Comparative Evaluation of Micronuclei in Buccal Mucosal Smear of Mobile Phone Users: (Original Research)

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ABSTRACT

Introduction:
Mobile phones have become a necessity in recent times such that its users are estimated to be around 6 billion in number. This majority of population are constantly exposed to non-ionising electromagnetic radiation as transmission and reception of signals take place by transfer of radio waves through cellular base stations.

Aim:
The aim of the study was designed to evaluate the occurrence of micronuclei (MN) in the buccal mucosal exfoliated cells of mobile phone users and to compare the micronuclei occurrence frequency between headset and non-headset mobile phone users which is the need of the hour in the current scenario.

Objectives:
- To evaluate the occurrence of micronuclei (MN) in the buccal mucosal exfoliated cells of mobile phone users.
- To compare the micronuclei occurrence frequency between headset and non-headset mobile phone users.

Materials and Methods:
The study population consisted of 100 participants between the age group of 18-22 years. Group A had 50 subjects who were not using headset for mobile phone usage and Group B had 50 subjects who were using headset for mobile phone. All subjects were verbally explained the nature of the study and an informed written consent was obtained (as per Helsinki declaration). Cell sampling was done on the slides and all the slides of 100 subjects were observed for a total of 1000 cells for the number of MN in each cell.

Conclusion:
Mobile phone radiation can cause significant genotoxicity, when used for a longer duration. The level of genotoxicity increases when mobile phones are frequently used on the same side so usage of headsets or using mobile phones in the contralateral sides can decrease the intensity of genotoxicity. Hence, headphone usage reduces the genotoxic effect to some extent.

Keywords: Cell, micronuclei, mobile phone, Papanicolaou

I. INTRODUCTION:
Electromagnetic fields (EMF) at extreme levels for longer duration are carcinogenic, as described by the International Agency for Research on Cancer (IARC). The mobile phone releases a power of 0.4–0.8 W, 30% of which is absorbed in the hands and the head due to its site approximation to the mobile phone.\(^1\) It will be considered as a “high intensity radio-transmitter” when used for long duration. Its emission is stronger than reaching the head of a user standing within 50 m range of a relay transmitter\(^1\).

The radiofrequency (RF) or microwave fields from natural terrestrial electromagnetic environment will not significantly comprise, however the artificial RF/microwave fields emitted
from wireless communication and the technology have very high intensity.[2]

The identity of the waveform has a specific frequency and wavelength, which is decided by the radiation. The specific absorption rate of tissue absorbed by a unit is known as electromagnetic energy. The whole-body threshold level of exposure in terms of SAR is 4 W/Kg was identified. The WHO stated mobile phone radiation on Cancer scale as Group 2B – “increased risk of glioma formation since it is possibly carcinogenic.”[1]

An association with gliomas with 10-year exposure criteria was found by Schuz et al. Papilloma occurrence was associated with ipsilateral mobile phone usage found by Hepworth et al. Mobile radiation on oral mucosa causes genotoxicity was confirmed by Sharma et al. Re-evaluation of the effect of mobile radiation on oral epithelium is needing considering all these.[3,4,5,6]

The micronuclei (MN) index in the buccal exfoliated cells was to evaluate the mobile phone radiation effect was conducted by Thomas et al., in 2009.[7] Hence, our present study was aimed to evaluate the occurrence of micronuclei (MN) in the buccal mucosal exfoliated cells of mobile phone users and to compare the micronuclei occurrence frequency between headset and non-headset mobile phone users which is the need of the hour in the current scenario.

II. MATERIALS AND METHODS

The study population consisted of 100 participants between the age group of 18-22 years. Group A had 50 subjects of who were not using headset for mobile phone usage and Group B had 50 subjects who were using headset for mobile phone. All subjects were verbally explained the nature of the study and an informed written consent was obtained (as per Helsinki declaration). Cell sampling was done on the slides and all the slides of 100 subjects were observed for a total of 1000 cells for the number of MN in each cell.

Inclusion criteria

The inclusion criteria of this study were as follows:
- Individuals in the age limit of 18-22 years
- Mobile phone usage for calls were considered
- Average usage of 4 hours per day was considered.

Exclusion criteria

The exclusion criteria of this study were as follows:
- Subjects having any deleterious habits
- Subjects having any oral lesions.

Cell sampling and preparation

Exfoliated oral mucosa cells were collected from all the subjects as per the protocol derived from the World Health Organization/International Agency for Research on Cancer guideline “Common Minimal Technical Standards and Protocols.” 15 Saliva samples were obtained in the morning after an overnight fast, during which subjects were requested not to drink any beverages except water. The subjects were given drinking water (bottled) and asked to rinse their mouth out well (without drinking water). 5 min after this oral rinse, a moistened wooden spatula was used to collect the exfoliated cells from the buccal mucosa and was fixed using cytotoxic, and the slides were stained with Papanicolaou nuclear staining solution. Two cytological smears were collected from each subject both from right and left cheek respectively.

Scoring Method:

The light microscope was used to examine the slides. Under low power magnification screening was done and under high power magnification counting the micronuclei in the cells was done. As per standard protocols Zig – Zag method was used for screening the slide. Intact cells with intact nuclei and cell borders were counted. A total of 1000 cells were counted from the smear in both the sides of the cheek buccal mucosa. Each subject’s slides were examined for the presence and number of micronuclei in each cell. Tobert et al criteria for designating an extra nuclear body as micronucleus includes,

- Micronucleus with rounded smooth perimeter of the membrane.
- Micronucleus with one third diameter of the associated nucleus
- Micronucleus with large enough to discern shape and colour.
- Micronucleus with staining intensity as same as that of nucleus.
- Micronucleus with texture same as nucleus.
- Micronucleus with focal plane same as nucleus.
- Micronucleus with absence of overlap
- Micronucleus with absence of bridge to the nucleus.

Statistical Analysis:

The frequency of micronuclei was recorded in percentage scale. Comparative evaluation was done with Unpaired t – test. Mean ± standard deviation was used for the representation of values. The P value was calculated by one-way analysis of variance using SPSS software and P < 0.05 was considered to be statistically significant.
III. RESULTS

In Group A, average number of micronucleated cells in the right side was 17.20, in Group B it was 22.27, with p value of 0.001, in Group A average number of micronucleated cells in the left side was 17.30, in Group B it was 22.00 with p value of 0.001, all of which were significant.

<table>
<thead>
<tr>
<th>Table 1-Micronuclei Count - Right side</th>
</tr>
</thead>
<tbody>
<tr>
<td>Micronuclei Count_RT</td>
</tr>
<tr>
<td>N</td>
</tr>
<tr>
<td>---</td>
</tr>
<tr>
<td>Head Phone Users</td>
</tr>
<tr>
<td>Non Head Phone Users</td>
</tr>
</tbody>
</table>

** Highly significant

Graph 1-Micronuclei Count - Right side

<table>
<thead>
<tr>
<th>Table 2-Micronuclei Count - Left side</th>
</tr>
</thead>
<tbody>
<tr>
<td>Micronuclei Count_LT</td>
</tr>
<tr>
<td>N</td>
</tr>
<tr>
<td>---</td>
</tr>
<tr>
<td>Head Phone Users</td>
</tr>
<tr>
<td>Non Head Phone Users</td>
</tr>
</tbody>
</table>

** Highly significant

Graph 2-Micronuclei Count - Left side

IV. DISCUSSION:

Mobile phones play an important role in our routine day to day life. Mobile phones were considered the mandatory gadgets even during the Covid pandemic situation it was used for children’s education for online classes and for work from home by the business people. Estimated mobile phone users were 4.93 billion in 2018 and with increase to 7.33 billion by 2022. Though various adverse effects have been reported that include physiological, behavioural, genetic changes, carcinogenesis potential and cognitive changes we all need mobile phones in our day to day life. MN counts in the exfoliated cells are used as a marker caused by aberrant mitosis in an abnormal cell cycle. Aberrant mitosis results in the failure of the entire chromosome to reach the spindle pole.
MN counts are the best indicators of interference in mitosis and chromosomal mutation or breakage. The advantages of the MN index includes its sensitivity, simplicity and cost effectiveness.\(^{[8]}\) In the present study the genotoxicity caused by mobile phone users were analysed by the MN index.

In the present study the count of MN was low in low mobile phone users in comparison to the high usage of mobile group. Questionnaire session was conducted on all the mobile phone users for the usage of wired headset and wireless headsets. There was a significant increase in MN count in non head phone users in comparison to the headset users. Headphones help to keep the mobile phones away from the body and direct contact of receiver is reduced. Local temperature around the ear was also getting reduced. Our study also proves a higher frequency of MN observed in habitually used side of mobile phones in relation to the excessive heat production due to phone battery which was noted in long term users. This excessive heat is also responsible for the genomic damage.\(^{[10]}\)

**Precaution:**
Increasing distance between the mobile phones and the user
- Using a head set / Blue tooth
- Switch off phone at night
- Usage of EMF (Electromagnetic shielding device.)

These precaution greatly tend to decrease the detrimental effect of radiation hazard produced by the mobile phones.\(^{[10]}\)

**V. CONCLUSION:**
Significant genotoxicity can be caused even with permissible range of radiation when used for increased time of usage of mobile phones. The accentuation of genetic damage was observed with mobile phone users which may be due to more amount of radiation and increase in the temperature due to longer duration of use of mobile phones. Mobile phone used with headsets will decrease the genetic damage to some extent. Hence it is advised to use the mobile phones with headsets preferably wireless headset to increase the distance between the mobile phone and the user and also to decrease the radiation exposure.

**REFERENCES**
[2]. Kuster N, Balzano Q, Lin J.