbial contaminants in Class I cavity preparation contaminants was at 30-second exposure. The researchers recommend that the LED light curing machine be used as an alternative method of cavity disinfection to using chemical substances.**

Keywords: **led light, cavity debridement.**

1. INTRODUCTION

Background of the Study

Cavity medication or disinfection is a common procedure after cavity preparation in order to reduce the number of bacterial pathogens in the preparation before placement of any restorative material. Studies have shown that if disinfecting agents-like silver nitrate, phenols or ethyl alcohol -are allowed to remain for a longer time in the cavity, it would result to irreparable pulpal damage.

Light Emitting Diode (LED) light specifically at the blue light spectrum had been extensively studied and results have shown that between 405-525 nm wavelengths, it is bactericidal. LED light curing machine is a readily available equipment in all dental clinics with the usual spectral distribution of 470-500nm. It has not been studied for its antibacterial effect.

Hence, this study is focused on LED light curing machine as an alternative for cavity disinfection agents.

Statement of the Problem

The study aimed to answer the question, "At what time of exposure does LED light curing machine have an antimicrobial effect on class I cavity preparation contaminants?"

Objectives

General: To determine the time LED light curing machine has an antimicrobial effect on Class I cavity preparation contaminants

Specific:

1. To count the number of microorganisms, present on a class I cavity preparation using colony count.
2. To measure and compare the number of microorganisms present in a Class I cavity preparation after a 20-second, 30-second, and 40-second light exposure.

Hypothesis

The LED light curing machine has an antimicrobial effect on a Class I cavity preparation contaminants.

Scope and Limitations

This study aimed to determine the exposure time at which an LED light curing machine would have its antimicrobial effect on a class I cavity preparation contaminants. The researchers tested the antimicrobial effect through three (3) different exposure times: 20 seconds, 30 seconds, and 40 seconds. Ten (10) prepared teeth were exposed for 20 seconds, another 10 for 30 seconds, and another 10 for 40 seconds, for a total of 30 teeth. The study is limited to a class I cavity preparation on any posterior tooth, regardless of depth. This was done in a pairwise comparison of treatment and control variable using only one tooth to obtain the pre-treatment sample (mesial wall) and the post-treatment sample (distal wall) after being exposed to COXO LED light curing machine (Model No. 686 DELI) emitting a wavelength of 420-480 nm and intensity of 1600 mW/cm^2.

Significance of the Study

The study would provide dentists an alternative method for cavity debridement which is safe for the patients because LED light curing machine is already utilized for curing composite restorations and convenient for the dentist because it is easily accessible and present in all dental clinics. It is also a non-chemical form of debridement which eliminates risk of allergies and pulpal damage.

2. LITERATURE REVIEW

After a cavity preparation, the dentin tubule lumen varying from 1 to 4 μm in diameter at varying distances between the dentino-enamel junction and the pulp, presents sufficient size for the entrance of microorganisms. (Roberson et. al, 2006) On a study conducted by Lado and Stanley (1987), individual microbial samples were aseptically collected from the base of deep carious lesions in each of six human teeth, the microorganisms present in these samples consisted predominantly of Lactobacillus, Actinomyces, and Streptococcus species. Juhl M (1983) also stated that pit-and-fissure caries has the highest prevalence of all dental caries. The pits and fissures provide excellent mechanical shelter for organisms and harbor a community dominated by S. sanguis and other streptococci.

In accomplishing the final procedures before insertion of the restorative material, disinfection of the preparation may be considered. Some operators place medicaments in preparations for disinfection purposes, based on empiric factors. (Roberson et. al, 2006) Simply swabbing the preparation with such agents as silver nitrate, phenol, or ethyl alcohol only leads to a false sense of security. If some of these agents were allowed to remain for a longer time, to permit penetration of the tubules, irreparable pulpal damage would result. (Muntz JA, et al., 1893) Extrinsic tooth stain is sometimes a side-effect observed with the chronic use of chlorhexidine-containing oral care products as observed both in humans and experimental animals. (Charbonneau and Snider, 1997)

Visible light lasers have been used as bactericidal agents to remove bacterial biofilms (Nandakumar et al., 2006). Several studies like that of Mclean et al. (2009) showed that bacterial pathogens, all commonly associated with hospital-acquired infections, when exposed to the 405-nm LED array, were successfully inactivated, with the general trend showing gram-positive species to be more susceptible than gram-negative bacteria. Another study conducted by Kim et al (2013) demonstrated that irradiation at 425 and 525 nm had bactericidal effects to S. aureus, E. coli, and P. gingivalis. On a study

conducted by Guffey and Wilborn, (2006) 405 and 470 nm blue light produce dose dependent bactericidal effects on Pseudomonas aeruginosa and Staphylococcus aureus. According to Vermuelen et. al (2007), visible wavelengths of 458 and 488 nm were able to produce significant mortality to E. coli cells but at 515 and 532 nm it did not produce a significant reduction. Sutton (2011) stated that, the general ranges in common acceptance for countable numbers of colonies on a plate are 30-300 and 25-250 and the United States Pharmacopeia (2011) recommends a range of 20 to 300 colonies, but not less than 6".

The emission spectra of the LED lights more closely mirrored the absorption spectrum of the commonly used photoinitiator camphorquinone which is between 450 and 500 nm. (Leonard et al, 2002) Radiometric and spectrophotometric analysis of third generation LED light curing units using different light curing meters showed that LED light curing units revealed a narrow spectral distribution from 470-500 nm. All LED light curing units were within the manufacturers specifications. (Owens and Rodriguez, 2007)

Feuerstein et al (2004) showed that blue light (400~500 nm) is phototoxic to *P. gingivalis* and *Fusobacterium nucleatum*. The minimal inhibitory dose for *P. gingivalis* and *F. nucleatum* was reported to be 16~62 J/cm. On a study conducted by Mclean et al. 2009, several bacterial pathogens like Staphylococcus aureus, Methicillin Resistant Staphylococcus aureus, Staphylococcus epidermis, Clostridium perfringens and Streptococcus pyogenes were all inactivated at 405 nm at a dose lower than 64 J/cm$^{2.}$ Guffey and Wilborn used treatment doses of 1, 3, 5, 10, and 15 Jcm2 doses in their study and the 470-nm light effectively killed Pseudomonas aeruginosa at all dose levels, but only killed Staphylococcus aureus at 10 and 15 J cm2. With this wavelength, as much as 96.5% and 62% reduction of Pseudomonas aeruginosa and Staphylococcus aureus was achieved, respectively (Kim S. et al, 2013) According to Craig and Powers, (2002), a typical resin composite requires an energy density of 16 J/s^2 (16,000 mW/cm^2 for 40 sec) for polymerization. Manufacturer specifications for the Coxo LED light curing machine is 1600 mW/cm^2 and has an equivalent dose of 64 J/cm^2.

At wavelength of 460 nm, photons can disrupt C-S bond (Vermuelen et. al, 2007) resulting to the removal of sulfhydryl groups which is a key component for coenzyme A (a major metabolic enzyme). LED's produce less heat and so it may have lower potential for gingival and pulpal irritation. (Leonard et al, 2002)

Based on the above literature, there is a need to conduct a study on the bactericidal effect of LED light curing machine as an alternative for disinfecting a cavity preparation.

Theoretical Framework

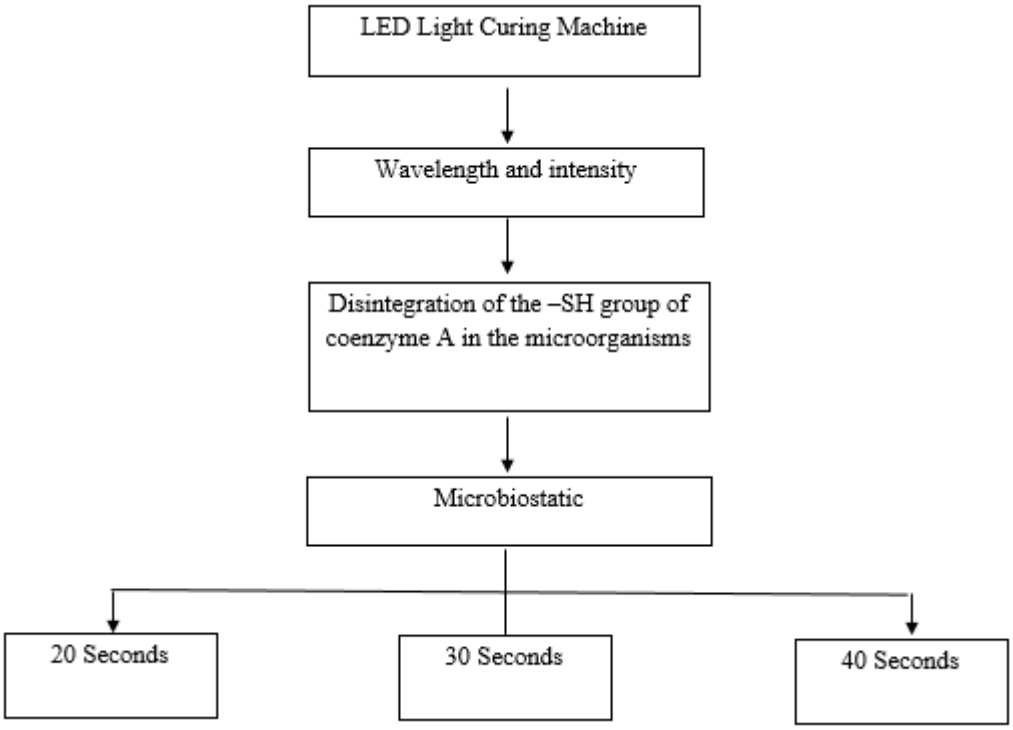

Fig 1

3. MATERIAL AND METHODS

Participants

Thirty (30) Class I cavity preparations done at the UE Dental Infirmary for adult patients.

Research Design

The study is of quantitative and experimental in nature since we utilized colony counting to determine the number of microorganisms present before and after exposure to LED light curing machine under controlled conditions.

Sampling

Purposive and quota sampling was utilized because the study only involved thirty samples of class I cavity preparations in the UE Dental Infirmary.

Preparation of treatments

Experimentation of the study used three (3) treatments with ten (10) replicates. The three (3) treatments used in the study will be as follows:

Pre-treatment: Untreated

- A_{20}, A_{30}, A_{40}

Treatment: Exposure to LED light

- B_{20} – 20-second LED light exposure
- B_{30} – 30-second LED light exposure
- B_{40} – 40-second LED light exposure

Instrumentation

The researchers used COXO Light Cure Machine (Model No. 686 DELI). Thirty (30) class I cavities were obtained from different patients at the University of the East Dental Infirmary. Sterile disposable petri dishes were bought from TKL Lab Supply and Brain-Heart Infusion Agar from Himedia Laboratories; sterilizing pouches from dental store; cotton and distilled water from the grocery. The researchers also used Erlenmeyer flasks, autoclave, digital weighing scale, refrigerator, and incubator of University of the East College of Dentistry. Sterilized custom-made cotton swabs were also utilized in the study.

Procedure

The researchers selected thirty (30) teeth with class I cavity. After cavity preparation, the cavity was dried and isolated and a microbial sample was gathered using a cotton swab. For the first 10 samples, the researchers swabbed the cavity on the mesial side as a pre-treatment right after cavity preparation then, inoculated the swab onto a spread plate, and counted the colonies present after incubation at 37^{o}C for 24 hours. The cavity was then exposed to LED light curing machine for 20 seconds, and then the researchers swabbed the cavity on the distal side as a post-treatment, inoculated the swab onto a spread plate, and counted the colonies present after incubating for 24 hours. For the next 10 samples, the researchers swabbed the cavity on the mesial side as a pre-treatment right after cavity preparation then, inoculated the swab onto a spread plate, and counted the colonies present after incubating at 37^{o}C for 24 hours. The cavity was then exposed to LED light curing machine for 30 seconds, and then the researchers swabbed the cavity on the distal side, as a post exposure, inoculated the swab onto a spread plate, and counted the colonies present after incubating at 37^{o}C for 24 hours. For the remaining 10 samples, the researchers swabbed the cavity on the mesial side as a pre-treatment right after cavity preparation then, inoculated the swab onto a spread plate, and counted the colonies present after incubating for 24 hours. The cavity was then exposed to the LED light curing machine for 40 seconds, and then the researchers swabbed the cavity on the distal side, as a post-treatment, inoculated the swab onto a spread plate, and counted the colonies present after incubating at 37^{o}C for 24 hours. After the researchers counted the number of colony-forming units, the former recorded the results.

Gathering of Data and Statistical Analysis

To determine the antimicrobial activity exhibited by LED light curing machine at different exposure times on microbial contaminants in a cavity preparation, plate count was performed to obtain quantitative results and the results were tabulated. Plates that showed less than 30 and more than 300 colonies were excluded from the results.

Students' T-test was utilized to determine the significance of the antimicrobial activity of LED light curing machine on cavity preparation contaminants at various exposure times.

Ways of Proper Disposal

After the analyses and experimental activities, the petri dishes were turned over to the University of the East College of Dentistry biochemistry section for proper disposal of the bacterial specimens. The researchers disposed the cotton swabs and sterilizing pouches on infectious waste bins at the UE Dental Infirmary. Any borrowed materials were sterilized and returned to the laboratory.

4. RESULTS

The results of the experimentation on the antimicrobial effect of LED light curing machine on class one cavity preparation contaminants is shown in Table 2 (refer to page 21)

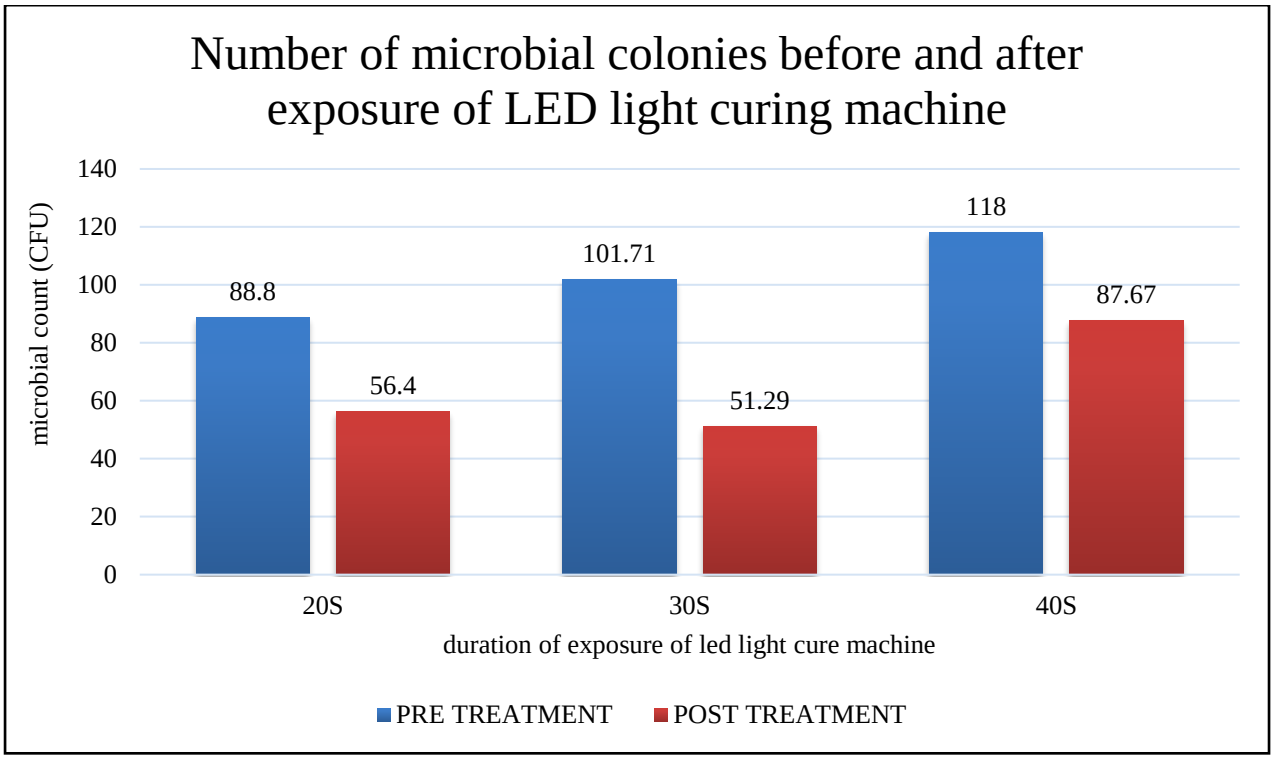

Figure 1: Graph showing the average mean of the number of microbial colonies before and after exposure of LED light curing machine.

The mean number of microbial colonies after 30 sec. exposure (mean= 51.29, SD=67.98) was lower than the pretreatment sample (mean 101.71, SD 84.82). The same is also true for the 40 sec. exposure (mean=87.67, SD=90.13) with lower values than the pretreatment sample (mean= 118, SD=100.90). Statistical analysis using one-tailed Students T-test showed the difference to be significant at $p \leq 0.05$ where p value of 30s is 0.05 and p value of 40s is 0.04. (refer to table 2 on page 21).

5. CONCLUSION

Results showed that microbial counts from the cavity preparations were generally lower after LED light treatment in all exposure times. Using one-tailed Student's t-test, the lowest exposure time that showed a significantly lower number of microbial counts was at 30 seconds (mean = 51.29, SD = 67.98, p = 0.046), with an average percent reduction equal to 53.5 %

With this we can suggest that the minimum time required for the LED light curing machine to have a microbiostatic effect on Class I cavity preparation contaminants is at 30-second exposure. Moreover, there is an improved microbiostatic effect on the contaminants at 40-second exposure.

The researchers recommend that the LED light curing machine be used as an alternative method of cavity debridement rather than using chemical substances. Furthermore, extensive research could be done to test the antimicrobial effect of the LED light curing machine with more samples and at a longer exposure time.

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