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## Evaluation of resting mouth salivary flow rate and dental caries In E-cigarette users.

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### Abstract:

**Introduction:** Components of cigarette smoke are said to be amongst the first to affect the saliva and its production. Repeated exposure of tobacco smoke may result in functional changes of saliva.

**Objective:** To evaluate the effects of E-cigarette on resting mouth salivary flow rate and dental caries.

**Methodology:** This cross-sectional study conducted at Altamash institute of Dental Medicine for a duration of six months from Feb'2019 to Sept'2019. Data was collected using a well-structured and validated questionnaire. SPSS 22 was used to analyze the data. A p-value  $\leq 0.05$  was taken as significant.

**Results:** The mean score of e-cigarettes consumption per day and Clinical Oral Dryness score was found to be  $5.13 \pm 2.483$  and  $1.08 \pm 1.172$  respectively, which indicates mild dryness amongst the participants. Furthermore, the mean unstimulated salivary flow rate in participants was  $0.406 \pm 0.1027$  ml/minute. It was also observed that 247(68.61%) participants had carious lesion in one or more teeth.

**Conclusion:** E-cigarettes cannot be considered as a safe substitute to traditional cigarette smoking and it can have detrimental effects on oral health just like other tobacco products as results indicated that participants had mild to moderate dryness and were prone to carious lesion with reduced salivary flow rate.

**Keywords:** Caries, Saliva, Smoking, Xerostomia

### Introduction:

The salivary flow rate (SFR) is a great indicator of overall oral health status of an individual<sup>1</sup>. Nicotine in cigarette has been shown to be an immunosuppressive agent that make person vulnerable to multiple diseases.<sup>1,2</sup> Cigarette smoke has countless chemical components that may be described as possessing cytotoxic, mutagenic, carcinogenic and antigenic characteristics. These toxic ingredients of cigarette smoke rapidly disband in fluids of oral and airway epithelial lining, and are fully absorbed in the individual active or passive. Toxins such as Nicotine, Carbon monoxide,

ammonia, tar and formaldehyde which are known components of cigarette smoke are said to be amongst the first to affect the saliva and its production.<sup>2,3</sup> Research has proved that smoking plays a major role in the progression of periodontal diseases, which also has an effect on the attachment apparatus and alveolar bone loss compared with nonsmokers.<sup>4</sup> In addition to this, smokers have the tendency to allow the colonization of gram-negative bacteria in their oral cavity and promotes the adhesion of Streptococcus mutans and Candida albicans to the acquired pellicle on tooth surface. Streptococcus mu-

tans is aciduric, has acidogenic tendency, high association with dental caries and is highly related to the progression of caries.<sup>5</sup>

An electronic cigarette is a battery-operated device that emits a vaporized solution to inhale. It simulates tobacco smoking.<sup>6</sup> It was initially thought that e-cigarette caused fewer negative effects in comparison to traditional ones.<sup>7</sup> But further studies confirmed that its cartridges contain up to 24 mg/mL of nicotine extracted from tobacco<sup>8</sup> which is enough to generate oxidative stress and an upsurge of pro-inflammatory cytokines causing cytotoxic changes which in turn can lead to the death of oral epithelial keratinocytes and periodontal fibroblasts.<sup>3, 8-10</sup>

Ambiguity exist in the literature regarding the effects of E-cigarettes on the oral microbiome as few in vitro studies showed that it has an indefinite effect on the growth and survival of oral streptococci when compared with traditional cigarette smoking.<sup>11</sup> On the contrary, literature also proved that its exposure can cause transcriptional changes in the related species without affecting their virulence properties.<sup>12</sup> There is scarcity and inadequacy of the data and the clinical implications of these effects and risk for oral disease have yet to be determined.

**Objective:**

This study was undertaken to evaluate the consequences of E-cigarettes smoking on SFR and dental caries.

**Methodology:**

This prospective study was conducted for duration of six months from Feb'2019 to Sept'2019 at Altamash institute of Dental Medicine after approval from Institutional Ethics and review committee (AIDM/EC/01/2019/03). The sample size of 360 participants was calculated using WHO sample size calculator, by taking statistics for SFR as  $2.94 \pm 1.38$  ml/min<sup>13</sup>, margin of error as 0.184 and 95% confidence level. Non-probability convenience sampling technique was used. Patients that were affected in any way with pre-existing dental conditions, using medications, e.g., steroids, beta blockers, anti-psychotics or anti-depressants; that might interfere with the natural salivary flow; were excluded from the sample set. In addition, pregnant women and individual using dentures were also excluded. The purpose of the study explained to each participant and informed consent ob-

tained. A self-administrated and well-structured questionnaire was used to gather the demographic details, inclusive of subjects' report about presence of sensation of unlikable taste or halitosis, along with the frequency and duration of smoking.

The participants were given few instructions before the start of the procedure including; avoidance of smoking, eating, drinking or performing any oral hygiene measure 60 minutes prior to the collection of the sample. They were asked to be seated on a dental chair in an upright position with their head prone forward so that the production of saliva is collected in floor of mouth. After thorough oral examination, each participant was requested to spit in the measured vessel every 1 minute for 5 minutes and SFR measured in ml/min. The caries index was measured by adding the values of decayed teeth. The clinical oral dryness (COD) score<sup>14</sup> used in the present study, consisted of a 10-point scale, each representing a feature of dryness in the mouth.

SPSS-22 was used for data analysis. Mean and SD was calculated for quantitative variables. Qualitative variables including gender and smoking status were reported as frequency and percentage. Independent t-test was used to compare SFR, COD score between active smokers and nonsmokers. Chi-square test was used to compare active caries with active smokers and nonsmokers. The effect of modifiers like age and gender were addressed through stratification. P value of <0.05 was considered statistically significant.

**Results:**

Out of the total (360) participants, 170 (47.22%) were males and 190 (52.77%) females. The mean age of participants was  $33.18 \pm 6.442$ . It was observed that 150 (41.66%) were active e-smokers while 210 (58.33%) were nonsmokers. The mean score of e-cigarettes consumption per day was found to be  $5.13 \pm 2.483$ . The mean COD score was  $1.08 \pm 1.172$ , which indicates mild dryness amongst the participants. In addition, 104 (28.9%) participants opted for Score 1 "mirror sticks to buccal mucosa". Score number 2 "mirror sticks to tongue" was evident in 53 (14.7%) participants. While COD score 3 "saliva frothy" was prevalent in 40 (11.1%) and score 4 "no saliva pooling in mouth" was found in 15 (4.2%) of participants. Whereas in 148 (41.1%) participants no signs of dryness were noted.

Moreover, the mean unstimulated SFR in participants was  $0.406 \pm 0.1027$  ml/minute, which indicates a normal SFR amongst participants. The carious lesion in teeth was found in 247(68.61%) participants while 113(31.38%) had no carious lesion. Furthermore, the carious lesion in SFR range of 0.2-0.4 was found in 203 (56.38%) and for 0.5-1 range, it was 44 (12.22%) which indicates that the participants were more prone to carious lesion with reduced salivary flow rate as shown in table 1. The carious lesions were observed in 171 (69.51%) e-cigarette smokers out of 246 participants while the rate of carious lesion was 76 (66.66%) out of 114 non-smokers. Furthermore, no carious lesion was seen in 157 (43.61%) participants with 0.2-0.4 SFR and 316 (87.77%) with 0.5-1 SFR. However, 75 (30.48%) had no caries with e-smoking habit while 38 (33.33%) had no caries in non-smokers. The frequency of caries was more with e-smokers compare to non-smokers is depicted in Table 1.

**Table 1: Distribution of carious and non-carious teeth with respect to SFR and e-cigarette smoking**

Variable	SFR (0.2-0.4)	SFR (0.5-1)	Smoker	Non-smoker
Carious	203 (56.38%)	44 (12.22%)	171 (69.51%)	76 (66.66%)
Non-carious	157 (43.61%)	316 (87.77%)	75 (30.48%)	38 (33.33%)

*SFR: Salivary flow rate*

Moreover, the COD score 1 with respect to SFR (0.2-0.4) and SFR (0.5-1) was observed in 82 (78.84%) and 22 (21.15%) participants respectively. Whereas the COD score 2 with respect to SFR (0.2-0.4) and SFR (0.5-1) was noted in 52 (98.11%) and 1 (1.89%) in participants respectively. COD score 3 of SFR (0.2-0.4) was noted in 40 participants while COD score 4 of SFR (0.2-0.4) was noted in 15 participants as shown in table 2. The COD score 1 with respect to 1-5 cig / day was noted in 21 (72.41%) participants while COD score 1 with respect to 6-11 cig / day was recorded in 8 (27.58%) participants. The COD score 2 with respect to 1-5 cig / day and 6-11 cig / day was recorded in 9 (31.03%) and 20 (68.96%) participants respectively whereas the COD score 3 with respect to 1-5 cig / day and 6-11 cig / day was noted in 4 (33.33%) and 8 (66.66%) participants respectively. Finally, the COD score 4 with respect to 1

5 cig / day and 6-11 cig / day was noted in 6 (50%) participants each respectively as depicted in table 2.

**Table 2: Comparison of COD to SFR score and number E-cigarette smoking**

COD	SFR (0.2-0.4)	SFR (0.5-1)	1-5 cig / day	6-11 cig / day
Mirror sticks to buccal mucosa	82 (78.84%)	22 (21.15%)	21 (72.41%)	8 (27.58%)
Mirror sticks to tongue	52 (98.11%)	1(1.89%)	9 (31.03%)	20 (68.96%)
Saliva frothy	40	0	4 (33.33%)	8 (66.66%)
No saliva	15	0	6 (50%)	6 (50%)

*COD: Clinical oral dryness, SFR: Salivary flow rate*

Additionally, a significant difference (chi square;  $p=0.001$ ) between male and female smokers was noted. Male were 102 (68%), female smokers 48 (32%). A significant difference was also found in COD scores of both sexes (chi square;  $p=0.001$ ) and amongst male and female participants in terms of caries frequency (chi square;  $p=0.001$ )

#### **Discussion:**

Electronic cigarettes have gained popularity due to their use as an initial step towards quitting cigarette smoking. In addition, they are assumed to do less harm to health as compared to traditional cigarette smoking.<sup>15</sup> E-cigarettes basically use aerosols by heating e-liquids and by-passing the traditional combustion used in tobacco cigarettes. In our study we assessed the mean unstimulated salivary flow rate in participants and found that it was within normal limits ( $0.406 \pm 0.1027$  ml/minute). This finding is contrary to studies which have reported decreased SFR with increasing frequency of tobacco use.<sup>1, 16</sup> Whereas, Hijjaw O et al. observed a slightly higher mean stimulated whole SFR as  $0.46 (\pm 0.44)$  mL/5min as compared to our study.<sup>17</sup> Literature proves that e-cigarettes indirectly modulates the oral microbiome by altering the levels of antimicrobial proteins and cytokines in saliva.<sup>18, 19</sup> In addition, studies report that consistent exposure to contents of tobacco especially nicotine does not only exert a vast difference in the pattern of salivary secretion<sup>20, 21</sup>, but also may cause morphological and functional anomalies of the

salivary glands.<sup>21</sup> It stimulates the sympathetic nerves to produce neurotransmitters, including catecholamine causing vasoconstriction which in turn, can decrease the saliva secretion and other functions of salivary gland, along with a reduction in bicarbonate ions as well. This is further supported by a study by Fitriyani et al, which showed that higher nicotine content in conventional cigarettes may cause a reduction in production of saliva.<sup>22</sup> Lesser duration of e-cigarette use or lower amount of absorbed nicotine in our study population may be the reason of an insignificant effect on the salivary flow rate.

The carious lesion in SFR range of 0.2-0.4 was found in 56.38% of participants whereas in SFR of 0.5-1, 12.22% of participants showed carious lesions. This finding indicates that the participants with lower SFR were more prone to carious lesion. A significant difference ( $p=0.001$ ) was seen amongst male and female participants in terms of caries frequency. The carious lesion was seen in 69.51% e-cigarette smokers out of 246 participants while the rate of carious lesion was 66.66% out of 114 non-smokers. Association of caries with tobacco smoking is consistent with literature.<sup>5,23</sup> In-vitro studies have reported that nicotine induces vasoconstrictor effects on the blood vessels of gingiva in addition to the oxidative stress and causes a surge of destructive-inflammatory cytokines.<sup>24,25</sup> Ebersole J et al suggested that critical changes in oral microbiome may be induced by e-cigarettes, facilitating development of carious lesions.<sup>26</sup> The increased propensity to develop caries due to tobacco exposure may be due to stimulation of oral cavity by smoke that changes blood flow and decreases salivary flow. The resultant dry mouth may promote anaerobic conditions in oral cavity, making it easier for anaerobic bacteria to grow in plaque.<sup>27</sup> Another reason for high frequency of carious lesions in e-cigarette smoker's may be low salivary pH which is caused by the growth of acid producing bacteria.<sup>28-30</sup> These bacteria metabolize in low pH and induce demineralization of dental hard tissues.<sup>28</sup>

In this study, the association of dry mouth with e-cigarette was assessed using COD score. It was observed that clinical oral dryness score (COD1 and 2) was prominent in reduced SFR (0.2-0.4 range). Mor

over, COD score 2 and 3 were associated with increased use of e-cigarette per day. This finding between COD score and SFR is in agreement with study of Correia et al<sup>30</sup> that mentioned significant negative correlation ( $r=-0.515$ ,  $p<0.05$ ) between clinical oral dryness score and salivary flow. It has been showed that patients with no oral dryness had an increased rate of unstimulated SFR  $>0.5\text{ml}/5\text{min}$  while patients with moderate dryness had decreased rate of unstimulated SFR  $>0.1\text{ml}/5\text{min}$  ( $p<0.01$ ).<sup>30</sup> Moreover, a weak negative correlation between COD score and reduced salivation group ( $r=-0.33$ ,  $p<0.01$ ) and moderate correlation between COD score and adequate & high salivation groups was noted respectively ( $r=-0.56$ ,  $p<0.01$  &  $r=-0.55$ ,  $p<0.01$ ) by Jager et al.<sup>31</sup> A limitation of our study is small sample size which was due to our strict inclusion criteria. For example, only those E-cigarette users were included who were solely vaping and had never consumed any other form of tobacco in the past. Saliva itself has specific limitations as a suitable sample for analytical purpose, as the SFR and salivary pH due to certain other reasons.

#### **Conclusion:**

E-cigarettes can have detrimental effects on oral health just like other tobacco products as results indicated that participants had mild to moderate oral dryness and were prone to carious lesion with reduced salivary flow rate. Dental practitioners and other community health workers should make an effort to create awareness about the harmful effects of electronic cigarettes.

#### **Conflict of interest:**

None

#### **Financial Disclosure:**

None

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