Effects of mobile phone radiation on buccal mucosal cells based on specific absorption rate: a cross sectional study

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ABSTRACT

Background: The usage of mobile phone had been increased drastically which created a major health concern among the people about the radiation emitted from mobile phones. Hence this study was aimed to evaluate the Micronuclei (MN) frequency in exfoliated oral mucosal cells in high and low mobile users based on Specific absorption rate (SAR) value.

Methods: The total of 60 subjects were divided into two major groups: low SAR mobile phone users and high SAR mobile phone users. Further, Subjects who use mobile phone for more than 3 hours a week was considered as high talk time users and less than 3 hours a week was low talk time users. The buccal mucosa cells extracted by slightly scraping the oral cavity with a wooden spatula. For staining, Giemsa stain was used. Micronuclei were evaluated in 1000 cells per individual at the microscope.

Results: The result show prolonged talk time may interfere with the development of micronuclei in individuals who use mobile phone for more than 3 hours per week rather than high SAR value showing a significant increase in frequency of micronuclei formation.

Conclusions: The study showed mobile phone radiation had adverse effects on buccal mucosal cells.

Keywords: Mobile phone, Radiation, Micronuclei, Buccal mucosa

INTRODUCTION

Over the past decade, the use of mobile phones has increased drastically. The rapid increase in the use of mobile phone has raised the health concerns about potential risks associated with exposure to electromagnetic fields produced by the mobile phone radiation.1

In India, according to Telecom regulatory authority of India, New Delhi, September 2019, total wireless Telephone Subscribers were 1168.31 Million with an increasing monthly growth rate of 0.24%.2 As of present scenario, mobile phones are used at an enormous number by all the age-groups, particularly highest in the age group of 25-34 years.3 According to Dagli et al, it has been noted that an average person spends 90 min a day on their phone which is used not only for having conversations but also for various purpose.4

Mobile phones emit electromagnetic radiation in the microwave range 300 MHz and 300 GHz.2 The rate at which the electromagnetic radiation is absorbed by human bodies is called as Specific Absorption Rate (SAR). It is a standardized unit which measures the impact of radio frequency electromagnetic waves on the human body and it is expressed as Watt/kg. The FCC (Federal Communication Commission) has limited the maximum legal SAR of any handheld cell device to 1.6 Watt/kg in India.5
According to International Agency for Research on Cancer, the mobile radiations are classified as Group-2B - possibly carcinogenic radiations i.e. there “could be some risk” of carcinogenicity. This leads to the concern of possible adverse health effect on mobile phone users from exposure to electromagnetic radiation.

The possible oral cell changes, according to Fenech et al include DNA damage like micronuclei or nuclear buds, cytokinetic defects like binucleated cells, proliferative potential, and/or cell death have been related to a high risk of cancer, aging and to the carcinogenic process.

The cytomorphological changes, such as micronuclei (MN), is indicative of genomic damage, are biomarkers of genotoxicity and can be tested in exfoliated cells, especially in oral mucosal radiation.

As a matter of fact, the oral mucosa is the tissue that is present close to mobile phone while in use and has chances to show possible genotoxic changes by the mobile phone radiation. Few authors like Daroit et al, Banerjee et al, have confirmed the genotoxicity of mobile radiation on oral mucosa, while authors like Ros-Llor et al, Hintzsche et al had apparently denied the genotoxic effect of mobile phone radiation.

Considering all these mixed views, re-evaluation of the effect of mobile radiation on the oral epithelium is warranted. Hence this study was designed to evaluate the MN frequency in exfoliated oral mucosal cells in high and low mobile users based on SAR value.

**METHODS**

The cross-sectional study was approved by the Institutional Ethical Committee and the samples were collected as per the ethical guidelines and with prior consent of mobile phone users. A pre tested structured questionnaire was filled up with information including demographic details, cell phone model, talk-time period for past 1 week, SAR of the model and brand in use, age, occupation, diet, disease (if any), addiction (if any), allergy etc. Exposed individuals included both males and females who use smart phone.

Sample size calculation was performed according to study conducted by Daroit et al. Considering a study with an 80% power and α=0.05, the minimum sample size was arrived as 54 individuals using G-POWER software version 3.1.

**Inclusion and exclusion criteria**

Subjects were selected in the age limit of 19-33 years, owning a mobile phone whose specific absorption rate was in the range of 0.1 to 1.6 watt/kg of body weight with mobile phone use for minimum of 5 years and who gave consent to participate were included in the study. Subjects who had any history of long term medications, deleterious oral habits and subjects who used hands free devices like microphone or Bluetooth for voice calls were excluded from the study.

**Sample collection**

A total of 60 subjects between the age group of 19-33 years were selected. Participants were grouped into two groups based on the mobile SAR value (SAR value between 0.1 to 0.8 watt/kg (Group I) and SAR value between 0.9 to 1.6 watt/kg (group II)). Further these groups were sub grouped into low and high mobile talk time users. Low talk time users used mobile phone for less than three hours a week as talks time. The high mobile phone users, used mobile more than 3 hours a week as talk time.

**Scoring of micronuclei**

The criteria for identifying and scoring of Micronuclei (MN) were based on the proposed description by Tolbert.

The MN has rounded smooth perimeter suggestive of the membrane. It stains in the same intensity as the nucleus. It is located within the cytoplasm of the cell, and usually, the diameter is 1/3 to 1/6 of the nucleus. It has texture similar to nucleus. It is located in the same focal plane as nucleus.

From each slide 1000 cells were observed under microscope under 400X magnification for MN identification.

**Smear collection**

The subjects were initially asked to rinse the mouth with water to remove the debris. Exfoliated cells from the buccal mucosa were collected using a moistened wooden spatula. An attempt was made to collect samples on the preferential side used during phone calls and the cells were spread evenly on clean microscopic glass slides and air dried. The samples were fixed using 95% isopropyl alcohol and the slides were stained with Giemsa staining. Giemsa stain is easy to prepare, less time consuming, reduces the effects of poor techniques and increases cell yield.

Quantification was performed by a single blinded and calibrated observer. The nuclear changes were quantified as follows: micronuclei (round or oval structures with well-defined borders, having 1/3 to 1/6 the diameter of the main nucleus, staining intensity and texture similar to that of the main nucleus, found in the same cytoplasm of the main nucleus.

**Statistical analysis**

The nonparametric Mann-Whitney test was used to compare values of micronuclei at the buccal mucosa and SAR and Kruskal-Wallis test was used to compare the value of micronuclei between low and high talk time users with in the group. The Spearman correlation coefficient
was used to analyse data on weekly cell phone use and on the count of nuclear changes and SAR. Statistical analysis was performed using the Statistical package for the social sciences (SPSS) version 20.0. The level of significance was set at 5%.

RESULTS

We evaluated 60 healthy individuals. The study population consisted of 28.3% men and 71.7% women; mean age was 24.57 years, ranging from 19 to 33 years (Table 1).

<p>| Table 1: Characteristics of the participants. |</p>
<table>
<thead>
<tr>
<th>Variables</th>
<th>N (%) (n=60)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>17 (28.3)</td>
</tr>
<tr>
<td>Female</td>
<td>43 (71.7)</td>
</tr>
<tr>
<td>SAR</td>
<td></td>
</tr>
<tr>
<td>Low (0.1-0.8 watt/kg)</td>
<td>30 (50)</td>
</tr>
<tr>
<td>High (0.9-1.6 watt/kg)</td>
<td>30 (50)</td>
</tr>
<tr>
<td>Talk time (each group)</td>
<td></td>
</tr>
<tr>
<td>Low (≤3 hours/week)</td>
<td>15 (25)</td>
</tr>
<tr>
<td>High (&gt;3 hours/week)</td>
<td>15 (25)</td>
</tr>
</tbody>
</table>

| Table 2: Association between of specific absorption rate and talk-time in formation of micronuclei. |
| Variables | MN (Mean Rank) | P value | Correlation value | P value |
| SAR |  |
| High SAR | 21.02 | 0.000 | 0.551 | 0.000 |
| Low SAR | 39.98 |  |
| TALK TIME |  |
| Low SAR low TT | 19.10 | 0.000 | 0.619 | 0.000 |
| Low SAR high TT | 22.93 |  |
| High SAR low TT | 32.20 |  |
| High SAR high TT | 47.77 |

SAR=specific absorption rate, TT= talk-time, *non-parametric Man Whitney test and kruskal-Wallis test with spearman’s correlation

The sample was divided into two groups based on the mobile SAR value (SAR value between 0.1 to 0.8 watt/kg (Group I) and SAR value between 0.9 to 1.6 watt/kg (group II)). Mean age was 24.63 years in the first group and 24.5 years in the second one. A statistically significantly higher number of micronuclei were observed in the buccal mucosa in the group that used mobile with high SAR value compared to individuals who owned mobile phone with low SAR value (Table 2).

Another analysis was performed by splitting the groups in to low and high mobile talk-time users. Low talk time users used mobile phone for less than three hours a week as talk time. The high mobile phone users, used mobile more than 3 hours a week as talk time. Significant differences were observed between the low & high talk time and the micronuclei count and Spearman’s correlation was also performed to find the correlation between the SAR, talk time and micronuclei count (Table 2).

DISCUSSION

The rapid growth in the number of mobile phone users has raised questions about possible biological effects of the radiation emitted by these gadgets. The buccal mucosa is located within an area exposed to radiation emitted by cell phones; therefore, it is important to investigate its effects on buccal mucosal cells. The findings of this study suggest that long-term exposure to cell phone radiation and high SAR value can slightly increase the frequency of micronuclei.

The primary aim of this study was to evaluate the cytotoxic effects caused by mobile phone radiation based on the specific absorption rate in the buccal mucosa. Since the buccal mucosal cells would be highly exposed to radiations while talking.

In this study micronuclei count in exfoliated buccal mucosal cells were used to evaluate the genotoxic effect of mobile phone radiation. MN count in the exfoliated cells can be used as a marker for an abnormal cell cycle as it is formed as a result of aberrant mitosis when the whole chromosome or chromatid fragment fails to reach the spindle pole. It is one of the best indicators of mitotic interference and chromosomal mutations or breakage.5 The MN index is preferable for mass screening as it is rapid, simple, sensitive and cost-effective.10 Hence, in the present study, MN index was used to analyse the genotoxicity caused by mobile phone usage based on the specific absorption rate. Counting of MN is very technique sensitive, and different staining methods cause significant variations for the evaluation of its frequency.11 Hence, considering all the above factors, Giemsa staining was used to stain as it is single step, cost-effective procedure provides better stain intensity and fine cellular details and also supports the idea of utilizing this stain in oral exfoliative cytology for a decisive result.12

As stated in the result, MN count was found to be significantly higher in high SAR mobile phone users in comparison to low SAR users, and in the high talk time users, which directly indicates the genotoxic effect of prolonged mobile phone use for longer period. In our study, all probable causes for the increase in the MN count were excluded (tobacco, alcohol, recent medication, systemic factors etc.). Therefore, mobile phone radiation was expected to be the immediate and possible cause for the increased MN count in mobile phone users with high SAR, and the talk time of this finding was similar to various research groups.1,5,11 On the contrary, other research groups have directly denied any significant increase of MN count in mobile phone users.7,13 In this
study, the high and low mobile phone users were also evaluated by specific absorption rate of the mobile phone. This result indicates that when mobile phone used within the permissible range with the low SAR strength of radiofrequency radiation is not a major factor for genotoxic damage.

On the other hand, the increase in the micronuclei count can also be directly associated with localized hyperthermia. So, the increased micronuclei count in subjects complaining of warmth around the ear may be synergistic effects of both mobile radiations induced genotoxicity and local thermal effects. In our study any visible oral mucosal changes were not observed but it should be taken into consideration that mobile phones are being used extensively by the general population and increased micronucleus count indicate that in future days it may cause visible oral lesions.

A study conducted by Kesari et al reported an increase in the MN count, caspase 3 levels and apoptosis rate in the mice model due to 3G mobile phone exposure for two hours a day for 60 days.

Yadav and Sharma found an increased frequency of micronucleated exfoliated cells in 85 cell phone users compared to 24 non-users (controls). Gandhi and Prabhjot also found a positive correlation between the number of micronuclei and increasing exposure to cell phone radiation, comparing 25 users with 25 controls. Souza et al did not find any correlation between micronuclei and radiofrequency exposure; however, this study reports an increase of broken eggs in the group with greater exposure. Meanwhile, the studies conducted by Hintzsche et al and Ros-Lior et al did not find such correlation.

Previously studies have been reported on mobile phone radiations on Peripheral blood lymphocytes by Bisht et al, and Agarwal et al, showed harmful effects of cell phone radiation on sperm function and reactive oxygen species. Non-thermal DNA breakage in human fibroblasts as well as rat granulosa cells in vitro due to exposure to cell phone radiations were also reported by Diem et al in 2005.

In the present study Buccal Cytome Assay was done as described by Tolbert et al in 1992 are being reported and also the exposed individuals have been grouped according to the mobile phone SAR and talk time. This is a direct in vivo study on the effect of cell phone radiations on buccal cells. Our results showed significant increase in the micronuclei in samples using high SAR mobile phone compared to the Low SAR mobile users, and the increase was directly proportional to the duration of calls, as seen.

The complete avoidance of the mobile phone technology is not possible in the present world scenario but with few precautionary steps such as: keeping the mobile phone away from the body when not in use; use of headphones and bluetooth; keeping the phone in switched off mode when possible and mainly selecting a mobile phone of low SAR may help us to reduce the deleterious effects of mobile radiation.

**CONCLUSION**

The results of our study showed significant increase in formation of micronuclei after exposure to mobile phone radiations. Also, a strong co-relation was observed between the micronuclei formation and mobile phone exposure time. Based on the present findings, we suggest that Mobile phones when used for prolonged periods can cause genotoxicity. Although according to the SAR values most of the mobile phones emit radiofrequency radiation within safety limit but the prolonged talk time may interfere with the development of micronuclei in individuals who use mobile phone for more than 3 hours per week rather than the high SAR value. This can be used as an endpoint to detect the cytotoxic damage in exposed individuals and this information can be helpful as an early sign of potential risk of genetic damage due to mobile phone radiation. Counselling and awareness of the mobile phone users becomes vital to protect them against damaging effects of mobile phone emitted radiations.

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**Conflict of interest:** None declared

**Ethical approval:** The study was approved by the Institutional Ethics Committee

**REFERENCES**


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