Original Article

Genotoxicity in Analog and Smart Mobile Phone Users by Assessing Micronuclei in Oral Exfoliated Cells – A Comparative Study

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Abstract

Aim: The number of micronuclei in oral exfoliated buccal mucosal cells of analog (basic) and smart mobile phone users was evaluated and compared. **Materials and Methods:** The study population constituted 30 individuals using basic and smartphones within the age group of 45–55 years. They were divided into two groups: Group 1–15 participants using basic mobile phones and Group 2–15 participants using smart mobile phones. Exfoliated buccal mucosal cells were collected from both right and left buccal mucosa, stained with Papanicolaou stain, and evaluated microscopically for the estimation of micronuclei count. Mean micronuclei count was compared statistically between the study groups and also between the sides of frequent usage and opposite sides within the study groups. **Results:** There was a significant increase in the mean micronuclei count in Group 2 compared to Group 1 and the comparison of mean micronuclei count between the side of frequent phone usage (right side) and opposite side (left side) also showed significant difference in both Groups 1 and 2. **Conclusion:** Despite technological advance and high-end features, the use of smartphones causes more genotoxicity compared to basic model or analog phones. Although this technology-dominated era mandates use of such phones as a part of routine lifestyle, it is imperative to adopt safety precautions such as use of headphones while talking, carrying a separate pouch for mobile phones to minimize the genotoxic damage.

Keywords: Genotoxicity, micronuclei, smartphone

Introduction

In modern days, mobile phones have become an indispensable part of life worldwide.[1] Mobile phones are continuously gaining recognition, especially within the younger generation. India has become world's second-largest population of mobile phone users with teledensity of 80%. Mobile phones use microwave radiation in the carrier frequency range of 900–1800 MHz.[2] Apart from basic services such as calls and messages, use of mobile phones for data transfer, music, games, and other applications are becoming increasingly popular in recent times.[3]

This technology is based on microwave-frequency electromagnetic radiation (both microwaves and radiofrequency waves). Depending on the area of the world, the radiation frequency and modulation standards range from 300 to 2100 MHZ.[3]

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The International Commission on Nonionizing Radiation Protection (ICNIRP) has established guidelines for exposure limitations for all of these frequencies (up to 300 GHz) (ICNIRP, 2009), which have been incorporated into national regulations in a numerous nation.[3] With respect to biological systems, the electromagnetic energy absorbed by a unit of tissue is known as the specific absorption rate (SAR, w/kg). The identified

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whole-body threshold level of exposure in terms of SAR is 4 w/kg.[1]

In 2012, the WHO has classified mobile phone radiation on the International Agency for Research and Cancer scale as Group 2B agent, which is considered as – "Possibly carcinogenic" to humans.[2] There is a dearth of studies on health hazards of mobile phone radiation and numerous studies do substantiate the genotoxic effect of mobile phone radiation. However, many have denied the same.[2]

Based on the data derived from previous literature, taking into consideration that oral cavity is in the closest vicinity to the area of mobile phone while in use, and the increased probability of oral mucosa presenting with genotoxic changes, the current study was formulated.[2] The presence and extent of chromosomal damage can be determined by measuring the Micronucleus (MN), damage to DNA or chromosomal misalignments are manifested as micronuclei, which is regarded as an accurate marker for genotoxic damage.[2]

Micronucleus (MN) is eccentric chromosomes or chromatin fragments formed due to abnormal mitosis.[4] Microscopically, they appear as visible chromatin mass in the cytoplasm near the nucleus with no direct communication with the nucleus. The presence of increased number of micronuclei is an indicator of DNA damage.[5] The increased frequency of micronuclei in chronic inflammatory diseases reflects the amount of DNA damage.[6]

The micronuclei (MN) assay in exfoliated buccal cells is a useful and minimally invasive method for monitoring genetic damage in humans. The aim of this study was to evaluate the extent of genotoxicity in oral epithelial cells by estimating the number of micronuclei in exfoliated buccal mucosal cells of analog and smartphone users.

Materials and Methods

The study protocol was approved by the Institutional Ethical Committee. A total of 15 males and 15 females between the age group of 45 and 55 years were selected from the Outpatient Department of Thai Moogambigai Dental College and Hospital, Chennai, Tamil Nadu, India. The participants were divided into two groups. In Group 1, 15 participants used analog phones or basic mobile phones and in Group 2, 15 participants used smartphones were selected.

The study group includes healthy individuals in the age group of 45–55 years, individuals with continuous usage of mobile phones for 4 years or above and participants who are right-handed, who use right side while talking in mobile phone were included in the study. Participants having any systemic diseases, oral lesions, and mobile phone usage of <4 years have been excluded from the study. Individuals using hands free or air pods are also excluded.

After obtaining written informed consent from participants, exfoliated buccal mucosal cells were collected from both left and right buccal mucosa. Before sample collection, subjects were asked to rinse their mouth with 1% glacial acetic acid to remove saliva, food particles, and any other debris. A moistened wooden spatula was used to collect the exfoliated cells from the buccal mucosa and was fixed, and the slides were stained with Papanicolaou nuclear staining solution. Two cytological smears were collected from each participant (right and left buccal mucosa).

All the slides were observed under low power (×10) for screening of micronuclei and high (×40) for the counting of the same. A total of 500 cells from right buccal mucosa and 500 cells from left buccal mucosa were assessed for the presence of micronuclei and the mean count was calculated.

The following criteria by Tolbert *et al*. were considered to designate any extranuclear body detected as micronucleus:[9]

- Rounded smooth perimeter suggestive of a membrane
- Less than a third the diameter of the associated nucleus but large enough to discern shape and color
- Staining intensity similar to that of the nucleus
- Texture similar to that of nucleus
- Same focal plane as nucleus
- Absence of overlap with, or bridge to, the nucleus.

Results

The mean micronuclei count in analog mobile phone users was mean ± standard deviation (SD) = 29.43 ± 6.163 whereas in smart mobile phone users was mean ± SD = 33.77 ± 7.977 (P = 0.022*). The mean micronuclei count in right side (frequently used) mobile phone users was mean ± SD = 37.7 ± 7.9 and in the left side was mean ± SD = 25.5 ± 3.37 (P < 0.001*). The mean micronuclei count was increased in smartphone users and also in frequently used side. There was significant difference in mean micronuclei count when we compared between phones and also used sides [Tables 1, 2 and Figures 1-6].

Discussion

Mobile (cellular) phones have been in use since the early 1980s and research into their potential health hazards has been carried out with increasing intensity for decades.[8] Between 1999 and 2004, there were six reports that directly addressed the issue

Table 1: Mean micronuclei count in different phone types and also used sides (right and left side)

	n	Mean MN count	SD	*P*
Phone type				
Basic phone	30	29.43	6.163	0.022*
Smartphone	30	33.77	7.977	
Buccal smears				
Right side (frequently used side)	30	37.70	4.793	<0.001*
Left side (other side)	30	25.50	3.371	

*P<0.05 significant. SD: Standard deviation, MN: Micronucleus

of rodent brain cancer and the type of radio frequency energy used by mobile phones.[10] These mobile phones radiate an average power of 0.2–0.6 w/kg, in which 40% is absorbed in the head-and-neck region thus resulting in various serious complications.[1]

Table 2: Comparison of mean micronuclei count between basic phone and used sides and also between smartphone and the used sides

Types of phone	*n*	Mean MN count	SD	*P*
Basic phone				
Right side (frequently used side)	15	34.6	3.481	<0.001*
Left side (other side)	15	24.27	3.058	
Smartphone				
Right side (frequently used side)	15	40.80	3.858	<0.001*
Left side (other side)	15	26.73	3.305	

*P<0.05 significant. SD: Standard deviation, MN: Micronucleus

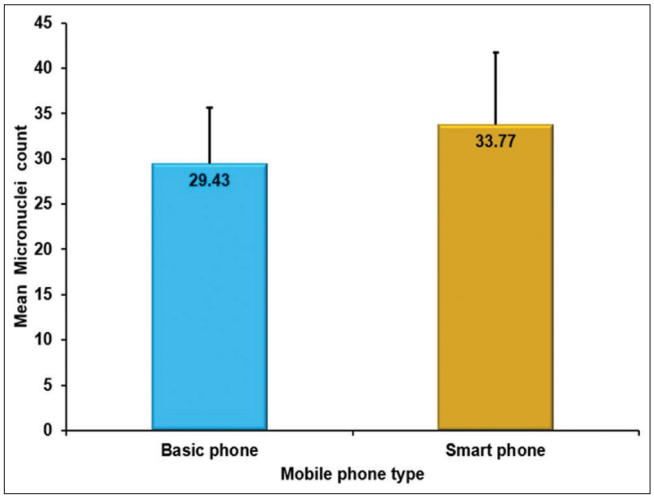

Figure 1: Mean micronuclei count of basic phone and smartphone users

Radiofrequency ranging from 100 kHz to 300 GHz can generate electromagnetic fields that may affect living cells through thermal or nonthermal mechanisms, although usually only at very high exposure levels. DNA damage may lead to cell senescence, cell death, or malfunction. DNA damage caused by genotoxic agents can be detected by cytological observation of morphologic changes such as micronuclei which are one of the biomarkers of environmental genotoxicity and cancer risk.[7]

The findings of the present study are in accordance with that reported by Vanishree *et al*. who found a statistically significant difference in the mean micronuclei count between high mobile phone users when compared to low mobile users.[1] Our results also corroborate with the findings of Banerjee *et al*. who have evaluated the mean micronuclei count between high and low mobile phone users in a larger sample size.[2] However, both studies yielded a statistically significant difference in the mean micronuclei count between the side of frequent usage of mobile phones and the opposite

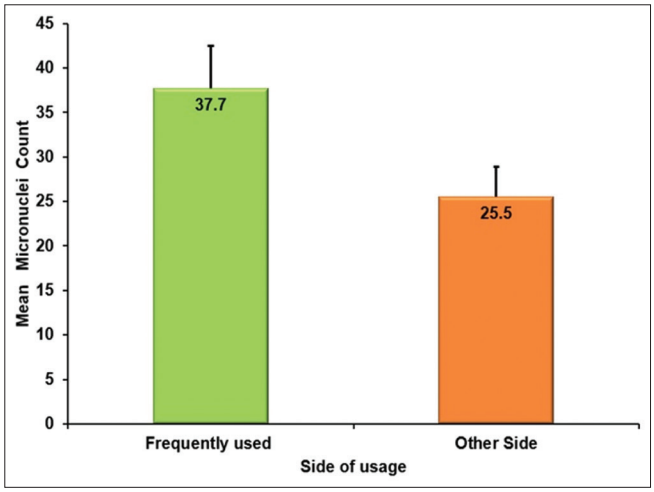

Figure 2: Mean micronuclei values of basic and smartphone users with respect to sides

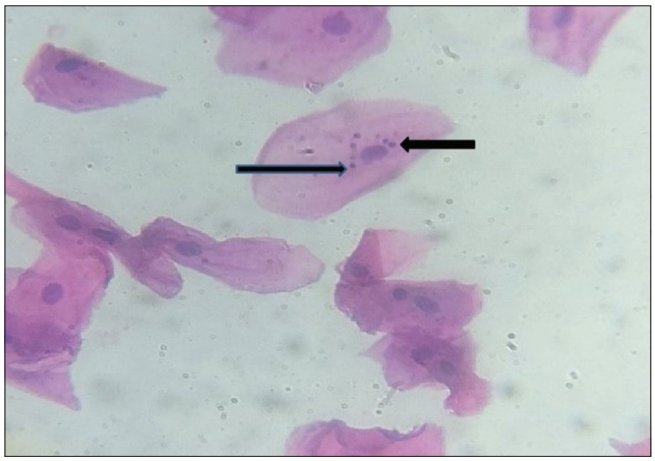

Figure 3: Micronuclei[arrow] in oral exfoliated cells of Smart phone users [Frequently used side]

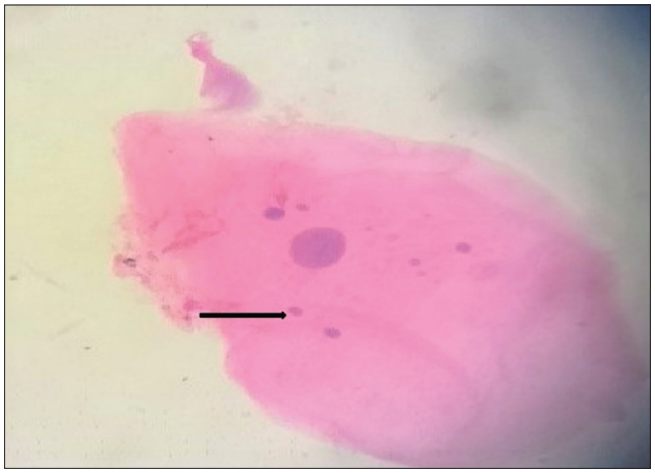

Figure 4: Micronuclei[arrow] in oral exfoliated cells of Smart phone users [Other side]

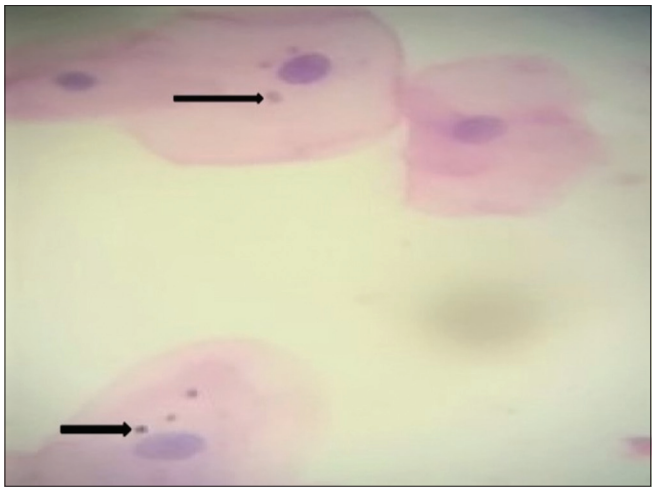

Figure 5: Micronuclei[arrow] in oral exfoliated cells of Basic phone users[Frequently used side]

side. In case of our study, we also concluded that the results revealed a statistically significant difference between the sides of frequent usage and the opposite side. These types of positive result show that further studies are required to completely elicit the elaborate effects associated with the long-term usage of mobile phones.

Our study recorded statistically significant change in micronucleus frequency according to basic and smart mobile phone type. This is in agreement with most of the studied that have used oral exfoliated buccal mucosal cells for micronucleus test. We have also compared the results according to the side of the face on which mobile phones were placed and found that there was a significant increase in the number of micronuclei in dominant side of face which is usually right side of most of people when compared to other side of the face. Incorporation of participants more than 40 years might be considered as a limitation for the study due to the age which is highly significant for the micronucleus frequency. On the contrary, other research groups have directly denied any significant increase of MN count in mobile phone users.[10-12]

Future research can be considered on the use of headphones with mobile phones. These wired headphones help to keep the device away from the body and prevent direct radiofrequency contact with the body. Long-term mobile phone users without headphones frequently experience a local temperature rise around the ear, which is decreased by using headphones. The local temperature increase may encourage heat stress-induced mitochondrial membrane damage, which in-turn leads to cytochrome c release, and caspase-9 and-3 activation,[13] which can be seen as a necessary step for cytotoxicity. The other variables such as brand of smartphones and amount of radiation of each brand can also be considered in further studies while evaluating micronuclei.

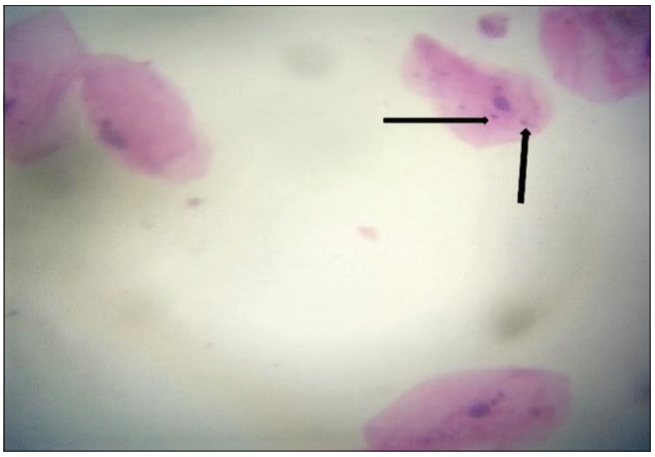

Figure 6: Micronuclei[arrow] in oral exfoliated cells of Basic phone users[Other side]

Conclusion

Individuals using high-end mobile phones are at higher risk for radiation-induced complications. Micronuclei assay in oral exfoliated cells is a noninvasive, cost-effective mass screening tool to detect genotoxicity.

Micronucleus assay of oral exfoliated cells can also be used a tool for education to encourage the practice of safety precautions in high-end mobile phone users such as usage of headphones for talking, usage of separate pouches for carrying mobile phones, and basically to maintain a safety distance with the radiation emitting devices.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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