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Research Paper

Effects of Exposures of Mobile Phone Radiation on Cellular Architecture and Redox Status of Mammalian Brain Tissues

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Abstract

Several reports have described the potential deleterious effect of radiofrequency electromagnetic radiation (RF-EMR) from mobile phone on cerebral redox status. However, the frequency and duration of exposure to such radiation in real life situation can vary widely thus suggesting that experimental simulation and evaluation of mammalian interaction with RF-EMR is seemingly endless and thus very open. In this study, Male Wistar albino rats were continuously exposed to RF-EMR with frequency 900 MHz for 0, 4, 8 and 12 h per day for 30 days. Thereafter, the enzymic and non-enzymic antioxidant defense statuses and histology of the brain were evaluated. The results showed that frequent exposure to RF-EMR diminished the antioxidant defense status and evoked distorted structural integrity of brain cells and this effect was exacerbated with increased daily duration of exposure. Within the limit of the present data, it appears that unwitting frequent occupational, intentional and inadvertent exposures to RF-EMR may evoke cerebral dysfunction in predisposed individuals. Hence, this study demonstrates the potential health risk associated with prolonged and frequent use of mobile phones.

Keywords: RF-EMR; Antioxidant defence Status; cerebral dysfunction

Introduction

The development of mobile phone technology has become one of the fastest growing technologies today. Mobile phones use electromagnetic radiation (EMR) in the radiofrequency band at frequency (RF) ranging from 900 MHz to 2200 MHz [1, 2]. Radiofrequency electromagnetic radiation [RF-EMR] from mobile phone can be absorbed by different organs of the body depending on how they are placed [3]. Evidences have shown that prolonged exposure to EMR from mobile phone could have effect on human's health [4]. In fact, based on findings from studies that were centered on the possible link between mobile phone use and brain cancer, the World Health Organisation have declared that radiation from mobile phones may be a potential source of increased incidence of brain cancer [5]. Furthermore, it has also been shown that increased incidence of brain tumor and acoustic neurinoma is linked with prolonged exposure to mobile phone radiation [6].

Generally, the mechanism by which RF-EMR produce deleterious effects on cerebral function have been intrinsically related to the modulation of redox homeostasis in animals [7].

Oxidative stress results from an imbalance between antioxidants and overproduction of reactive oxygen species (ROS), that exceed cellular enzymic and non-enzymic antioxidant defense mechanisms [3, 8, 9]. Consequently, it has been suggested that antioxidants such as melatonin, caffeic acid phenyl ester, vitamin C and vitamin E may counteract oxidative stress caused by RF-EMR in animal tissues [3].

Although emerging studies describing the involvement of acute or chronic exposure to RF-EMR in oxidative

stress is growing astronomically [3, 9, 10, 11, 12, 13], such studies are still inconclusive. Besides, mode and manner of exposure to RF-EMR can vary considerably depending on individuals. In some cases, a sizeable number of the populace are occupationally exposed to non-ionizing radiation on regular basis at different exposure duration daily. Consequently, experimental models of non-ionizing radiation that can simulate real day to day human exposure can vary widely. Hence, this study sought to evaluate the influence of different exposure duration of mobile phone radiation on brain redox status and cellular integrity in rat.

Materials and Methods

Animals

Thirty-two (32) Male albino Wistar rats weighing 100-120 g were kept in plastic cages and were allowed free access to water and normal pellet diet and were maintained under controlled conditions of humidity, temperature and a diurnal environment. Animals were acclimatized to laboratory before starting the experiment and during the irradiation period. All animal procedures were carried out in accordance with standard practice of the use of experimental animals.

Electromagnetic Source

The Mobile phone which have a personal communications service code division multiple access (PCS CDMA) frequency band of 2G network, 900MHz was used as a source of RF radiation. The mobile phone used in the study was Itel 5600 (China).

Irradiation Procedure

The rats were divided into four groups (A, B, C and D) of eight rats each and were exposed to different time durations (0, 4, 8 and 12 hours) of 900 MHz RF from mobile phone respectively. In order to ensure that all rats received same frequency radiation, they were confined in a plastic cage (11 cm x 15 cm) with a metal lid that allowed free air flow. The mobile phone was placed centrally on the metal lid.

Preparation of Tissue Homogenates

Rat testes were removed, placed on ice and homogenized in cold 50 mM Tris–HCl of pH = 7.4. The homogenates were centrifuged at 4,000x g for 10 min to yield the low-speed supernatant (S1) fraction that was used for lipid peroxidation assay. For all other antioxidant assays, protein inS1 were precipitated in 10 volumes of cold 4 % trichloroacetic acid (TCA) solution to give supernatant S2 which was used for other antioxidant assays.

Estimation of Antioxidant defense systems

Glutathione (GSH): Homogenate was mixed with 0.2 M Tris buffer, pH 8.2 and 0.1 mL of 0.01 M Ellman's reagent, (5, 5'-dithiobis-(2-nitro-benzoic acid)) (DTNB). Each sample tube was centrifuged at 3000 g at room temperature for 15 min. The absorbance of the clear supernatants was measured using spectrophotometer at 412 nm in one-centimeter quarts cells Units

Glutathione Transferase (GST): The reaction mixture for the assay of GST consisted of 1.0 mM GSH, 1.0 mM CDNB, 0.1 M phosphate buffer (pH 7.4) and 0.1 mL of PMS in a total volume of 3.0 mL. The change in absorbance was recorded at 340 nm by using Shimadzu spectrophotometer UV-1601 and enzyme activity was calculated as nmol of CDNB conjugate formed min-1 mg-1 protein using molar extinction coefficient of 9.6×103/M/cm.

Superoxide Dismutase (SOD): The activity of SOD was estimated, with the aid of nitroblue tetrazolium as the indicator. Superoxide anions were generated by the oxidation of hydroxylamine hydrochloride. The reduction of nitroblue tetrazolium to blue formazon mediated by superoxide anions was measured at 560 nm under aerobic conditions. The SOD activity was expressed as units/mg protein as compared to a standard curve.

Glutathione Peroxidase Activity (GPx): GPx activity in homogenate was evaluated spectrophotometrically at 340 nm by the modified method of Tappel [14]. The reaction mixture for the assay of GPx included GSH (0.39mM), NADPH (0.19mM), glutathione reductase (1.55 U/ml) in assay buffer (50mM Tris, 0.1mM EDTA; pH 7.6) and sample (10m l). The enzyme reaction was initiated by cumen hydroperoxide (0.1%, v/v). Activity was expressed as nmol/min/g tissue.

Catalase: Aliquot of 0.5 mL post-mitochondrial supernatant was mixed with 2.5 mL of 50 mM phosphate buffer (pH 7.0) and 20 mM H2O2. Catalase activity was estimated spectrophotometrically following the decrease in absorbance at 240 nm. The specific activity of catalase was expressed in terms of units/mg protein as compared to a standard curve.

Histological Study

Slices of the brain tissue of each rat were carefully removed and fixed in 10 % formalin, dehydrated with ethanol, and embedded in paraffin. Tissue sections of 5 to 7 µm thickness were cut and stained with hematoxylin and eosin (H and E). The slides were examined under Nikon Eclipse C1 Photomicrography at the Central Research laboratory, Federal University of

Technology, Akure, Nigeria.

Data Analysis

Data were analyzed using SPSS program (statistical package for social sciences Inc. Chicago, Illinois). Means were compared by independent sample t-test. A difference was considered significant at probability $p \leq 0.05$.

Results and Discussion

A The largest most extensive and intensive human biologic experiment ever has been described in terms of the deliberate and voluntary exposure of the human brain to RF-EMR derived from mobile phone radiation. In fact, more than a quarter of the world's population expose their brain to microwaves through hand-held mobile phones [15]. Hence, the need to evaluate the possible risks associated with the human exposure to RF-EMR has been a major concern for more than five decades even before the advent of mobile phones, when radar and microwave ovens posed a possible health problem [16]. Because RF-EMR can induce redox imbalance in living systems, one plausible scientific approach has consistently been the need to determine the influence of these radiations on redox homeostasis in different life forms. Consequently, this study adopts similar approach in determining the influence mobile phone radiation on rat brain antioxidant and cellular integrity using of a model of mobile phone radiation exposure in rats.

Effect of RF-EMR on non-enzyme and enzyme antioxidant status

Figure 1 shows the results of different exposure durations to RF from mobile phone on the level of the antioxidant biomolecule, glutathione. It is evident that increased exposure to mobile phone radiation markedly decreased the level of GSH content in the brain of rats (with $p < 0.05$). In Figure 2, the results presented show that increased exposure to RF-EMR caused a decrease in the activity of superoxide dismutase. The activity of the enzymes is lowest in animals exposed to 12-hour mobile phone radiation. The influence of the RF-EMR on the activity of the glutathione-dependent antioxidant enzyme, glutathione peroxidase is shown in Figure 3. Similarly, it is obvious that prolonged exposure to RF-EMR posed a stress on the activity of the antioxidant enzyme. Figure 4 shows the effect of RF-EMR on the activity of catalase. As revealed in the result, the activity of the enzymes was inhibited, and the extent of inhibition is dependent on the duration of exposure to the RF-EMR. The activity of glutathione transferase is in animals exposed to RF-EMR is presented in Figure 5.

Generally speaking, in all the exposure duration (4, 8 and 12 hours) reported in this study, the activity of the detoxifying enzymes was inhibited, and the degree of inhibition is dependent on the exposure duration to RF-EMR from the mobile phone. Specifically, the increase in exposure duration to RF-EMR from mobile phone is accompanied with simultaneous decreases in the level of non-enzymic (glutathione) and the activities of enzymic (catalase, superoxide dismutase, glutathione peroxidase and glutathione transferase) antioxidant indices (Figures 1-5).

Antioxidants are known to continuously neutralise reactive oxygen species (ROS) in the body tissues. Thus, decrease in antioxidant capacity will lead to increase in ROS with a consequent increase in oxidative stress (OS) which arises from disequilibrium between the production of free radicals and the scavenging capacity driven by various antioxidant compounds and enzymes. All these antioxidant defense systems can be specifically diminished by the RF radiation, thus amplifying oxidative stress [13]. The result further corroborates earlier reports indicating that oxidative stress develops in response to mobile phone radiation [3, 17]. The result showed that, increasing exposure to RF radiation may increase the oxidative stress possibly via generation of free radicals which ultimately leads to induction of lipid peroxidation and this effect is associated with concomitant reduction in the activities of antioxidant enzymes such as SOD and GSH-Px which are equally regarded as free radical scavengers [18]. It is to be noted that *indicates a significant difference from control at $p < 0.05$.

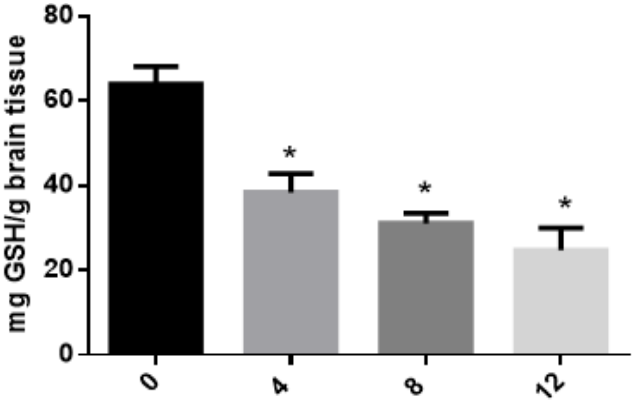

Figure 1. **Effect of RF radiation exposure on the level of cerebral GSH. Data are expressed as means ± SEM (n=8).**

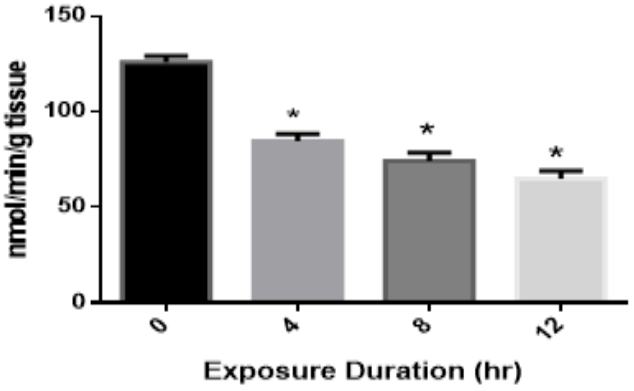

Figure2.**Effect of RF radiation exposure on the activity of cerebral superoxide dismutase. Data are expressed as means ± SD (n=8).**

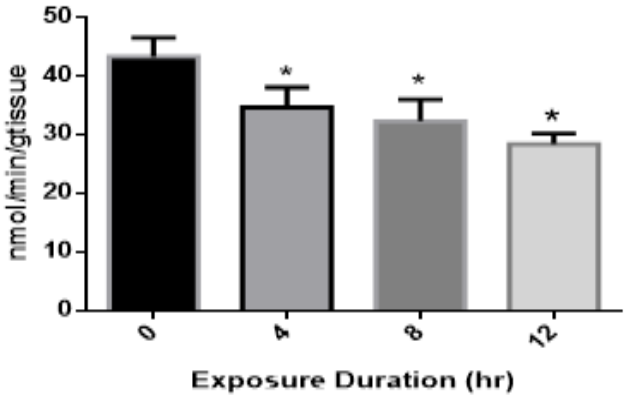

Figure3.**Effect of RF radiation exposure on the activity of glutathione peroxidase in rat brain. Data are expressed as means ± SD (n = 8).**

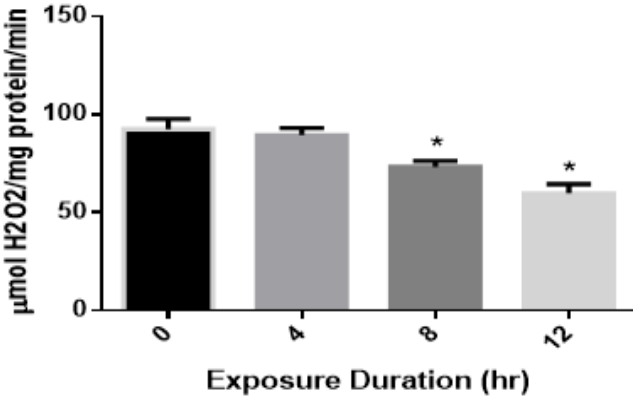

Figure4.**Effect of RF radiation exposure on catalase activity in the rat brain. Data are expressed as means ± SD (n = 8).**

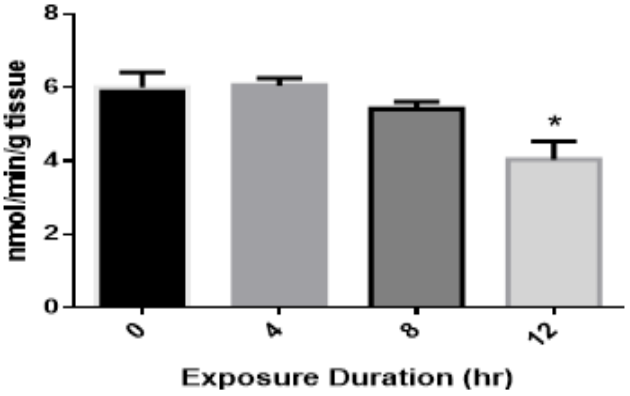

Figure5.**Effect of RF radiation exposure on catalase activity in the rat brain. Data are expressed as means ± SD (n = 8).**

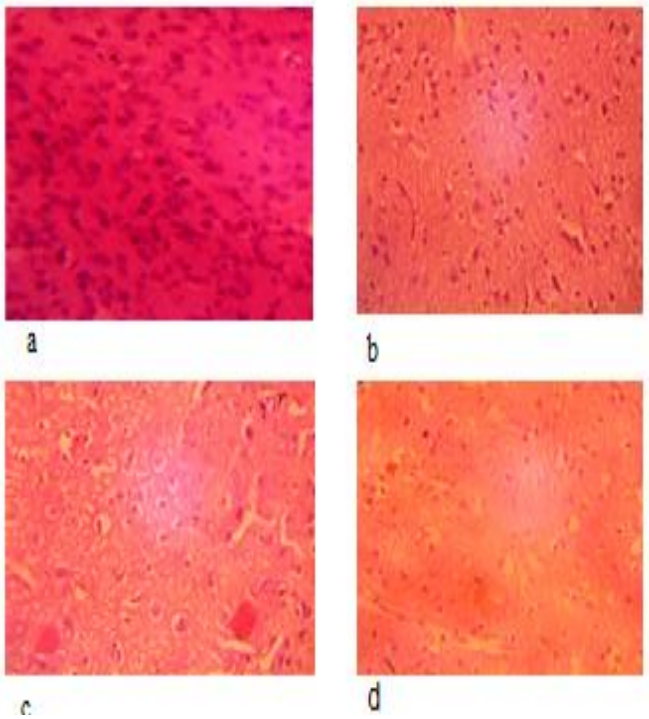

Plate1.**Histological Architecture of the brain after (a) 0 hours (control), (b) 4 hours, (c) 8 hours and (d) 12 hours of RF radiation exposure.**

Effect of RF Radiation Exposure on the Histological Architecture of the Brain Tissues

The cellular architectures of the brain tissues studied at varied durations are provided in Figures 6 (a - d). The histological evaluation of the control group (figure a) revealed a normal architecture of brain (neuronal) cells, there was no visible lesion seen in the tissues.

Figure (b) shows the effect of 4 hours exposure of albino rats to RF Radiation exposure from mobile phone on the neuronal cells, the Plate showed that the neuronal cells appeared normal with a mild gliosis and mild spongiosis of the parenchyma.

In the 8 hours exposure duration (Plate 1c), there was a disarray that defined a moderate to severe neuronal cell degeneration, some cells appeared inflamed.

The architectural observation on the neuronal cells of

the animals exposed to RF radiation from mobile phone for 12 hours (Plate 1 d) shows severe degeneration of the neuronal cells compared to the brain specimen of the control group. There were obviously inflamed cells seen and several cells had collapsed. This result agrees with previous reports indicating that 900 MHz RF radiation exposure to mobile phone produce observable histological changes in the brain tissue [2, 18]. Prolonged exposure to RF radiation from mobile phone can cause degenerative changes, shrunken cytoplasm and extensively dark pyknotic nuclei [19]. An epidemiological survey found that RF radiation caused human fatigue, headache, excitement, dreams, memory loss and other symptoms of neurasthenia [20]. RF radiation exposure may also lead to neuronal shrinkage, nuclear condensation, mitochondrial swelling, an expanded endoplasmic reticulum, alterations to the synaptic gaps and widened vascular endothelial connections, where mitochondrial injury occurred earlier and more severely [21, 22].

Conclusions

The results of this work show that frequent exposure to RF radiation diminished the antioxidant defense status and evoked distorted structural integrity of brain cells and this effect was exacerbated with increased duration of exposure. Within the limits of the present study, it appears that unwitting frequent occupational, intentional and inadvertent exposures to RF radiation may evoke cerebral dysfunction in predisposed individuals. Hence, this study serves as public awareness of the potential health risk associated with prolonged and frequent use of mobile phones.

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Abbreviations

RF-EMR: Radiofrequency electromagnetic radiation; GSH: Glutathione; GST: Glutathione Transferase; SOD: Superoxide Dismutase; GPx: Glutathione Peroxidase Activity.

Author Contributions

The author gave final approval for the publication of this study.

Competing Interests

The author has declared that no competing interest exists.

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