EFFECT OF ELECTRONIC CIGARETTES SMOKING ON COLOR STABILITY OF CAD/CAM ACRYLIC RESIN: IN VITRO STUDY

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ABSTRACT

INTRODUCTION: The most common electronic nicotine delivery system is electronic cigarettes. Most of electronic cigarettes consist of a battery and electric heater used to aerosolize a liquid to produce vapor which the user inhales. Poly methyl methacrylate comes in contact with a variety of foods and beverages of different temperatures, as a result, it is prone to absorbing a variety of pollutants, altering its physical structure and appearance.

OBJECTIVES: This study aimed to investigate the effects of electronic cigarettes smoking on color stability of CAD/CAM acrylic resin.

MATERIAL AND METHODS: This study was in vitro study in which sixty specimens of CAD/CAM acrylic resin was exposed to aerosols which were flavored and non-flavored with nicotine content (0 mg, 6 mg and 12 mg). A portable suction machine was modified to simulate smoking in vivo. Each specimen was exposed to 20 cycles of aerosol released by a single ECIG device using the smoking chamber. Color measurements were performed using a calibrated spectrophotometer on the specimens before and after ECIG exposure

RESULTS: Specimens exposed to flavorless 0 mg. nicotine aerosol showed the lowest delta E value 2.39 (±0.16), while Specimens exposed to flavored 12 mg. nicotine showed the highest values 4.14 (±0.19). No significant difference in color change found between groups exposed to flavorless e-liquid, while a significant difference found between flavored 0 mg. nicotine and 12 mg. nicotine groups (P = 0.002). Also, a significant difference found between flavored 6 mg. nicotine and 12 mg. nicotine groups (P = 0.002). According to Two Way ANOVA, both flavor and nicotine contents have a significant effect on color change (P < 0.0001).

CONCLUSION: Exposure to flavored and non-flavored electronic cigarettes aerosol of different nicotine concentrations changed the color of acrylic resin. Both flavor and nicotine contents have a significant effect on color change.

KEYWORDS: E-cigarette, Vape smoking, Acrylic resin, Color change, CAD/CAM, E-liquid.

RUNNING TITLE: Electronic cigarettes smoking effect on acrylic resin discoloration.

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INTRODUCTION

Although smoking prevalence has declined since 1990, population expansion has resulted in a significant increase in the overall number of smokers. In 2019, there were 1.14 billion smokers worldwide who used 7.41 trillion cigarette-equivalent of tobacco. (1) Many electronic nicotine delivery systems were introduced as smoking cessation tools.(2)

Electronic nicotine delivery devices have grown in popularity, with electronic cigarettes (ECIG) being the most common.(3) As smoking rates in many developed nations continue to decline or plateau, international tobacco companies have shifted their focus to the vaping business. Since the first commercialized product was produced in China in 2003, ECIG has seen a rapid increase in

2003, ECIG has seen a rapid increase in popularity.(4)

The majority of ECIGs are made up of a battery and an electric heater that is used to aerosolize a liquid (eliquid) into a vapor that is inhaled by the user.(5) An e-liquid is a mixture of nicotine, a solvent (typically propylene glycol and/or vegetable glycerin), and flavors (neutral, tobacco, menthol, candy, and others). It comes in a variety of nicotine doses as well as nicotine-free options.(6) Even though ECIG was promoted as a safe alternative to conventional cigarettes, they contain hazardous components; which lead smokers who are trying to quit smoking to raise questions about the advantages and disadvantages of using it.(7) Literature reveals that ECIG aerosol and e-liquid components induce and promote chronic inflammatory conditions.(8)

For decades, polymer materials have been in dental treatment. Temporary crowns and bridges, occlusal appliances, complete denture fabrication, as well as overdentures, are all examples of applications where the polymer base is employed to replace missing soft and hard tissues.(9)

Polymethyl methacrylate (PMMA) comes in contact with a variety of foods and beverages of different temperatures, as a result, it is prone to absorbing a variety of pollutants, altering its physical structure and appearance. Also, since acrylic resin represents organic materials, its transparency and color may deteriorate. Discoloration or color deterioration is an aesthetic issue that is viewed critically from the perspective of patient acceptance and poses challenges to the workability and skill of the prosthesis.(10)

Complete dentures, record bases, immediate dentures, and implant-supported overdentures can now be made in two clinical appointments using computeraided design/computer-aided manufacturing (CAD/CAM) technology.(11) CAD/CAM technology is employed in the production of prostheses because of its benefits, which include enhanced efficiency, automation, and accuracy throughout the treatment process.(12)

For denture milling that are colored and polished like traditional dentures, CAD/CAM PMMA has been the material of choice. When CAD/CAM PMMA dentures were compared to traditional heat-cured PMMA dentures, the strength and surface roughness attributes of CAD/CAM PMMA were found to be superior, indicating a more durable denture.(13-15)

Many studies aimed to explore the effects of Conventional cigarettes smoking on the color stability of dental materials; Conventional cigarette smoke causes significant color changes in artificial dentures, which are visually identifiable and considered clinically unacceptable.(3)

While the effects of conventional cigarettes on the teeth and dental prosthesis as regard to color change have been studied, the effects of ECIGs using have not been studied in detail.

Therefore, the aim of this study was to investigate the effects of ECIG smoking on the color stability of CAD/CAM acrylic resin. The null hypothesis was that ECIG smoking would have no effect on the color stability of CAD/CAM denture acrylic resin.

MATERIAL AND METHODS

Study design

This study was in vitro experimental study investigating the effects of ECIG smoking on the color stability of acrylic resin. In this study, the acrylic resin was exposed to aerosols that were flavored and non-flavored with nicotine content of 0 mg, 6 mg, and 12 mg. The study was approved by the Research Ethics Committee of the Faculty of Dentistry of Alexandria University, Egypt to be executed (IORG 0008839) according to the declaration of Helsinki (2013).

Randomization

A total of sixty discs were randomized with an equal allocation ratio with a block size of 6 using random allocation software. To assign samples to one of six categories according to the flavor and nicotine concentrations. A trial independent individual created a computer-generated randomization list that was maintained in an opaque sealed envelope till the commencement of the study.

Sample size estimation

The sample size was estimated based on assuming 5% alpha error and 80% study power. The mean enamel color alteration (ΔE) in the unflavored group with 0 and 12 mg of nicotine was 2.40 and 1.90 while the mean ΔE of 6 mg of nicotine was calculated to be 1.7. The mean ΔE of the flavored group was 4.60 and 2.40 for 0 and 12 mg of nicotine. The mean of 6 mg flavored nicotinic E-cigarettes was calculated to be 0.7. Higher variability was expected to be shown on resin materials and the highest SD of 2.10 reported by Pintado-Palomino et al (16) was used. Sample size was calculated to be 8 specimens per group which was increased to 10 specimens to make up for processing errors. Total sample size = Number per group \times Number of groups = $10 \times 6 = 60$ specimens. Sample size was based on Rosner's method calculated by Gpower 3.0.10.

Specimen preparation

Total numbers of (60) specimens were constructed from acrylic resin discs (Lucitone 199® Denture Base Disc, Dentsply Sirona, USA) with a minimum thickness of 2mm to resemble the thickness of the denture base clinically. (17) The acrylic resin specimens of a diameter of 10 mm and thickness of 2 mm were prepared by CAD/CAM (ED5X, EMAR, Egypt). The acrylic specimens were polished and treated in the same way as an actual acrylic resin denture base would be. (Fig. 1)

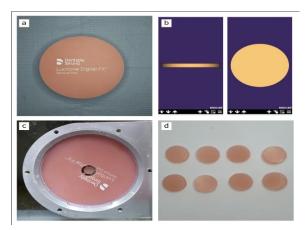


Figure 1: Specimen preparation. **a** CAD/CAM PMMA blank; **b** Specimen design; **c** Specimen milling; **d** Finished specimens.

E-Liquid preparation

Nicotine e-liquid formulations were prepared at (0, 6 and 12 mg/mL) in 50:50 propylene glycol: vegetable glycerin with and without flavor.

Smoking chamber

A portable suction machine (H003-c, Folee, China) was modified to receive specimens and to simulate smoking in vivo.

A perforated metal base was constructed to receive specimens. The base permits a metal pipe, which is attached to negative pressure of the suction machine, to penetrate it. The inlet of the suction machine was attached from inside with a custom made distributor with openings directly opposite to the specimens' positions. (Fig. 2)

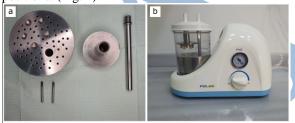


Figure 2: a components of smoking chamber; b Smoking chamber.

Exposure to ECIG aerosol

Sixty specimens were exposed to 20 cycles of aerosol released by a single ECIG device (Vboy 222W Box Mod, GTRS, China) using the smoking chamber. The ECIG device was used in this study contains a cartridge (Cascade Baby SE Tank, Vaporesso, China) that was filled entirely with e-liquids. The batteries of the ECIG were charged completely before each experiment. The ECIG was then connected to the smoking machine through a silicon tube. Specimens were placed in front of the ECIG's aerosol inlet inside the chamber. Negative pressure was applied by the vacuum system.(16)

Each cycle consists of 10 puffs with duration of 4 seconds and an interpuff interval of 20 seconds, for a total of 3.6 minutes. To avoid overheating, the liquid was resupplied after 10 cycles.(16)

Color measurement

Color measurements were performed using a calibrated spectrophotometer (VITA Easyshade, VITA, Germany) on the specimens before and after ECIG exposure. Specimens were subjected to gentle washing with distilled water after 200 puffs for 1 minute, dried with absorbent paper and final color measurement was performed. (16)

The color evaluation was based on a threedimensional CIE L*a*b* color system established by the "Commission Internationale de l'Eclairage-CIE." According to this system, L* indicates lightness, a* indicates green-red, and b* indicates yellow-blue.(18) Three consecutive measurements without replacement were taken for each parameter and the mean after measurements was considered the baseline value. The CIELab color difference ΔE was calculated as follows: $\Delta E = [(\Delta L^*) 2 + (\Delta a^*) 2 +$ $(\Delta b^*)2]1/2$; where ΔL^* , Δa^* , and Δb^* correspond to the difference between final and initial L*, a*, and b* values, respectively.(16)

The discoloration of acrylic resin was measured with a spectrophotometer in this study, which is more ideal for standardization and numerical expression than visual evaluation.(18) ΔE values are used to define discoloration in dental materials, and an ΔE value of more than 3.5 is considered undesirable clinically.(19)

Statistical analysis

Data were analyzed using SPSS version 24.0 for Windows (IBM Corporation, Armonk, NY, USA). Normality was checked using Shapiro Wilk test, box plots and descriptive. Mean color change was normally distributed and presented using Mean and Standard deviation (SD). Two Way ANOVA was applied to assess the effect of flavor and nicotine content and their interaction effect on color change. Estimated Marginal Means, Adjusted R Squared values were calculated. Pairwise comparisons were done to assess differences in delta E between groups.

RESULTS

The data presented in figure (3) shows Mean ΔE and Standard deviation of the CAD/CAM PMMA discs according to flavor and nicotine content. Regarding specimens which exposed to flavorless e-liquid, delta E values increased through groups with 0, 6, 12 mg nicotine respectively; 0 mg. nicotine group showed the lowest delta E value 2.39 (±0.16) of flavorless groups, while 12 mg. nicotine group showed the highest values 2.69 (±0.19).

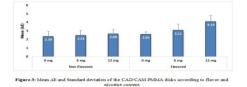


Figure 3: Mean ΔE and Standard deviation of the CAD/CAM PMMA disks according to flavor and nicotine content.

On the other hand, delta E values increased with wider range through groups exposed to flavored eliquid of 0, 6, 12 mg nicotine concentrations respectively. The group exposed to 0 mg. nicotine showed the lowest delta E value 2.65 (\pm 0.16) of flavored groups, while 12 mg. nicotine group showed the highest values 4.14 (\pm 0.19).

According to Two Way ANOVA, both flavor and nicotine contents have a significant effect on color change (P <0.0001). Flavor caused the color change with Partial Eta Squared (η 2) 0.348. There was a significant interactivity between flavor and nicotine concentration (P = 0.004). (Table 1)

The data in the table (2) shows the pairwise comparison between different nicotine concentrations according to flavor content. No significant difference in color change found between groups exposed to flavorless e-liquid, while a significant difference found between flavored 0 mg. nicotine and 12 mg. nicotine groups (P = 0.002).Also, a significant difference found between flavored 6 mg. nicotine and 12 mg. nicotine groups (P = 0.002).

Table 1: Two Way ANOVA assessing the effect of flavor and nicotine content on color change

	Mean	F test	P value	Partial Eta
	Square			Squared
Flavor effect	8.87	28.83	<0.0001*	0.348
Nicotine	4.15	13.48	< 0.0001*	0.333
effect				
Flavor *	1.86	6.04	0.004*	0.183
Nicotine				
effect				

R Squared=0.557, Adjusted R

Squared=0.516

*Statistically significant different at p value < 0.05

Flavor content	Nicotine Conc.	Compared to	Mean Difference	P value
Non- Flavored	0 mg Nicotine	6 mg Nicotine	0.123	1.00
		12 mg Nicotine	0.301	0.615
	6 mg Nicotine	12 mg Nicotine	0.179	1.00
Flavored	0 mg Nicotine	6 mg Nicotine	0.453	0.289
		12 mg Nicotine	1.484	0.002*
	6 mg Nicotine	12 mg Nicotine	1.030	0.002*

Table 2: Pairwise comparison between different

 nicotine concentrations according to flavor content

*Statistically significant different at *p* value≤0.05

DISCUSSION

This study evaluated the effects of ECIG smoking on the color stability of acrylic resin. A significant change in the color of acrylic resin specimens exposed to 12mg. nicotine flavored e-liquid aerosol was visually observable and considered clinically unacceptable. In contrast, all acrylic resin specimens exposed to other e-liquid formulations displayed a minute color change, which was considered clinically acceptable. Thus, the null hypothesis that ECIG smoking would not affect the color of acrylic resin was rejected.

When dental materials are exposed to staining agents like tea, coffee, or cigarette smoke, they discolor.(18) Resin color and surface roughness can be affected by both external stains caused by food, beverages, and intrinsic elements like the chemical composition of the material.(20-22)

Smoking cigarettes causes the combustion of elements, resulting in the emission of metals as well as dark smoke components. Cadmium, arsenic and lead have been detected on the resin composite and dental structures exposed to ordinary cigarette smoke. (3, 16) Color change happens as a result of pollutants impregnating the surface.(23) ECIG devices, unlike traditional cigarettes, do not burn or release smoke, but instead it produces an aerosol. Fibers that absorb e-liquid come into contact with heating elements that contain heavy metals in ECIG devices. ECIGs release trace elements such as nickel (Ni), lead (Pb), zinc (Zn), aluminum (Al), arsenic (As), cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), and manganese (Mn). These metals are produced in higher concentrations compared to the nonaerosolized e-liquid form. Furthermore, the concentration of some of these metals is higher in ECIG aerosol when compared to conventional

cigarettes smoke.(24) These could possibly be a cause of discoloration in samples exposed to ECIG aerosol.

In this study, Nicotine e-liquid formulations were prepared at (0, 6, and 12 mg/mL) in 50:50 propylene glycol: vegetable glycerin with and without flavor. The tobacco flavor is one of the most preferred flavors among ECIG users, therefore it was the flavor of choice in our study.(25)

According to Two Way ANOVA, both flavor and nicotine contents have a significant effect on color change (P <0.0001). Nicotine is colorless in its neutral condition, yet oxidized nicotine cause discoloration of teeth.(3) Due to the inherent alkalinity of nicotine, pH values for each e-liquid correlated with the measured total nicotine concentration.(26)

Besides nicotine, ECIG aerosol contains toxicants created by the thermal breakdown of e-liquid ingredients. Reactive oxygen species (ROS; mainly radicals and peroxides) can be found in ECIG aerosol.(6) Peroxides were found to cause damage to the acrylic resin; oxidation in combination with an alkaline solution can be deleterious.(27) Exposure of acrylic resin to alkaline solution was reported to cause surface pitting and formation of polymer beads.(28) Rough surfaces are more susceptible to discoloration than smooth surfaces.(29)

Another explanation for these findings is that the tobacco flavor used in the study was brown in color. This might explain the higher color change noticed in flavored groups.

The results of our study were consistent with a study held by Pintado-Palomino K., et al. where the effect of electronic cigarettes on dental enamel color has been studied.(16) Also, Chotimah C., et al. found that ECIG aerosol affects the color of acrylic teeth.(30) Alnasser H., et al. concluded that ECIG smoking changes the color of composite resins.(31)

According to the study conducted by Vohra F., et al. ECIG aerosol causes discoloration of ceramic and composite specimens. While ceramic specimens showed ΔE similar to those of our study, composite specimens showed much higher ΔE values.(32) These differences in results may be due to different types of material, the concentration of nicotine in e-liquid, the flavor used, or the method of exposure of specimens to ECIG aerosols.

There are some limitations for this study which includes that the experimental specimens have not been subjected to any means of cleaning like brushing and denture cleansers which are cleaning methods commonly used by dentures wearers because of their effectiveness in removing stains and deposits. Also, saliva, lips, and cheeks movement could remove stains and debris. Therefore, the change of acrylic resin color by ECIG intraorally could be less than that displayed in the present study. Moreover, ECIG devices differ broadly in regards to battery, and nicotine delivery which may affect particles distribution even within an established puffing protocol.

CONCLUSION

Based on the findings, it was concluded that exposure to flavored and non-flavored ECIG aerosol of different nicotine concentrations has a significant effect on the denture's CAD/CAM acrylic resin color. Higher concentrations of flavored nicotine had visually perceptible color changes and were deemed clinically unacceptable.

Conflict of interest:

The authors declare that they have no conflicts of interest.

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