ABSTRACT

Introduction: The tremendous growth in telecommunication world has increased the number of mobile phone users to many folds. The benefits of cell phones are just half the picture. The other half may be a darker one. The main aim of this study is to observe the effect of electromagnetic radiation being emitted by mobile phones on adrenal gland microanatomy and the hormones released by the gland on male albino rats.

Materials and methods: Forty-two male albino rats of Spargue-Dawley species were divided into two equal groups. The experimental group rats were exposed to mobile phone radiation operating at 900 MHz while the control group rats were not. At the end of every two months, seven rats were sacrificed to analyse histological and hormonal changes.

Results: No changes were seen at the cellular level of the adrenal glands even after six months of radiation exposure. Hormonal assay showed a variation in the cortisol levels of the rats but the changes were within normal range.

Conclusions: Lack of appreciable changes in the cellular morphology and hormonal levels even after six months of radiation exposure signify that the adrenal glands are not affected by exposure to mobile phones.

Keywords: Mobile Phone, Albino Rats, Adrenal Gland, Cortisol.

INTRODUCTION

GSM stands for Global System for Mobile communications. It is a digital mobile telephone system used in most parts of the world. GSM uses a time division multiple access which enables more people to communicate simultaneously with a station. According to GSM world, there are now more than 3 billion GSM mobile phone users worldwide with China referenced as the largest single GSM market with more than 370 million users [1].
Tremendous growth in the use of cell phones has led to more demand for land to site telecommunication base stations and related infrastructures. However, as cell phone usage skyrockets, the concern over potential health risks rises as well. This is a consequence of the radiation emitted by handsets and base stations that receive and transmit the signals. Although some scientists and researchers across the globe claim there is no adverse effect from the use of mobile phones and its base stations, others in different countries in different laboratories, are finding disturbing results that point to far greater health implications than anyone is ready for [2].

Having suggested the potentially harmful effects of EMW radiation on some biological systems, recent studies have dealt with the concerns regarding the safety of radio frequency (RF)-EMW exposure. For example, the microwaves emitted by mobile phones have been linked to several genetic defects [3-5].

Microwave radiation may induce chromosomal instability and may lead to increased risk of cancer as suggested by Sykes et al. in 2001 and 2002, by Mashevich et al. in 2003 and Agarwal in 2007 [6-8]. Scientific researches have highlighted some extremely hazardous effects of exposure to radiation emitted from cell phones on the human body. These effects range from those at the molecular level manifested as an increase in single and double strand DNA breakages [9], change in Ornithine decarboxylase activity [10], increased risk of brain tumors [11] to disruption of learned behavior, dysaesthesia, etc. [12] and an increase in chick embryo mortality [13].

So according to the above effects of microwave radiation, the risk on adrenal gland as an essential organ becomes most important in the study of this issue [14]. Results from epidemiological studies indicate that cell phone radiation power density even below the standard level can cause symptoms such as headache, heat sensation in the ears, memory loss and fatigue and show significant relationship with the duration of call / time of the day [15-20].

Regarding the endocrine system, the sensitivity of pineal gland, pituitary gland, adrenal gland and thyroid gland as well as of the endocrine pancreas, testicles and ovaries to EMWs have been investigated [21]. Various papers were published on different endocrine glands but adrenal gland was the least explored. Hence, considering the lack of data on the effects of GSM mobile phone-induced electromagnetic fields on the adrenal gland and cortisol hormone in humans, the aim of the present study was to assess the potential alterations of cortisol hormones and microanatomy of adrenal gland after exposure to microwave radiation emitted by mobile phones.

**MATERIALS AND METHODS**

**VENUE:** The entire study was conducted in the department of Anatomy, BRD Medical College, Gorakhpur in collaboration with the department of Pharmacology of the institute. **STUDY MODEL:** Forty-two male albino rats of Spargue-Dawley species were chosen for the experiment. **INCLUSION CRITERIA:** A) Male rats B) Knowingly disease-free C) Age about 1.5 months-2 months D) Weight about 100-150 gms.
PROCUREMENT: All the rats were procured from Central Drug Research Institute, Lucknow along with their health certificates. After the procurement of the rats they underwent the process of acclimatization for one week during which they were fed with recommended pellet diet and water ad libidum. Twelve-hourly day and night cycle was maintained with appropriated temperature and humidity. Ethical clearance was taken from institutional animal ethical committee and experiment was performed as per the recommended guidelines.

GROUP DIVISION: The rats were divided into 2 groups having 21 rats each. Group A was the control group and Group B was the experimental group. Both the groups of rats were given exactly the same conditions and environment. The only difference was in the fact that Group-B rats were exposed to mobile phone radiation while Group A rats were not. In fact, the control group of rats were kept in different room in order to avoid any exposure to radiation. RADIATION SET UP: Plastic cages normally available for rat housing were used and the base of the cage was floored with thermocol sheet. A slot was cut at the corner of the cage in thermocol sheet to adjust the mobile phone in it. The cage was partitioned using a wooden plank in order to restrict the movement of rats during the experiment. A GSM model mobile phone with frequency bandwidth of 900 MHz and power of 2 watt with a SAR value of 0.38 W/Kg was used to carry out the experiment. When the experiment was ‘ON’, a call was made from another cell phone on the mobile phone being kept inside the cage and the call was received in order to make the phone on answering mode for the next one hour. During this period, the rats remained in touch with the mobile phone and were receiving the radiations just like humans [22-25]. The entire experiment was repeated for 6 months and at the end of every two months, seven rats from both the group were scarified and the tissue of adrenal gland and their blood sample were collected for analysis.

HORMONAL ANALYSIS: After the sacrifice of rats from both the groups, the blood sample of each rat was collected directly from the heart and was sent immediately to the lab for hormonal analysis of cortisol. Reports were collected and the data obtained was tabulated and analysed.

HISTOLOGICAL ANALYSIS: Just after the sacrifice of rats the adrenal gland was procured and was kept in 10% formalin solution. The tissues collected were processed using routine tissue processing technique used for Hematoxylin and Eosin staining methods. Slides were prepared and visualized under microscopes.

RESULTS

A. MICROSCOPIC ANALYSIS

CONTROL GROUP: (Fig. 1-a)
- The interior of the adrenal gland was divided into outer cortex and inner medulla.
- Cortex further exhibited three concentric zones: - zona glomerulosa, zona fasiculata and zona reticularis.
- Zona glomerulosa was a thin zone interior to the adrenal gland capsule. It consisted of cells arranged in small clumps.
- Zona fasiculata was intermediate and the thickest zone of the adrenal cortex. This zone exhibited vertical columns of one cell thickness adjacent to straight capillaries. It
consisted of pale staining cells due to increased lipid droplets.

- Zona reticularis was the innermost zone that was adjacent to the medulla. The cells here were arranged in cords or clumps.
- Medulla lied toward the centre of the gland, cells here were also arranged in small cords and were secretory in nature (epinephrine and nor-epinephrine).
- Medulla contained a lot of capillary network.

**EXPERIMENTAL GROUP:**

At the time of 2\textsuperscript{nd} month of sacrifice: (Fig. 1-b)

- No appreciable changes were observed after 2\textsuperscript{nd} month of radiation doses to the rats.
- The microscopic picture observed was more or less similar to the control group of rats.

At the time of 4\textsuperscript{th} month of sacrifice: (Fig. 1-c)

- No appreciable changes were observed after 4\textsuperscript{th} month of radiation doses to the rats.

At the time of 6\textsuperscript{th} month of sacrifice: (Fig. 1-d)

- No appreciable changes were observed after 6\textsuperscript{th} month of radiation doses to the rats.
- Lack of changes at the cellular level in adrenal gland even after the exposure of 6 months was a clear indication of the fact that whatever other changes in parameter observed were not because of stress. This fact was very well supported by the level of corticosteroid hormones which showed no rise during the experiment.

Fig. 1. Microscopic picture of Adrenal gland (40x) (a) Control group, (b) Experimental group at 2 months (c) Experimental group at 4 months and (d) Experimental group at 6 months
B. HORMONAL ANALYSIS

- The analysis done from the data obtained for the cortisol level suggested that there were no changes occurring in the level of cortisol throughout the experiment.
- After 2nd month of experiment, the tables and bar diagram showed that there was variation in the level of cortisol in both the groups of rats but the data obtained from both the groups remained within the normal range. (Fig. 2).
- The same result was obtained from the data analysis after four months (Fig. 3) and after six months of the experiment (Fig. 4).
- Whether it was the control or the experimental group of rats, the level of cortisol remained within the limits of normal range.
- This data also indicated that throughout the experiment there was no increase in the stress of rats.
- Hence the changes being observed at any level were not due to any stress to the rats.

DISCUSSION

Various researches have suggested that electromagnetic radiation emitted from base stations and cell phones have destructive effects on tissues in two ways. To begin with, warm impacts happen by means of increment in bodily warmth by electromagnetic vitality, which is consumed by the body. Second, non-warm impacts show up as changes in cerebrum capacities and headaches [26].

Studies concentrating on the impacts of EMR radiated by mobile phones have yielded dubious outcomes. Past investigations have detailed that radiofrequency waves produced especially by the third era mobile phones may affect the immunological status, sensory system, hematological status, cardiovascular capacities, urinary framework, typical development and genes [27], cells [28], tissues [29], organs and embryonic improvement [30]. Alongside this, EMR may likewise prompt DNA damage.

However, on the other hand there are studies in the literature reporting that cell phones do not have an effect on blood-brain barrier, testes, sperm morphology, semineferous tubules and Leydig cells of the rats and do not cause a significant change in mean fetal heart rate [31].

Similarly, in comprehensive studies conducted in the USA and Denmark, it was reported that cell phone use was not associated with increased risk of brain tumor [32], direct genotoxic, mutagenic or cytotoxic effects [33]. To date such studies reporting hazardous as well as non-hazardous effects have used different experimental animal models like rats [34], mice [35], chick [36], etc.

In our study, we investigated the effect of mobile phone radiation operating at 900 MHz upon one important organ, that is the adrenal gland, over a period of 6 months and found that there was not much appreciable change occurring in the histology or physiology of the adrenal gland. However, Sima and Imam et al found alterations in the cortisol level of rats after exposure for 6 hours daily for 8 weeks. They also reported that the zona fasciculata layer of adrenal cortex eventually thickened following mobile RF radiation. The quantity of cells in
Comparison of Cortisol level in Control and Experimental groups of rats after 2 months

<table>
<thead>
<tr>
<th>R.no</th>
<th>1st day</th>
<th>2nd month</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Experimental</td>
</tr>
<tr>
<td>1</td>
<td>0.29</td>
<td>1.42</td>
</tr>
<tr>
<td>2</td>
<td>0.31</td>
<td>1.38</td>
</tr>
<tr>
<td>3</td>
<td>0.44</td>
<td>0.76</td>
</tr>
<tr>
<td>4</td>
<td>2.01</td>
<td>0.82</td>
</tr>
<tr>
<td>5</td>
<td>1.46</td>
<td>0.97</td>
</tr>
<tr>
<td>6</td>
<td>0.91</td>
<td>2.10</td>
</tr>
<tr>
<td>7</td>
<td>1.78</td>
<td>1.21</td>
</tr>
<tr>
<td>Mean</td>
<td>1.028571</td>
<td>1.237143</td>
</tr>
<tr>
<td>SD</td>
<td>0.72299</td>
<td>0.46046</td>
</tr>
<tr>
<td>P value</td>
<td>0.5278</td>
<td>0.6676</td>
</tr>
</tbody>
</table>

Table 1. Comparison of Cortisol in Experimental and Control Group of rats after 2nd month

Fig. 2. Comparison of Cortisol in Experimental and Control groups of rats after 2nd month

(0.2-2.6) Normal cortisol level
0.2-2.6 µg/dl
Mobile Phone Radiation Effect on Adrenal Gland

Table 2. Comparison of Cortisol in Experimental and Control groups of rats after 4th month

<table>
<thead>
<tr>
<th>R.No</th>
<th>Control</th>
<th>Experimental</th>
<th>Control</th>
<th>Experimental</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.10</td>
<td>1.43</td>
<td>1.78</td>
<td>1.23</td>
</tr>
<tr>
<td>2</td>
<td>2.20</td>
<td>1.92</td>
<td>2.01</td>
<td>1.65</td>
</tr>
<tr>
<td>3</td>
<td>1.89</td>
<td>1.94</td>
<td>1.77</td>
<td>1.74</td>
</tr>
<tr>
<td>4</td>
<td>1.32</td>
<td>2.56</td>
<td>0.76</td>
<td>2.00</td>
</tr>
<tr>
<td>5</td>
<td>1.40</td>
<td>2.53</td>
<td>0.87</td>
<td>2.32</td>
</tr>
<tr>
<td>6</td>
<td>1.23</td>
<td>1.38</td>
<td>1.02</td>
<td>1.09</td>
</tr>
<tr>
<td>7</td>
<td>1.86</td>
<td>2.52</td>
<td>1.65</td>
<td>2.34</td>
</tr>
<tr>
<td>Mean</td>
<td>1.714286</td>
<td>2.04</td>
<td>1.408571</td>
<td>1.76714286</td>
</tr>
<tr>
<td>S.D</td>
<td>0.392762</td>
<td>0.512022</td>
<td>0.508312</td>
<td>0.49151854</td>
</tr>
<tr>
<td>Pvalue</td>
<td>0.1989</td>
<td>0.1987</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Fig. 3. Comparison of Cortisol in Experimental and Control groups of rats after 4th month
Table 3. Comparison of Cortisol in Experimental and Control groups of rats after 6th month

<table>
<thead>
<tr>
<th>R.No</th>
<th>1st day Control</th>
<th>1st day Experimental</th>
<th>4th month Control</th>
<th>4th month Experimental</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2.56</td>
<td>2.34</td>
<td>2.31</td>
<td>2.01</td>
</tr>
<tr>
<td>2</td>
<td>2.39</td>
<td>2.56</td>
<td>2.12</td>
<td>2.34</td>
</tr>
<tr>
<td>3</td>
<td>2.32</td>
<td>2.30</td>
<td>2.03</td>
<td>2.10</td>
</tr>
<tr>
<td>4</td>
<td>2.09</td>
<td>1.12</td>
<td>1.67</td>
<td>0.98</td>
</tr>
<tr>
<td>5</td>
<td>1.96</td>
<td>2.51</td>
<td>1.89</td>
<td>2.33</td>
</tr>
<tr>
<td>6</td>
<td>1.81</td>
<td>2.48</td>
<td>1.65</td>
<td>2.08</td>
</tr>
<tr>
<td>7</td>
<td>1.54</td>
<td>2.55</td>
<td>0.87</td>
<td>2.31</td>
</tr>
<tr>
<td>Mean</td>
<td>2.095714</td>
<td>2.265714</td>
<td>1.791429</td>
<td>2.02142</td>
</tr>
<tr>
<td>S.D</td>
<td>0.356224</td>
<td>0.515165</td>
<td>0.470334</td>
<td>0.478589</td>
</tr>
<tr>
<td>P-value</td>
<td>0.4811</td>
<td></td>
<td>0.3780</td>
<td></td>
</tr>
</tbody>
</table>

Fig. 4. Comparison of Cortisol in Experimental and Control groups of rats for 6th month
zona fasciculata stayed steady, in spite of the expanding phone size and edge during RF presentation.

Alteration in results may be due to the species of rats, their morphology, environmental conditions, amount and duration of exposure to radiation, etc. Any conclusive result to be quoted requires more extensive study with more parameters.

**CONCLUSION**

The results obtained from our experiment showed that even after an exposure of rats to mobile phone radiation for 6 months, there were no significant changes obtained in adrenal gland morphology at microscopic level and even the hormonal level. Though variation in the level of the hormone was very well reported but the data obtained after the analysis was all within the limit of normal range of cortisol level. This also proves that the rats were not stressed during the experiment and hence, any other parameter studied will have minimum chances of error due to stress factor.

**REFERENCES**


