Effect Of Electromagnetic Radiation On Vital Organs In Rats
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INTRODUCTION
Every technological development comes with some element of health concern, and cell phones are of no exception. The global system for mobile communications (GSM) standard operating in the 900 MHz frequency band is largely used, over the past decade. EMR are harmful to public health. In 2011, the World health organization, International Agency for Research on Cancer (IARC) has been identified electromagnetic from mobile phones and other wireless devices constitutes “a possible human carcinogen” 2B (IARC 2011). EMR may be absorbed by various body organs according to places the mobile are carried especially liver and kidney (Topali et al. 2015).

Electromagnetic radiations affect biochemical processes of all the vital organs via causing alterations in the cellular membrane potential that further disturb the regular distribution of ions and dipoles (Lerchl et al. 2002; Illhan et al. 2004; Nawal et al. 2016), the causative factor in every exposed associated organ dysfunction found to be similar. (Huber et al. 2002; Krause et al. 2007; Hao et al. 2015) neurotransmitter systems (Mausset-Bonnefont et al., 2004), and blood-brain barrier permeability (D’Andrea et al. 2003; Reddy et al. 2016). The toxicity of 900 MHz electromagnetic produced by mobile phones has been previously analyzed in vital organs (Acar et al. 2009). EMF has been reported to be genotoxic and carcinogenic (Gandhi et al. 2005). There is an increased risk of several other types of cancers following prolonged exposure to mobile phone/ tower radiation, such as, salivary gland tumors, uveal melanoma, lymphoma, facial nerve tumors, skin, blood, testicular and breast cancer (Elisabeth et al. 2011; denis et al. 2011; Kundi et al. 2009; Ahlborn et al. 2004).

This study analyzed mobile induced hazards in terms of antioxidants level, enzymological activity, biochemical parameters, and hematological profiles of various organs. The understanding of toxicological effects of EMR radiation in a mammalian model is necessary for studying human health hazards and could help in the establishment of guidelines for the safe use of the mobile phones.

MATERIALS AND METHODS

Animals and Chemicals
Male wistar rats, approximately 120±10 g body weight, were used for the study. Before the tests, they were acclimatized to laboratory conditions (12-h light/12-h dark cycle, 22°C temperature) for a minimum of 10 days. The rats were divided into two groups; each group contained six male rats housed in cages that have sterile pady husk as bedding materials. Standard food pellets (provided by the animal facility) and clean tap water were provided ad libitum. The experimental protocols were carried out according to the approved guidelines set by the Institutional Animal Ethics Committee.

Experimental Design

Animals were allocated into two groups: group I (n = 6), control without near source of electromagnetic radiation; and group II (n = 6) were exposed to the mobile phone radiation 2h/day constantly for 90 days.

Exposure system
The electromagnetic radiations were produced by a real GSM mobile phone electromagnetic radiation. Mobile phones operated by local network at 900 MHz frequency band and specific absorption rate (SAR) equivalent to 1.10 W/kg. The mobile phone was put in a small perforated polycarbonate cell in the center of cage in order to prevent any damage caused by rats. A call was given with another mobile phone and it will ensured that mobile phone is switched on and with call accepting mode and rats is in close proximity to the mobile phone. False mobile phones will put in the control. Conditions of exposure were similar to those to which a mobile phone user is exposed and were determined according to previous study (Motawi et al., 2014).

At the end of each experimental period rats were sacrificed and the blood was drawn from the animal’s eye by puncturing the retro-orbital venous sinus. The brain, liver, and kidney were dissected out, rinsed in a cold 0.9% sodium chloride solution, placed in pre-chilled homogenization buffers for further procedure.

Indices of oxidative stress
Tissue lipid peroxidation (LPO) was measured by estimating the thiobarbituric acid reactive substances (TBARS) as described by Sharma and Krishna-Murti (1968). Tissue-reduced glutathione (GSH) was measured following the method described by Brehe and Burch (1976).

Assay on antioxidant and associated enzymes
Catalase (CAT) activity was estimated following Aebi (1984). Total superoxide dismutase (SOD) activity was estimated by measuring the inhibition of the auto-oxidation of epinephrine according to the method described by Misra and Fridovich (1972). Tissue glutathione peroxidase was measured following the method described by Paglia and Valentine (1976).

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Blood biochemistry
Serum was isolated from the blood samples by centrifugation at 2000 r/min for 15 min and was collected in vials and used to determine the levels of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) following the methods described by Reitman and Frankel (1957). The levels of albumin, bilirubin, creatinine, urea, and uric acid in the serum were determined by standard procedures used in clinical biochemistry laboratories based on manual biochemical kits following manufacturer’s
directions (E-Merck Limited, Mumbai, India) and using an auto analyzer (Micro Lab 200, Merck, Germany).

**Hematological analysis**
Determination of hematological parameters (RBCs, WBCs, Hb concentration and platelets count), by an automated hematologic analyzer using whole blood sample.

**Statistical analysis**
Values are expressed as mean ± SE for six rats in each group and significance of the differences between mean values were determined by the Student’s t-test according to the Snedecor and Cochran (1994). The minimal level of significance was considered at $P \leq 0.05$.

**RESULTS**

**Oxidative stress marker in tissue**

**Lipid Per-oxidation and Reduced glutathione**
Lipids are susceptible to oxidation and lipid peroxidation products are potential biomarkers for oxidative stress. A significantly elevated LPO level follows electromagnetic radiation exposure for 3 months in all tissues (Fig 1A). Enhanced LPO was expressed in terms of thiobarbituric acid reactive substances in electromagnetic exposed groups. However a considerable slump in the reduced glutathione contents of liver, kidney and brain after EMR exposure (Fig 1/B).

**Antioxidant and associated enzymes in tissue**
A fall was observed in SOD activity in both exposed group which indicates susceptibility of vital organs against ROS produced by EMR (Fig 1/C). The defensive antioxidant enzyme next to SOD is catalase. CAT traps the harmful hydrogen peroxide and converts into water and oxygen. A mixed trend was observed in CAT activity. EMR exposure enhanced CAT activity in brain and liver while depleted activities observe in kidney tissue (Fig 1/D). Glutathione peroxidase plays important biochemical role in reducing most organic and inorganic hydroperoxides in many tissues and cells. Electromagnetic radiation exposure significantly inhibited the enzymatic activity of GPx in liver, kidney and brain (Fig.1/E). In exposed group decrease in GPX activity observed statically significant compared with control ($P \leq 0.05$).

![Graphs showing oxidative stress markers and antioxidant enzymes in tissues](image1.png)

Figure 1. Effects of microwave radiation oxidative stress markers and antioxidant enzyme in rat liver, kidney, and brain. (a) representation of lipid peroxidation (B) Representation of reduced glutathione (C) ) Representation of superoxide dismutase (D) ) Representation of catalase (E) ) Representation of glutathione peroxidase. Value represent the mean±SE of six animal in each group. * Exposed vs control significant at $P \leq 0.05$.

![Graph showing oxidative stress markers in blood](image2.png)

Figure 2. Effects of microwave radiation on oxidative stress markers in blood. GSH; Reduced glutathione, LPO; Lipid per oxidation, CAT; Catalase. Value represent the mean±SE of six animal in each group. * Exposed vs control significant at $P \leq 0.05$. 

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Sharma et al., 2017 / Effect Of Electromagnetic Radiation On Vital Organs In Rats

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Table 1: Depicts effect of electromagnetic radiations produced by mobile phone in serum biochemical parameters in rats.

<table>
<thead>
<tr>
<th>Liver function test</th>
<th>Kidney Function test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Groups</td>
<td>AST (IU/l)</td>
</tr>
<tr>
<td>Control</td>
<td>42.0±2.3</td>
</tr>
<tr>
<td>Exposed</td>
<td>65.1±3.5*</td>
</tr>
</tbody>
</table>

Data are mean ± SE, N=6;Exposed Vs control at p<0.05.

Blood Biochemistry
Changes in the levels of enzyme activity such as ALT and AST are presented in table. ALT and AST levels in rats exposed to EMR were significantly higher than the control (p≤0.05). Variations in biochemical parameters such as uric acid, urea, creatinine, albumin, and bilirubin levels indicate kidney dysfunction. Serum uric acid, urea, creatinine and bilirubin levels in experimental rats were higher than the control group (p≤0.05), whereas a significant depletion in albumin level in serum (Table 1) exposed to EMR compared with the control group (p≤0.05).

Hematological parameters
From a biological point of view, blood can be considered as a tissue comprising various types of cells (Hb, RBCs, WBCs and platelets) and a liquid intracellular material (Plasma). A significant effect in blood components due to the exposure to mobile phone electromagnetic radiations. Value of Hb, RBCs, WBCs, and platelets decrease significantly in exposed group compared to control group (Fig. 2).

DISCUSSION
The widespread mobile phone usage has raised the possibilities of inevitable exposure of electromagnetic radiation (EMR). Previously, there were several apprehensions were established in explaining the ill-effects of EMR on human health (Goldwein and Afinan 2010). Despite the several research related to harmful effect of mobile radiations toxicity in vital organs like brain, kidney, and liver is still poorly elucidated. Therefore, in this present study, we have investigated the effect of EMR in terms of antioxidants level, enzymological activity, biochemical parameters, and hematological profiles of various organs and the possible deleterious effects of EMR.

Alterations in the level of antioxidants, biochemical molecules, enzymological activity and hematological profile are well considered as the indicators of deteriorating animal homeostasis that further resulted in stress and decline functional ability (Gecti et al. 2014; Saravanan et al. 2012). Hence, we hypothesize an insufficiency of antioxidant defense capacity is a pathogenic pathway involved in all the organ dysfunction or disorders (Saravanan et al. 2012). Ample of studies have shown that EMF exposure is capable of causing the substantial oxidative damage to the body (Sharma et al. 2014; Aydin and Akar et al., 2011; Maaroufi et al., 2014). Similarly in our study we observed a regular alteration in the level of oxidative molecules like GSH, LPO, CAT, GPX and SOD in all the vital organs. Additionally the estimated level of GSH and LPO was also altered in blood directing towards the continual level of oxidative stress in the systemic flow.

In the present study the high level of oxidative stress in brain tissue account for the higher vulnerability of brain towards the EMR exposure. Theses results coincide with findings of Ragy et al. (2014).

Furthermore, we have also assessed the liver and kidney toxicity following EMF exposure. In response to any cellular injury in liver the level of intracellular enzymes, like serum alkaline phosphatase, transaminases and bilirubin get increased in the circulation (berrahal et al. 2009). During liver damage the elevated level of enzymes like ALT and AST are considered as the preliminary indicators of the structural and functional damage of both cellular and mitochondrial membranes as observed in this study following EMF exposure (rajesh and Latha 2004; Sharma et al. 2013). Serum urea, uric acid and creatinine were also found to be higher, directing towards renal damage, which also corroborates with other study (Moussa 2009; Tsi et al. 1998). These increments could be considered as a reflection of deteriorating renal performance (Gericat et al. 1990) due to the ammonia formed by deamination of amino acids in the liver which converted to urea or to increased breakdown of nucleic acids (Yamonenko 1998). Since irradiation may cause breaking of DNA molecules and destruction of their bases (the purines) which may be catalyzed into uric acid (Ganong 1992). As creatinine is formed largely in muscles and occurs freely in blood plasma and urine, its increased levels in plasma serve as an index of renal function impairment. Additionally, the abnormalities in hematological profile in terms of RBC counts, haemoglobin and monocyte count as observed in this study also provides corroboration to the detrimental effects of EMF exposure on blood. This means that these components of blood are broken due to irradiated by Mobile phone electromagnetic radiation. The decrease in the Hb may be associated with the hemolytic anaemia or the suppression of the bone marrows. Alongwith the apparent significant effect on bilirubin (a byproduct of red blood cell lysis) prove the possibility of hemolysis. Previously El-Bedawi et al. (2011) also reported an increased in the level of bilirubin in rats after exposure to mobile phone.

We would highlight that in this study to generate the reality of the human exposition the exposure was provided by the commercial cellular phone. Present study showed an evident oxidative insult in liver, kidney and brain. Most of the studies regarding EMF effects were targeted on a particular organ or mechanism. Here in this study we propose that during EMF exposure due to excessive mobile usage various vital organs get equally affected with the similar blood stream. Present study will provide an effectual consideration in developing therapeutic strategy regarding multiple organ conditions and relating mechanisms during EMF exposure. This study provides an insight regarding mobile emitted radiations could induced alteration in blood as well in vital organs like liver, kidney and brain tissue.

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