

Impact of Smoking and Smokeless Tobacco on Salivary Flow Rate, pH Levels and Salivary Electrolytes

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ABSTRACT

Background: Tobacco use, both in smoked and smokeless forms, poses a significant threat to public health. The specific impact of tobacco use on the parameters of saliva still needs to be explored. Hence, the current study aimed to identify the impact of smoking and smokeless tobacco on salivary flow rate, pH levels, and salivary electrolytes.

Methods: This cross-sectional study was conducted at community health centers, dental clinics, and public spaces in Hyderabad from January 2024 to July 2024. A total of 150 smokers and smokeless tobacco users who were >18 years old were recruited via convenience sampling technique. MedCalc software was used, and a one-way analysis of variances was run to identify the differences in the values of groups. The level of significance was kept at 95% CI.

Results: Salivary flow rate, pH levels, and electrolyte measurements were analyzed, and the findings revealed that levels of the unstimulated flow rate of saliva were noticeably less ($p < 0.001$) in both smoking and smokeless tobacco user groups compared to the control group.

Conclusion: Overall, the results underscore the dangerous impact of tobacco on oral health, highlighting the need for targeted interventions to reduce tobacco use and promote oral health awareness.

Keywords: Tobacco, Chewing Tobacco, Mouth Diseases, Salivary Gland.

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INTRODUCTION

Saliva is a complex yet essential bodily fluid critical for maintaining oral health. It supports the integrity of the oral mucosa, aids in mineralization and digestion, enhances taste sensation, and contributes to phonation. Saliva also held diagnostic value for various infections, making it vital for homeostasis^{1,2,3,4}. Salivary flow, whether unstimulated or stimulated, functioned as a reflex dependent on afferent stimulation and coordination from higher brain centers^{5,6}. On average, daily salivary secretion ranged from 0.75–1.5 liters, with an unstimulated salivary flow rate (SFR) of approximately 0.3–0.5 mL/min, increasing up to 10 mL/min when stimulated^{7,8}.

Smoking was a prevalent habit worldwide, with one-third of the adult population identified as smokers⁹. Although the number of smokers had been declining, those who continued to smoke tended to do so more frequently. Smoking was a well-established risk factor for oral health issues, including xerostomia, halitosis, enhanced calculus formation, periodontal diseases, and severe conditions such as oropharyngeal and respiratory cancers¹⁰. Saliva, as the first fluid to encounter smoking-related toxins, underwent structural and functional changes^{11,12}.

The relationship between smoking and salivary flow rate remained complex, with conflicting evidence regarding its effects¹³. Smokeless tobacco products, including chewing tobacco, snuff, betel quid, gutkha, and paan, were also widely used and linked to oral health problems such as periodontal diseases, lesions, and cancers^{14,15}. Both smoking and smokeless tobacco altered salivary flow rate, pH levels, and electrolyte composition, significantly affecting the oral environment.

Tobacco use, in any form, posed a significant public health threat, particularly in Southeast Asia, where prevalence rates remained high^{16,17,18}. This study addressed gaps in understanding by investigating the effects of smoking and smokeless tobacco on salivary flow rate, pH levels, and electrolytes among individuals in Hyderabad, Pakistan.

METHODS

The Ethical Review Committee of Isra University approved the study. Informed consent was obtained from all participants prior to data collection. Each participant was assigned a unique code, with all data securely stored and accessible only to the research team to maintain confidentiality. The study sticks to the ethical

principles outlined in the Declaration of Helsinki, ensuring the protection of participants' rights and well-being. It was a cross-sectional study conducted at different community health centers, dental clinics, and public spaces in Hyderabad, Sindh. A cross-sectional study was conducted from January 2024 to July 2024 at different community health centers, dental clinics, and public spaces in Hyderabad, Sindh. A sample size of N=150 was taken through convenient sampling by considering the prevalence of tobacco consumption in Pakistan equal to 13.4%, as reported in the study "Prevalence of tobacco use in Urban and rural areas of Pakistan; a sub-study from National Diabetes Survey of Pakistan"¹⁹. Using the following formula:

$$n = \frac{[DEFF * Np(1-p)]}{[(d2/Z21-a/2*(N-1)+p*(1-p)]}$$

The selection criteria involve participants aged >18 years, current smokers (using cigarettes or other smoked forms of tobacco) for at least one-year, current users of smokeless tobacco (such as chewing tobacco, snuff, betel quid, gutkha, or paan) for at least one year, and non-tobacco users (control group) who have never used any form of tobacco. Participants were excluded if they used both smoked and smokeless tobacco regularly; however, if a participant used both forms but reported a significantly higher frequency of one type over the other, they were assigned to the group corresponding to the predominantly kind of use. Other exclusion criteria having systemic diseases affecting salivary function (e.g., diabetes, Sjögren's syndrome), being on medications known to influence salivary flow (e.g., anticholinergic, antidepressants), being pregnant or lactating and having a history of head and neck radiation therapy.

Participants were divided into three groups: smokers, smokeless tobacco users, and non-tobacco users. After obtaining informed consent, participants underwent the following procedures:

Participants were asked not to eat or drink anything and not to use the tobacco for at least 1 hour before the collection. Unstimulated saliva was taken from participants, and they were asked to spit into a pre-weighed container for 5 minutes. The amount of saliva produced was measured to calculate the flow rate (mL/min)²⁰. The pH of unstimulated and stimulated saliva samples was measured immediately using a calibrated pH meter²¹. Collected saliva samples were analyzed for sodium (Na⁺), potassium (K⁺), and calcium (Ca²⁺) levels using ion-selective electrode analysis in a laboratory setting (Fig.1)²².

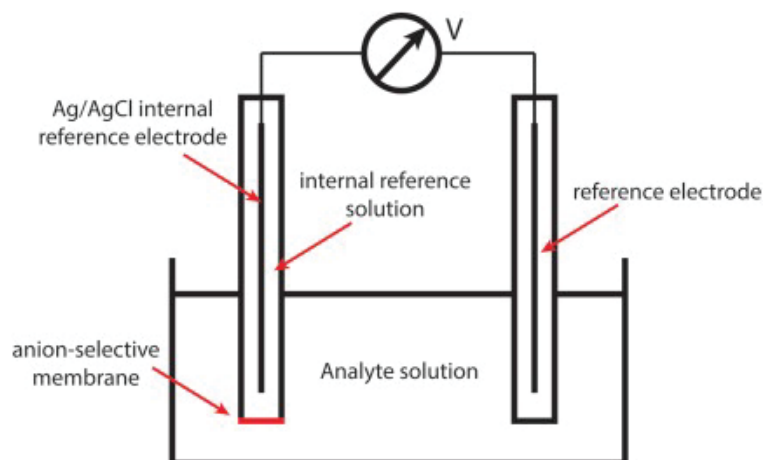


Figure-1: Diagram Shows Ion-Selective Electrode Analysis, Adapted from The Chapter of “Comprehensive Supramolecular Chemistry II”.

All salivary samples were labeled using unique participant codes to ensure anonymity and confidentiality. These codes were used throughout the analysis process to maintain privacy. Samples were securely stored in a temperature-controlled environment and were accessible only to authorized members of the research team. Participants were not provided with individual test results. However, those identified as belonging to the test groups (smokers and smokeless tobacco users) were informed about the study's general findings, including the observation of low salivary calcium levels. Participants were educated on the potential health risks associated with tobacco use and were provided with resources for cessation support.

A MedCalc statistical software was used to perform the data analysis for demographic description analysis frequencies were identified whereas inferential statistics were performed by applying a one-way analysis of variance at 95% of CI.

RESULTS

Descriptive analysis has revealed that the mean age of the participants in the control group was 46.18 ± 15.21 years, in the smoking group 45.10 ± 13.31 years and in the smokeless tobacco consumption was 46.80 ± 14.22 years ($p=0.83$) Table-1.

Table 1: Demographic Information of The Participants Included in The Study

Group	Male n (%)	Female n (%)	Age (Years) Mean±SD	F-Ratio	Level of Significance
Control (n=5)	28 (56%)	22 (44%)	46.18±15.21	0.182	0.83
Smokers (n=50)	40 (80%)	10 (20%)	45.10±13.31		
Smokeless Tobacco (n=50)	35 (70%)	15 (30%)	46.80±14.22		

One-way ANOVA was applied to determine differences in mean age among groups $p>0.05$

Further salivary flow rate measurements, pH levels, and electrolyte measurements were performed and the analysis of the findings revealed that the levels of unstimulated flow rate were less $p \leq 0.001$ in both smoking and smokeless tobacco user groups in comparison to the control group. Moreover, saliva pH levels were also compromised in both smokers and smokeless tobacco users in comparison to the control group $p \leq 0.001$ Table-2.

Table 2: Determining Difference in Salivary Flow Rate and pH Levels Among Group

Parameter	Groups	Average SFR (mL/min)	F-Ratio	Level of Significance	Difference among factors	p-value		
Salivary Flow Rate (mL/min)	Control (1)	2.65±0.69	322.56	p<0.001	1 > 2	*0.001		
					1 > 3	*0.001		
					2 < 1	*0.001		
	Smokers (2)	0.43±0.22			2 < 3	0.350		
					Smokeless Tobacco (3)	0.63±0.40	3 < 1	*0.001
							3 > 2	0.600
Saliva pH	Control (1)	6.02±1.28	9.24	p<0.001	1 > 2	0.220		
					1 > 3	*0.001		
					2 < 1	0.180		
	Smokers (2)	5.70±0.42			2 > 3	0.750		
					Smokeless Tobacco (3)	5.34±0.42	3 < 1	*0.001
							3 < 2	0.450

Post-hoc pairwise comparison

*Significance $p < 0.05$, $p \leq 0.001$

Levels of electrolytes were also analyzed and the findings provided evidence that the levels of sodium and potassium were changed among smokers and smokeless tobacco users but differences were non-significant $p > 0.05$ whereas for calcium; the levels were significantly different among the three-group $p \leq 0.001$ **Table-3, Fig-2.**

Table 3: Determining Difference in The Levels of Electrolytes Among Groups

Parameter	Groups	Mean±SD	F-Ratio	Level of Significance	Difference Among Factors	p-value		
Sodium (mEq/L)	Control (1)	20.96±4.90	1.301	0.275	1 < 2	0.350		
					1 < 3	0.220		
					2 > 1	*0.001		
	Smokers (2)	22.16±2.37			2 > 3	0.600		
					Smokeless Tobacco (3)	21.44±3.45	3 < 1	*0.020
							3 < 2	0.450
Potassium (mEq/L)	Control (1)	25.18±5.80	0.188	0.828	1 < 2	0.400		
					1 < 3	0.600		
					2 > 1	0.050		
	Smokers (2)	25.66±2.39			2 > 3	0.800		
					Smokeless Tobacco (3)	25.48±2.71	3 < 1	0.150
							3 < 2	0.250
Calcium (mg/dL)	Control (1)	1.28±0.177	111.577	< 0.001	1 > 2	*0.001		
					1 > 3	*0.001		
					2 < 1	*0.001		
	Smokers (2)	0.85±0.14			2 < 3	0.750		
					Smokeless Tobacco (3)	0.90±0.14	3 < 1	*0.001
							3 > 2	0.600

Post-hoc pairwise comparison

*Significance $p < 0.05$, $p \leq 0.001$

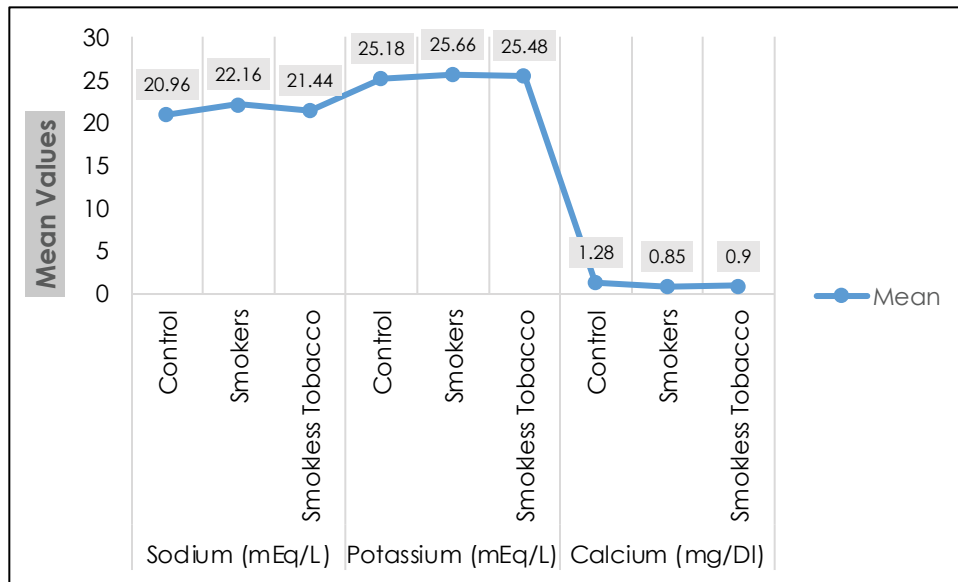


Figure - 2: Shows the Mean Difference Between Three Electrolytes Between the Groups

DISCUSSION

The reporting of this study reveals significant differences in salivary parameters and electrolyte levels among participants who smoke or use smokeless tobacco and those in the control group. Although the mean age of participants did not significantly differ across the three groups ($p=0.83$), there were notable changes in salivary flow rates and pH levels. Both smokers and smokeless tobacco users exhibited significantly reduced unstimulated salivary flow rates compared to the control group ($p<0.001$). Additionally, salivary pH levels were significantly lower in both smokers and smokeless tobacco users compared to the control group, indicating compromised oral health in tobacco users ($p<0.001$). Furthermore, while sodium and potassium levels showed no significant difference among the groups ($p>0.05$), calcium levels were significantly lower in both smokers and smokeless tobacco users compared to the control group ($p<0.001$).

A study conducted to assess the impact of tobacco use on salivary pH levels found that both tobacco chewers and smokers exhibited significantly lower salivary pH compared to non-users, highlighting the detrimental effects of tobacco on oral health. The results showed that tobacco chewers had a mean salivary pH of 6.59 ± 0.4399 , smokers had a mean pH of 6.87 ± 0.4835 , and non-users had a mean pH of 7.12 ± 0.144623 . These findings align with our results, which also demonstrate significantly lower salivary pH levels in both smokers and smokeless tobacco users compared to the control group ($p<0.001$).

In another study, it was found that smokers exhibit a significantly reduced salivary flow rate (SFR) compared to non-smokers, reinforcing the negative

impact of smoking on oral health. The study revealed a statistically significant decrease in SFR among smokers ($p<0.0001$), with further reductions observed as the duration ($p<0.001$) and frequency ($p=0.012$) of smoking increased²³. These findings are consistent with our results, where both smokers and smokeless tobacco users demonstrated significantly lower unstimulated salivary flow rates compared to the control group ($p<0.001$). The reduction in SFR associated with smoking is particularly concerning, as it can lead to dry mouth (xerostomia), increasing the risk of oral complications such as tooth decay, gum disease, and oral infections.

In a related study investigating the effects of Smokeless Tobacco (SLT) on periodontal health, it was found that SLT use significantly disrupts the balance between reactive oxygen species (ROS) and antioxidants (AO) such as glutathione in saliva²⁴. The study demonstrated that periodontitis patients using SLT had lower glutathione levels in their saliva at baseline and one month after non-surgical periodontal therapy compared to non-SLT users ($p<0.001$)^{25,26}.

In another study, it was found that while the mean bleeding on probing (BOP) was much higher in smokers, the mean calcium level of female smokers was significantly lower than that of non-smokers despite there being no statistically significant difference in the total daily calcium consumption, indicating that calcium levels may be influenced by factors other than intake. The study also found that salivary calcium levels and BOP decreased with increased smoking times²⁷. Compared to the control group, our results showed a substantial decrease in calcium levels among smokers and users of smokeless tobacco ($p<0.001$). The steady decline in calci-

um levels highlights the need for more research on the impact of smoking on calcium metabolism²⁸.

The strength of this study lies in its comprehensive approach, assessing multiple salivary parameters, including flow rate, pH levels, and electrolytes, to understand the impact of smoking and smokeless tobacco on oral health. However, one of the areas for improvement is our study design, which is a cross-sectional design that restricts various variables to be studied and limits the causal relationship between tobacco use and changes in the parameters of saliva.

CONCLUSION

The study concludes that both smoking and smokeless tobacco can cause a reduction in salivary flow rate and altered pH levels, along with a significant reduction in calcium levels. While sodium and potassium levels showed changes, these were not statistically significant. The results overall mention the harmful effects of tobacco on oral health.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

ETHICAL APPROVAL

The ethical approval for this study was granted by the Faculty of Dentistry & Allied Sciences, ISRa University, Hyderabad, under reference number: IU/DN(F-D)/IDC/RD/2024/02.

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AUTHOR'S CONTRIBUTIONS

SSH and **SHA** conceptualized the manuscript, **SMAB** and **AAS**, and **SAAZ** performed data collection. **AA** and **SSH** performed an analysis of data. **HS**, **SSH**, and **SHA** wrote the original draft **FR** performed final revisions and proofreading.

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