Review Article

Cytotoxic and genotoxic effects of cigarette and waterpipe tobacco smoking on buccal mucosa: A systematic review and meta-analysis

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Abstract

Background: Waterpipe tobacco smoking (WTS) is an issue all over the world, although it is particularly prevalent in the Middle East, and Southeast Asia. The genotoxic effects of smoking were reported to be associated with nucleus abnormalities such as micronuclei (MN), karyorrhexis (KR), karyolysis, pyknosis, binucleates, broken eggs, condensed chromatin in exfoliated buccal mucosal cells, and was believed to be associated with apoptosis of cells and was not correlated to the exposure time.

Aim: The aim of this study was to evaluate and compare the cytotoxic and genotoxic effects of cigarette and WTS on buccal mucosa.

Materials and Methods: The pertinent search was done through the computerized literature on MEDLINE, EMBASE, and PUBMED databases, which included case-control, clinical and observational studies regarding the mutagenic effects of cigarettes and WTS in oral tissues. The retraction of data in this study was undertaken from May 2010 to May 2022. A total of 60 articles from the search data were retrieved. This investigation was registered with the research center of Riyadh Elm University for institution review board approval (IRB) and obtained the IRB number "FRP/2021/448/733/707 and the systematic review registration number with respect to PROSPERO is 345417.

Results: After the removal of duplicates, 32 were evaluated for the inclusion and exclusion criteria. Out of 32 articles, twenty studies were evaluated for cytogenetic abnormalities in buccal mucosal cells of waterpipe tobacco smokers (WTS) and cigarette smokers, and 12 were excluded. The mean MN levels in the oral tissues of WTS were more (1.94 ± 0.39) than in non-smokers (1.68 ± 0.35).

Conclusion: Therefore, we conclude that the MN count can be employed as a biomarker and preliminary signal for the identification of changes in oral mucosa among smokers, which develop towards cancer formation.

Keywords: Buccal mucosa, cigarette, cytotoxic, genotoxic, micronuclei, water-pipe

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INTRODUCTION

Micronuclei (MN) frequencies corroborate the commonly recognized idea that the MN are products of early events in human oncogenic pathway development, especially in the oral cavity, which is immediately exposed to cigarettes and WTS.[1-6] The typical documented stable population with respect to MN frequency is 1–3 per 1000 cells.[7-9] Brown, J.E *et al*.[8] recognized that the evidence of genotoxic and cytotoxic effects of smoking on buccal mucosa was very limited and controversial. According to Jensen R.P *et al*.[10] mutagenic symptoms have been reported to be associated with nuclear dyscrasias. Few studies reported that the mutagenic abnormalities of oral tissues in cigarettes and WTS are > in nonsmokers.[11,12] Pop A.M *et al*.[13] reported that diagnostically healthy young tobacco users displayed an increased number of MN in the buccal epithelia, compared to non-smokers, suggesting the existence of histological alterations.

Early detection of cytological and genotoxic damages to the epithelia of cigarette and WTS smokers may assist in increasing longevity. Therefore, the purpose of the present review is to evaluate and compare the mutagenic aspects of cigarettes and WTS on dental epithelia. This investigation will help to detect the early alterations in the buccal mucosa and assess the risk for carcinomas formation in such individuals.[14]

MATERIALS AND METHODS

Study design

This present systematic review was carried out with a search in the literature that included original full-text articles, cross-sectional, observational, descriptive studies, published from May 2010- May 2022, which evaluated the cytotoxic and genotoxic effects of cigarette and WTS on buccal mucosa."

Protocol and registration

This investigation was registered with the research center of Riyadh Elm University for institutional review board approval (IRB) and obtained the IRB number "FRP/2021/448/733/707 and the systematic review registration number with respect to PROSPERO is 345417.

Search strategy

The present systematic review of the literature was carried out both electronically and manually. The relevant literature search was carried out through searches of the digitized literature on MEDLINE, EMBASE, and PubMed databases, and manual search irrespective of the date of publication using Medical Subject Headings (MeSH) terms. A total of 32 papers were identified with this method. Various keywords utilized in the search strategy included such as cigarette smoking, waterpipe smoking, cytotoxic, genotoxic, buccal mucosa, exfoliated buccal cells, and periodontal health.

Selection criteria

Initially, titles and abstracts of the records retrieved by the search were assessed in order to exclude those studies that were inappropriate. Retrospective studies were not included. For the remaining studies, full-text articles were recovered that met the inclusion criteria. Selected studies were screened using the STROBE checklist for observational studies.[15]

Inclusion criteria

Study selection was based on the following: (1) Studies published until May 2022 (2) full-text articles published in the English language (3) studies evaluating the cytotoxic effects of cigarette smoking on the buccal mucosa (4) studies evaluating the genotoxic effects of cigarette smoking on buccal mucosa. (5) Studies evaluating the cytotoxic effects of WTS on exfoliated buccal mucosa cells and buccal mucosa (6) studies evaluating the genotoxic effects of WTS on exfoliated buccal mucosa cells and buccal mucosa (7) cross-sectional, observational studies clinical, case-control studies and review articles were included.

Exclusion criteria

The studies that were excluded from the present review were: Studies published before May 2010, studies published in other than the English language, articles having only titles, conference abstracts, editorial letters and retrospective data, cytotoxic and genotoxic studies done in the oral cavity other than in buccal mucosa, animal and plant studies, studies on Nargile and Marijuana were excluded.

Control of bias assessment

The following issues were included in the risk of bias or quality assessment in the present systematic review: (1) Completeness of article information on cytotoxic and genotoxic effects of smoking on the buccal mucosa (2) selective outcome reporting (3) outcome measures (cytotoxic and genotoxic effects of smoking on buccal mucosa) (4) study design and (5) conflict of interest in the conduct of the study.

Collection and data extraction

The search retrieved 60 articles. After the removal of duplicates, 32 articles were identified. After title and abstract screening, 20 studies remained and 12 studies

were excluded. Out of 32 studies, 20 studies remained for qualitative analysis and five for the meta-analysis of the primary and secondary outcomes. All authors analyzed the selected studies and critically reviewed the main findings. This review was done according to the guidelines set forth by Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA).[16]

RESULTS

Study selection and characteristics

The search retrieved 60 articles from the search data. After the removal of duplicates, 32 were evaluated for the inclusion and exclusion criteria. After careful analysis, 20 articles were included[4,5,13,17-33] and after comprehensive evaluation of the titles, abstracts, resulted in the exclusion of 12 articles[1,3,9,12,34-41] The reason for exclusion was due to incomplete articles, conference abstracts, articles other than the English language, retrospective studies, animal studies, editorial letters, studies on Marijuana and Nargile users and studies on blood and saliva, gingiva and those not following inclusion criteria were excluded, as shown in Figure 1, according to PICO framework.

From 20 articles that were evaluated and included in this study, eight studies[5,22,23,24,26,29,31,33] compared the cytotoxic and genotoxic effect of WTS and conventional and electronic cigarettes on buccal mucosa/exfoliated buccal mucosal cells. Whereas, seven studies were among smokers and non-smokers[4,17,19-21,25,28] which compared the cytogenetic abnormalities in desquamated cells of the oral mucosa. The other five studies included those with a smokeless form of tobacco users and smokers of different tobacco products.[4,18,27,13,30] These studies have mentioned that smoking WTS and cigarettes cause cytotoxic damage to cells by causing an increase in MN compared to non-smokers/controls.

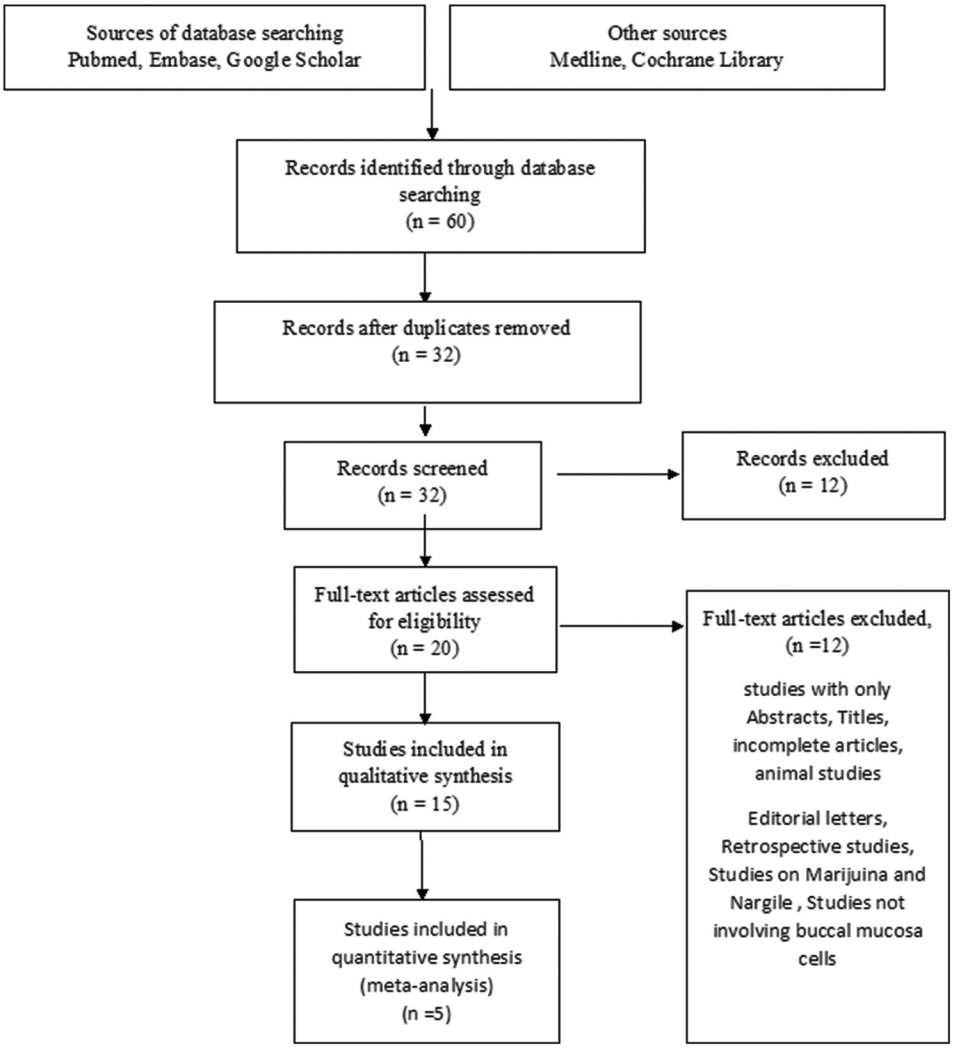

Figure 1: PRISMA Flow Diagram for data search strategy

This present review includes studies that were done in different countries such as USA, UK, Brazil, Bosnia, Romania, Iraq, Iran, UAE, Egypt, Jordan, India, and Pakistan as shown in Table 1. Meta-analysis of the five articles included as shown in Figure 2 and the Funnel plot of the odds ratio for publication bias of included articles was shown in Figure 3. The meta-analysis results included five articles (Bibars AR *et al*. 2015[5]; Jackson M, *et al*. 2020[22]; Javed H *et al*. 2017[25]; Jalayer Naderi N *et al*. 2017[23]; Prasad P, *et al*. 2018[31]) which fulfilled the criteria of inclusion of the comparison of the groups of cigarette smoking and water pipe smoking. The odds ratio favors the vales of water pipe smoking to greatly affect the buccal mucosa cells in these individuals.

DISCUSSION

Different terms are used to describe WTS depending on different regions and cultures. It is known as Shisha, Narghile, and Hookah in different countries. WTS is very common nowadays among young teenagers, especially among college students.[3] It is a frequent practice in Arabic countries and in several Asian ones. It commonly occurs among friends in social situations such as private residences or events that offer primed outlets to consumers for smoking purposes.[2] Most waterpipe smokers believed, that it is less harmful and less addictive than cigarette smoking. The available literature demonstrates that both types of smoking are dangerous, with many similar health impacts. Compared to cigarette users, waterpipe smokers have been discovered to have higher amounts of toxins and teratogenic chemicals that cause malignancies. *In vitro* investigations indicated that WTS exposure caused mutations in WBCs and oral tissues.[42]

A total of 20 studies were evaluated for cytogenetic abnormalities in the oral tissues of WTS and cigarette smokers in the present review. The MN test is a better indicator of genotoxicity damage and can be used as biomarker for the assessment of DNA damage.[27] Bansal *et al*.[4] have reported MN is higher in smokeless users than in smokers and non-smokers. Therefore, smokeless tobacco users have an increased risk for cancer due to an increase MN count. This study is similar to the study conducted by Devadoss S *et al*.[18] & Motgi AA,[27] which reported nuclear abnormalities such as prominent nucleoli and condensed chromatid among smokeless users than in smokers. A study by Da Siva VHP *et al*.[17] reported that MN incidence was high in the exfoliated cells of buccal mucosa of cigarette smokers than in non-smokers. This is consistent with other studies done by Farhadi *et al*. S[19] & Shafi FAA.[32] MN originate from chromosome fragments that lag behind at anaphase during nuclear division. MN studies on peripheral blood and exfoliated cells of buccal mucosa were reported by Fenech M *et al*.[20] and Haveric A *et al*.[21] MN is higher in the buccal mucosa of smokers than in non-smokers and is associated with duration, age, and intensity of smoking, unlike the MN in lymphocytes, which are not correlated or associated with these factors. Bonassi *et al*.[7] have reported MN frequency increased in heavy smoking and decreased with the daily intake of fruits. Jalayer NN & Pasha P[23] reported MN count increased in cigarette smokers than in WTS and controls and the mutagenic effects of tobacco smoking were not interlinked to exposure period and duration of smoking, However, WTS was correlated to the exposure time. This statement contradicts El-Setouhy M *et al*.[6] The MN count findings though contradict Jalayer NN & Pasha P's[23] study and are in agreement with El-Setouhy M *et al*.[6] & Moghaddam MR *et al*.[26] MN count not only depends on smoking habits but also on the occupation of the individual Javed H & Ghani N[25] & Fenech M *et al*.[20] Studies on MN Assay of buccal cells in water pipe smokers (WTS)/Hookah smokers by Nezhad MD *et al*.[29] & Taghibakhsh M *et al*.[33] reported that the mean number of MN is higher in WTS. Increased MN count in the buccal cells of cigarette smokers than in betel quid users and e-cigarette smokers.[13,30,31]

Limitations

The main drawback of our study is that it is primarily a systematic review with meta-analysis, which causes

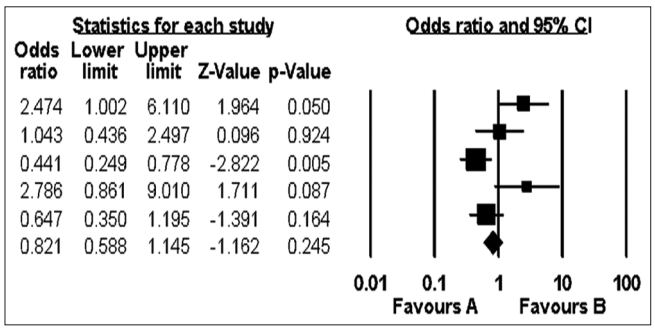

Figure 2: Meta analysis of the five articles included

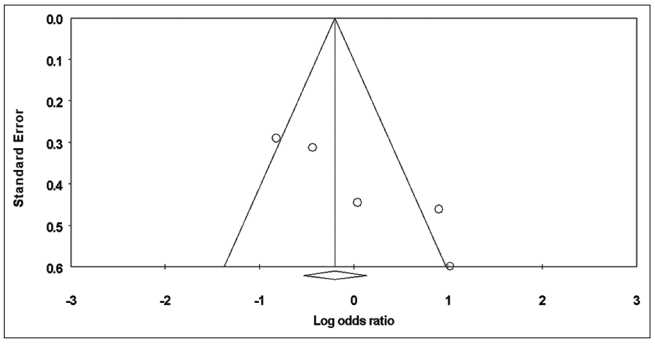

Figure 3: Funnel plot of odds ratio for publication bias

Table 1: Summary of studies under our investigation

Authors	Sample size	Study population	Study Setting	Study Region/ Location	Results
Bansal H, *et al*. (2012)[4]	75	Smokers, smokeless users and controls	Cross-sectional study	India	MN is higher in smokeless users than in smokers and non-smokers
Bibars *et al*. (2015)[5]	190	Cigarette smokers, waterpipe tobacco smokers (WTS), dual smokers and non-smokers	Comparison study	Jordan	OR=4.6 for cigarette smokers OR=4.3 WTS OR=4.9 dual smokers PPD (probing depth) >3 mm, CAL >1mm in smokers than in non-smokers. Me ta analysis of five types of forest plots are displayed for the mean of micronuclei
Da Silva VHP, Antonio RDL, Pompeias S, Ribeiro DA (2015)[17]	38	Cigarette smokers and non-smokers	Comparative study in exfoliated oral buccal mucosal cells	Brazil	Micronuclei incidence was 0.7±0.8 in smokers Whereas, in non-smokers it was 0.0++0.1
Devadoss S, *et al*. (2021)[18]	150	Smokers, smokeless tobacco and healthy controls	Case-control study	India	Smokeless tobacco users showed high rate of nuclear damages compared to smokers and health controls.
Farhadi S *et al*. (2016)[19]	26	Smokers and non –smokers related articles	Review study	Iran	Micronucleus assay plays significant role in assessing the genotoxic damages in oral buccal cells and can be used to assess risk for cancer in such individuals
Fenech M *et al*. (2011)[20]	29	Smokers and non-smokers	Review study	Australia	HUMN and HUMNxL studies on micronucleus assays in human buccal cells and lymphocytes
Haveric A, *et al*. (2010)[21]	87	Smokers and non-smokers	Case- control study	Bosnia	High frequencies of apoptotic cells were found in exfoliated buccal mucosal cells of smokers than in Non-smokers.
Jackson M *et al*. (2020)[22]	121	E-cigarettes, waterpipe tobacco smokers , (WTS) and dual smokers (WTS & cigarette smokers)	Pilot cross – sectional cohort study	USA	E-cigarette users showed increase in plasma IgE levels than non-tobacco users & WTS Dual users (WTS & smokers) showed increase in plasma IgG compared to other users.
Jalayer Naderi N, Pour Pasha P (2017)[23]	75	WTS, Cigarette smokers and healthy controls	Case control study	Iran	Cigarette smoking showed high cell death rates than in WTS.
Jalil S & Naderi NJ (2022)[24]	60	Water pipe smokers (WTS), Cigarette and non-smokers	Case- control study	Iran	Repair index is more in cigarette smokers than in WTS &Non-smokers. MN count is higher in WTS than in cigarette smokers
Javed H & Ghani N (2017)[25]	200	Smokers and controls (non-smokers)	Case-control study	Pakistan	DNA damage and cell death in workers of photocopy centers is directly associated with smoking and duration of exposure at work (occupation)
Moghaddam MR *et al*. (2020)[26]	90	Waterpipe tobacco smokers (WTS) and Cigarette smokers	Case-control study	Iran	Waterpipe smoking showed higher frequency of micronuclei (MN) than cigarette smoking
Motgi AA *et al*. (2014)[27]	100	Smokeless, smoked form of tobacco and control groups	Clinical case – control study	India	Smokeless users showed higher MN count in than in smokers and controls.
Naderi NJ *et al*. (2012)[28]	63	Smokers and controls	Cohort study	Iran	The mean number of cells with micronucleus was more in smokers with more than 10 years of smoking than in controls.
Nezhad MD *et al*. (2020)[29]	60	Waterpipe WTS (Hookah), smokers and non-smokers	Case –control study	Iran	The average amount of MN cells in WTS is greater than people who formerly smoked and was 1.94±0.39 & 1.68±0.35 . MN Count is dependent on duration and dose of WTS.
Pop AM *et al*. (2021)[13]	68	Conventional smokers, E- cigarette smokers and Non-smokers	Cross-sectional study	Romania	Conventional Smokers showed higher mean values of micronuclei than E-cigarette smokers and non-smokers
Pradeep MR *et al*. (2014)[30]	180	Cigarette Smokers. Betel quid smokers, smokeless tobacco users, control group	Comparative Case-control Study	India	Mean MN count in smokers is 3.11, which is higher than betel quid smokers (2.13) and smokeless tobacco users (1.67)
Prasad P *et al*. (2018)[31]	400	Hookah and tobacco smokers, dual smokers and non -smokers	Survey and observational study	UAE	WTS has the potential to cause pre-malignant lesions and oral cancer Dual smokers showed cardiovascular disorder (CVD) risk
Shafi FAA (2015)[32]	90	Smokers and non-smokers	Case-control study	Iraq	The average of MN cells in desquamated buccal tissues was (10.18±1.07) & (12.89±1.85) in nonsmoker and smokers.
Taghibakhsh M *et al*. (2019)[33]	72	WTS (Hookah) and control subjects	Cohort study	Iran	Hookah smokers showed increased MN cells in oral mucosa.

challenges with hypothesis testing because data on risk factors and outcomes are examined at the same time, although this does not appear to have an impact on our findings. Because the influence of smoking may fluctuate depending on sex, an imbalance in the proportion of males and females in our study sample could be a constraint. Future longitudinal studies are needed to better understand the long-term impact of WTS on the overall well-being of individuals with respect to a larger sample size; however, due to funding constraints and a lack of demographic records, these are challenging to conduct in poor countries.

CONCLUSION

The present investigation indicated a greater prevalence and count of MN cells in desquamated cells of buccal mucosa of WTS users compared to cigarette smokers and non-smokers among the included studies. Future studies are warranted to assess MN assays in the oral cavity of waterpipe tobacco and cigarette smokers and their correlation with the duration and dose of smoking.

Declaration of patient consent

The authors certify that they have obtained all appropriate patient consent forms. In the form, the patient(s) has/have given his/her/their consent for his/her/their images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity, but anonymity cannot be guaranteed.

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Nil.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Al-Halbosiy MM, Al-Jumaily RMK, Lafta FM. The exposure effect of water pipe smoke (WPS) on the total count leukocyte, mitotic index and micronucleus formation and chromosome aberration in albino male mice. J Biotechnol Res Center 2015;9:89-93.
2. Abdul NS, Alshehri NM, Bindawis HM, Alzahrani HK, Alanezi AF. Awareness of the effects of shisha and electronic smoking on oral health in Saudi population. Ann Dent Spec 2020;8:41-9.
3. Benvindo-Souza M, Assis RA, Oliveira EAS, Borges RE, Santos LRDS. The micronucleus test for the oral mucosa: Global trends and new questions. Environ Sci Pollut Res 2017;24:27724-30.
4. Bansal H, Sandhu VS, Bhandari R, Sharma D. Evaluation of micronuclei in tobacco users: A study in Punjabi population. Contemp Clin Dent 2012;3:184-7.
5. Bibars AR, Obeidat SR, Khader Y, Mahasneh AM, Khabour OF. The effect of waterpipe smoking on periodontal health. Oral Health Prev Dent 2015;13:253-9.
6. El-Setouhy M, Loffredo CA, RadwanG, RahmanRA, Mahfouz E, Israel E, *et al*. Genotoxic effects of waterpipe smoking on the buccal mucosa cells. Mutat Res 2008;655:36-40.
7. Bonassi S, Coskun E, Ceppi M, Lando C, Bolognesi C, Burgaz S. The human MicroNucleus project on eXfoLiated buccal cells (HUMN (XL)): The role of life-style, host factors, occupational exposures, health status, and assay protocol. Mutat Res 2011;728:88-97.
8. Brown JE, Luo W, Isabelle LM, Pankow JF. Candy flavorings in tobacco. N Engl J Med 2014;370:2250-2.
9. Celik A, Kanik A. Genotoxicity of occupational exposure to wood dust: Micronucleus frequency and nuclear changes in exfoliated buccal mucosa cells. Environ Mol Mutagen 2006;47:3-8.
10. Jensen RP, Luo W, Pankow JF, Strongin RM, Peyton DH. Hidden formaldehyde in e-cigarette aerosols. N Engl J Med 2015;372:392-4.
11. Peterson LA, Hecht SS. Tobacco, e-cigarettes, and child health. Curr Opin Pediatr 2017;29:225-30.
12. Tarbiah N, ToddI, Tighe PJ, Fairclough LC. Cigarette smoking differentially affects immunoglobulin class levels in serum and saliva: An investigation and review. Basic Clin Pharmacol Toxicol 2019;125:474-83.
13. Pop AM, CorosR, Stoica AM, Monea M. Early diagnosis of oral mucosal alterations in smokers and e-cigarette users based on micronuclei count: A cross-sectional study among dental students. Int J Environ Res Public Health 2021;18:13246.
14. Nersesyan AK, Vardazaryan NS, Gevorgyan AL, Arutyunyan RM. Micronucleus level in exfoliated buccal mucosa cells of cancer patients. Arch Oncol 2002;10:35-6.
15. von Elm E, Altman DG, Egger M, Pocock SJ, Gøtzsche PC, Vandenbroucke JP; STROBE Initiative. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. J Clin Epidemiol 2008;61:344-9.
16. Liberati A, Altman DG, Tetzlaff J, Mulrow C, Getzsche PC, Ioannidis JPA, *et al*. The PRISMA statement for reporting systematic review and meta- analysis of studies that evaluate health care interventions, explanation and elaboration. J Clin Epidemiol 2009;62:e1-34.
17. Da Silva VHP, Antonio RDL, Pompeia S, Ribeiro DA. Cytogenetic biomonitoring in buccal mucosa cells from young smokers. Acto Cyto 2015;59:474-8.
18. Devadoss S, Raveendranath MC, Kathiresan TS, Ganesan K. Genotoxic effect of various forms of tobacco on oral buccal mucosa and nuclear changes as a biomarker. J Pharm Bioallied Sci 2021;13:1141-8.
19. Farhadi S, Sadri D, Sarshar S. Micronucleus assay of Buccal mucosa: A useful noninvasive approach in screening of genotoxic nuclear damage. Adv Biores 2016;7:20-9.
20. Fenech M, Holland N, Zeiger E, Chang WP, Brurgaz S, Thomas P, *et al*. The HUMN and HUMNxL international collaboration projects on human micronucleus assays in lymphocytes and buccal cells–past, present and future. Mutagenesis 2011;26:239-45.
21. Haveric A, Haveric S, Ibrulj S. Micronuclei frequencies in peripheral blood and buccal exfoliated cells of young smokers and non-smokers. Toxicol Mechanism Methods 2010;20:260-6.
22. Jackson M, Singh KP, Lamb T, McIntosh S, Rahman I. Flavor preference and systemic immunoglobulin responses in e-cigarette users and waterpipe and tobacco smokers: A pilot study. Int J Environ Res Public Health 17:640.
23. Jalayer Naderi N, Pour Pasha M. Comparison of cytotoxic effect of cigarette and waterpipe smoking on human buccal mucosa. Int J Prev Med 2017;8:98.
24. Jalil S, Naderi NJ. Comparison of repair index in cigarette and water pipe smokers: A bio monitoring assessment using human exfoliated buccal mucosa cells. Int J Prev Med 2022;13:27.
25. Javed H, Ghani N. Cytogenetic damage in the buccal cells of photocopying workers in Lahore, Pakistan. J Pak Med Assoc 2019;67:275-9.
26. Moghaddam MR, Mirzamohammad MH, Yahyazadeh E, Gholinia H, Abbaszadeh H. Comparison of genotoxic effect in buccal exfoliated cells between cigarette and waterpipe smokers. Acta Cytologica 2020;64:1-6.

27. Motgi AA, Chavan MS, Diwan NN, Chowdhery A, Channe PP, Shete MV. Assessment of cytogenic damage in the form of micronuclei in oral epithelial cells in pateints using smokeless and smoked form of tobacco and non-tobacco users and its relevance for oral cancer. J Cancer Res Ther 2014;10:165-70.
28. Naderi NJ, Farhadi S, Sarshar S. Micronucleus assay of buccal cells in smokers with the history of smoking less and more than 10 years. Indian J Pathol Microbiol 2012;55:433-8.
29. Nezhad MD, Naderi JN, Semyari H. Micronucleus assay of buccal mucosa cells in waterpipe (Hookah) smokers: A cytologic study. Iran J Pathol 2020;15:75-80.
30. Pradeep MR, Guruprasad Y, Jose M, Saxena K, Deepa K, PrabhuV. Comparative study of genotoxicity in different of tobacco related habits using micronucleus assay in exfoliated buccal epithelial cells. J Clin Diagn Res 2014;8:21-4.
31. Prasad P, Hamed MS, Nahar P. Oral health effects in shisha smokers - A study among Arabs and Indians in UAE. Saudi J Oral Dent Res 2018;3:207-12.
32. Shafi F. The effects of smoking on micronucleus frequencies in buccal cells of health Iraqi individuals. World J Pharm Res 2015;9:406-15.
33. Taghibakhsh M, Farhadi S, Babaee A, Sheikhi M. The effect of Hookah use on buccal mucosa: Evaluation of repair index. Asian Pac J Cancer Prev 2019;20:1109-12.
34. Dalia E. Cellular and molecular biomarkers detected in the oral mucosa and saliva in water-pipe tobacco smoking compared to cigarette smoking: A systematic review. Available from: http://hdl.handle.net/11394/8754.
35. Fabian-Morales E, Fernández-Cáceres C, Gudiño Gómez A, Andonegui Elguera MA, Torres Arciga K, Escobar Arrazola MA, *et al*. Genotoxicity of Marijuana in mono-users. Front Psychiatry 2021;12:2183.
36. Jalayer Naderi N. Response to letter to the editor:" Is micronucleus assay suitable for cytogenetic biomonitoring the different ways to smoke?". Iran J Pathol 2020;15:352-4.
37. Jamil A, Rashid A, Naveed AK. Assessment of smoke induced genotoxicity in sprague dawley rats. PAFMJ 2019;69:577-81.
38. Khemiss M, Rouatbi S, Berrezouga L, Saad HB. Oral health effects associated with narghile use. La Tunisie Medicale 2016;94:401-2.
39. Onwukwe OS, Achukwu PU, Azubuike NC, Udeani TK, Okpukpara OB, Sofoluke OD. Frequency of buccal cell micronuclei in abattoir workers exposed to smoke from singeing animal hide in Enugu, South East Nigeria. J Environ Toxicol Public Health 2019;4:1-5.
40. Ramenzoni LL, SchneiderA, Fox SC, Meyer M, Meboldt M, Attin T, *et al*. Cytotoxic and inflammatory effects of electronic and traditional cigarettes on oral gingiva cells using a novel automated smoking instrument: An *in vitro* study. Toxics 2022;10:179.
41. Welz C, Canis M, Schwenk ZS, Becker S, stuckeV, Ihler F, *et al*. Cytotoxic and genotoxic effects of electronic cigarette liquids on human mucosal tissue cultures of the oropharynx. J Environ Pathol Toxicol Oncol 2016;35:343-54.
42. Ramôa CP, Eissenberg T, Sahingur SE. Increasing popularity of waterpipe tobacco smoking and electronic cigarette use: Implications for oral health care. J Periodontal Res 2017;52813-23.