

# Long-term effect of tobacco on resting whole mouth salivary flow rate and pH: An institutional based comparative study

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## ABSTRACT

**Background:** Saliva is the first biological fluid that is exposed to tobacco and is responsible for its changes especially the salivary flow rate (SFR) and salivary pH. **Aims and Objectives:** The aim of this study was to analyze and compare the long-term effects of tobacco on SFR and pH between tobacco chewers, smokers, and controls. **Materials and Methods:** A total of 60 Subjects were divided equally into tobacco smokers (group A), chewers (group B), and controls (group C). Saliva of each subject was collected under resting condition and SFR was expressed in mL/min for 10 min. Salivary pH was determined using the specific salivary pH strips. **Results:** The mean ( $\pm$ SD) SFR for group A; 4.34 ( $\pm$ 0.3), group B; 3.07 ( $\pm$ 0.26) and group C; 5.65 ( $\pm$ 0.44) mL/min when compared and a significant relation was obtained. The mean ( $\pm$ SD) pH for group A; 6.8 ( $\pm$ 0.1), group B; 6.7 ( $\pm$ 0.1) and group C; 7.04 ( $\pm$ 0.1) when compared and a non-significant relation was obtained though, lower salivary pH were observed in group A and B. **Conclusion:** Present study indicates that the SFR decreases appreciably among tobacco abusers especially more among smokeless form. A lower (acidic) salivary pH was observed in tobacco users as compared with control. These alterations in SFR and pH due to long-term effect of tobacco user can render oral mucosa vulnerable to various oral and dental diseases.

## Key words

Saliva, salivary flow rate, salivary pH, tobacco

## INTRODUCTION

The oral cavity is kept moist by a film of fluid called saliva that coats the teeth and the mucosa.<sup>[1]</sup> It is a complex and important body fluid, which is very essential for oral health.<sup>[2]</sup> It is the most easily accessible fluid in the human body and in the future it is probable that it will provide an easy tool for non-invasive measurements of various body parameters.<sup>[3]</sup>

Salivary parameters are supposed to be altered by drugs such as anti cholinergics, diuretics, antihistaminics, antihypertensive agents and psychoactive substances and conditions such as post-surgery, metabolic, nutritional, neurological abnormalities and hydration status.<sup>[4]</sup> Altered salivary gland function could be associated with

oral, pharyngeal, esophageal, neoplastic, metabolic nutritional inflammatory, genetic, autoimmune, nervous system disorders and require early diagnosis and intervention.<sup>[2,5]</sup> Alterations in salivary flow rate (SFR) and pH have a significant impact on oral and dental health and can be used for the diagnosis of a wide range of diseases.<sup>[2,4,5]</sup>

Based on the clinical and epidemiological evidence adverse effects of tobacco on oral health is already been established.<sup>[6,7]</sup> The main ingredient of tobacco is nicotine, which acts on certain cholinergic receptors in the brain and other organs causing neural activation leading to altered salivary secretion.<sup>[8]</sup> There are several studies concerning the effect of chewing tobacco and smoking on salivary secretion. Although some of these studies have shown an increase in SFR, especially in the short term.<sup>[7-9]</sup> Other Studies have shown that SFR remains unaffected with long-term tobacco use.<sup>[10]</sup> Though, long-term effect of tobacco use on SFR and pH is still unclear. Given the paucity of literature on the influence of tobacco use on SFR and pH, the present study was undertaken to analyze and compare the long-term effect of tobacco on SFR and pH in tobacco chewers, tobacco smokers, and control.

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## MATERIALS AND METHODS

Subjects in the present study comprised of 60 healthy adults, divided into 3 groups (20 each). Group A and B comprised of subjects consuming the tobacco for more than 10 years.

Group A: Smoked form (10 males and 10 females)

Group B: Smokeless form (10 males and 10 females)

Group C: Healthy control (10 males and 10 females).

The exclusion criteria included were age over 40 years, alcohol consumption, combination of tobacco (smoke and smokeless form), history of any other habits, history of trauma to the head and neck, denture wearers, pregnant and post-menopausal women, history of radiotherapy, patients with systemic or salivary gland diseases or under any drug therapy and patients with any lesions in the oral cavity.<sup>[2,7,11,12]</sup>

After obtaining informed written consent a through case history was taken followed by careful oral examination. Saliva of each subject was collected under resting condition and SFR was expressed in mL/min for 10 min. Salivary pH was determined using specific salivary pH strips.

### Saliva collection

Saliva collection was carried out between 9:00 am and 12:00 pm to avoid diurnal variation. Each subject was requested not to eat, drink or perform oral hygiene or chew or smoke 60 min before and during the entire procedure. Subjects were then seated in the dental chair and asked to spit in a graduated container every 1 min for 10 min [Figure 1]. During saliva collection subjects were instructed not to speak or swallow. After collection, the SFR was measured and expressed in mL/min for 10 min.<sup>[2]</sup>

Salivary pH was measured immediately after

measuring SFR using the Dental Salivary pH Indicator strips (pH 4.5-9.0, iGen pH test strips, China) [Figure 2]. Based on the color change of the indicator paper strip, the pH was assessed in comparison with a color chart. Manufacturer's instructions were followed while measuring salivary pH.

## STATISTICS

Data were analyzed using the Statistical Package for Social Service (SPSS) computer software. Student *t*-test, ANOVA and Z-test were applied to assess between group differences. *P* value of less than 0.05 was considered as statistically significant. Significance level of 0.05 and Confidence of 95% was considered.

## RESULTS

The subjects in our study were present in the age group of 30-40 years. The mean age ( $\pm$ SD) in the group A, was 36.85 ( $\pm$ 0.77), group B- 35.55 ( $\pm$ 0.56) and group C- 34.55 ( $\pm$ 0.85), when compared a non-significant difference was obtained ( $F=1.341, P=0.318$ ). Group A and B subjects consume tobacco for more than 10 years, with the Mean ( $\pm$ SD) duration, consumption and frequency of habit; 12.05 ( $\pm$ 1.16), 9.9 ( $\pm$ 0.8 pieces/day) and 8.75 ( $\pm$ 0.89) in group A and 10.1 ( $\pm$ 0.84), 1.18 ( $\pm$ 0.17 packets/day) and 7.6 ( $\pm$ 0.79) in group B respectively.

A non-significant relation was obtained when the mean salivary pH for groups were compared [Table 1 and Figure 3]. Whereas, a significant relation was obtained the mean SFR for groups were compared [Table 1 and Figure 4]. Moreover, a significant relation was obtained when SFR and pH were compared between the groups [Table 2].



Figure 1: Saliva collection



Figure 2: Salivary pH strips

**Table 1: Individual comparison of age, salivary flow rate (mL/min) for 10 min and salivary pH between the groups**

Groups	Age mean±SD	Mean salivary flow rate (mL/min)	Mean salivary pH	ANOVA age* flow rate* pH	ANOVA age* flow rate	ANOVA age* pH
Group A	36.85±0.77	4.34±0.35	6.8±0.11	$F=1364.4$ , $P=7.27E-49$ , NS	$F=1.610$ , $P=0.234$ , NS	$F=1.506$ , $P=0.266$ , NS
Group B	35.55±0.56	3.07±0.26	6.7±0.11	$F=2432.6$ , $P=6.55E-56$ , NS	$F=0.736$ , $P=0.661$ , NS	$F=1.705$ , $P=0.203$ , NS
Group C	34.55±0.85	5.65±0.45	7.03±0.14	$F=854.9$ , $P=3.15E-43$ , NS	$F=0.654$ , $P=0.742$ , NS	$F=0.807$ , $P=0.631$ , NS
ANOVA A*B* C*	-	-	-	$F=1.341$ , $P=0.318$ , NS	$F=13.01$ , $P=2.22E-05$ , S	$F=2.12$ , $P=0.13$ , NS

Confidence –95%;  $P<0.05$ ; S –Significant;  $P>0.05$ ; NS – Non significant; ANOVA – Analysis of variance; SD – Standard deviation

**Table 2: Comparison between the salivary flow rate (mL/min) for 10 min and salivary pH**

Groups	Mean salivary flow rate (mL/min)	Mean salivary pH	Wilcoxon signed rank test
Group A	4.34±0.35	6.8±0.11	$Z=-3.758$ , $P=0.000$ , S
Group B	3.07±0.26	6.7±0.11	$Z=-3.925$ , $P=0.000$ , S
Group C	5.65±0.45	7.03±0.14	$Z=-2.991$ , $P=0.003$ , S

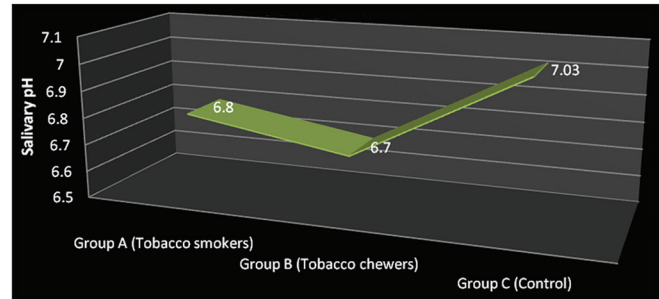
Confidence –95%;  $P<0.05$ =S (Significant);  $P>0.05$  – NS (Non significant)

## DISCUSSION

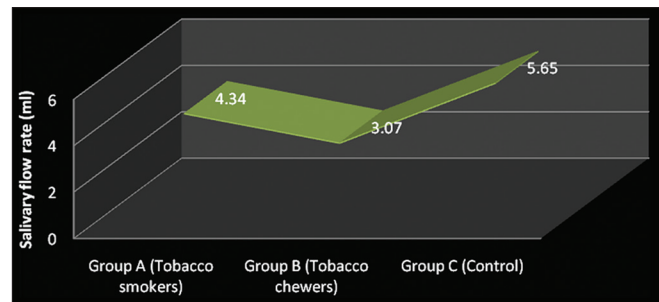
Saliva is a complex and important body fluid, which is very essential for oral health.<sup>[2]</sup> Its functions are protection of the oral mucosa, teeth remineralization, digestion, taste sensation, pH balance and phonation. It is composed of various electrolytes, peptides, glycoproteins, and lipids having antimicrobial, antioxidant, tissue repair, and buffering activities.<sup>[13]</sup> Therefore, altered whole-mouth SFR has an important role in the pathogenesis of oral and dental diseases.<sup>[4]</sup> There are many clinical and epidemiological evidences regarding the adverse effects of tobacco on oral health.<sup>[6,14]</sup> Saliva is the first biological fluid that is exposed tobacco (smoked/smokeless form), which contains numerous toxic compositions responsible for structural and functional changes in saliva.<sup>[11]</sup>

In the present study, the mean (±SD) SFR was found to be 4.34 (±0.35) in the group A, 3.07 (±0.26) in group B and 5.65 (±0.45) in group C, when compared a significant difference was noted ( $F=13.01$ ,  $P=2.22E-05$ ). This decrease in SFR in group A and group B subjects is probably due to the effect of nicotine on the taste nerve apparatus.

Rooban *et al.*,<sup>[2]</sup> observed that the raw form of areca nut (RAN) has a highest mean SFR (4.18 mL/10 min) as compared to the non-chewers (3.5 mL/min for 10 min) and other chewers. This finding is in contrast with the study where differences in mean SFR between smokers 3.12 (±1.56) and non-smokers 3.40 (±1.69) as well as between tobacco chewers and tobacco non-chewers were not significant.<sup>[2]</sup> Khan *et al.*, observed that some individuals develop tolerance to the salivary effects of smoking in the long-term use.<sup>[3]</sup> A number of studies



**Figure 3: Comparison of mean salivary pH between the groups**



**Figure 4: Comparison of mean salivary flow rate (mL/min) for 10 min between the groups**

have shown that cigarette smoking would typically cause a noticeable short term increase in SFRs, where as the long-term influence of tobacco use is still unclear.<sup>[9]</sup>

However, studies have shown that long-term consumption of tobacco in any form, especially smokeless form, is one of the risk factors for reducing saliva,<sup>[2,7]</sup> which was observed in the present study. These findings were also consistence with the finding of Rad *et al.*<sup>[7]</sup>

Moreover, in the present study it was also observed that the mean (±SD) salivary pH of whole saliva, was 6.8 (±0.11) in the group A, 6.7 (±0.11) in group B and 7.03 (±0.14) in group C. In the present study, salivary pH was found to be lower (acidic) in tobacco smokers and tobacco chewers than in controls, but the difference was statistically insignificant ( $F=2.12$ ,  $P=0.13$ ). Group B subjects has lowest salivary pH probably because of

use of lime in smokeless form, which can react with bicarbonate buffering system by the loss of bicarbonate, turning saliva more acidic. The alteration in electrolytes and ions alters the pH as they interact with the buffering systems of saliva.<sup>[1]</sup>

Khan *et al.*, also observed a lower salivary pH in smokers than in non-smokers.<sup>[12]</sup> which was consistent with the findings of the present study. Rooban *et al.*,<sup>[2]</sup> observed a mean pH of 6.77 in non-chewers and those who chew RAN, the mean pH turns acidic. In contrast Reddy *et al.*, observed no difference in salivary pH between the chewers and non-chewer.<sup>[15]</sup> This difference could be due to the amount of tobacco, lime, and other components. The role of lime in paan and beetal quid has been a source of concern. Lime (calcium oxide in aqueous forms calcium hydroxide) could cause a free radical injury or the high alkaline content probably reacts with the salivary buffering systems and alters the pH.<sup>[16]</sup>

SFR influences the pH of saliva.<sup>[17]</sup> It was observed in the present study when the SFR decreases the pH become acidic and vice versa. A statistical significant correlation exists between SFR and pH when compared, group A ( $Z=-3.758$ ,  $P=0.000$ ), group B ( $Z=-3.925$ ,  $P=0.000$  and group C ( $Z=-2.991$ ,  $P=.003$ ). An increase in SFR alters salivary pH by increasing bicarbonate secretion.<sup>[18]</sup> An increase in saliva bicarbonate increases the salivary pH.<sup>[19]</sup>

## CONCLUSION

Our observations are based on this preliminary study, in which the sample size was small with multiplicity of factors. SFR and pH can vary and are the limitations of the present study. From the present study, we can conclude that the long-term use of tobacco especially the smokeless form can cause significant alteration in SFR (decreases) and pH (acidic) These alteration in long-term tobacco users can render oral mucosa vulnerable to various oral and dental diseases.

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