The Effects of WiFi Network (2.45 GHz) on Rats with Induced Stroke Associated with an Increased Risk of Heart Attack

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Abstract

Introduction: Stroke and heart attack are the most common causes of death among humans. Troponin I, Creatine Kinase-MB (CK-MB) and Lactate Dehydrogenase (LDH) are the diagnostic markers of heart attack which can also be used as high risk biomarkers. WiFi is a cheap common technology which exposes its users to a spectrum of electromagnetic waves. Can weak electromagnetic waves affect human health?

Materials and Methods: In this study, stroke in rats has been induced, and then they were exposed to WiFi waves (2.45 GHz) and finally were examined for the risk of heart attack through analyzing three enzyme biomarkers related to heart attack (Troponin I, CK-MB and LDH).

Results: This study’s results confirm WiFi’s biological effects and shows WiFi’s contribution in stroke. WiFi exposure affects three cardiac enzyme markers of heart attack (LDH, Troponin I and CK-MB), considering the current data on WiFi exposure effects on the brain, heart and related enzymes.

Conclusion: Some of the WiFi wave’s cellular targets include cell membrane, cellular proteins and enzymes. Despite all the data and reports on biological effects of electromagnetic fields, the range and rate of these effects has not yet been determined.

Keywords: Stroke, Heart attack, Troponin I, CK-MB, LDH, WiFi

1. Introduction

Stroke is the basis for a series of serious clinical and biological crisis in the body. In most cases, the stroke happens following arterial blockage and the consequent temporary blood supply interruption. Blood supply cut-off results in cerebral infarction [1]. Besides stroke, cardiac damage (heart attack) is another important cause of death worldwide. A strong body of evidence exists suggesting that the occurrence of these two phenomena is related to each other. Biomarkers are considered a powerful tool in the high risk detection and prognosis of these fatal diseases.

Biomarkers can be used for rapid and precise detection of the disease, resulting in an efficient treatment plan [2,3]. Troponin I, Creatine Kinase-MB (CK-MB) and Lactate Dehydrogenase (LDH) are some of the biomarkers used for heart attack diagnosis and high risk detection [4-6]. Troponin I, responsible for regulating the intermediate interaction of calcium with actin and myosin fibers, is also a biochemical marker of cardiac damage. This calcium-binding protein is the unit of muscle contraction. Troponin I is not only used for the detection of myocardial necrosis, but can also be utilized for risk
classification and the staging of the disease [7,8]. Elevation of Troponin I levels occurs in two stages. The initial elevation of cardiac Troponin I results in cytosolic Troponin I release, followed by the release of Troponin I from repairing cardiac myofibrils [9,10].

Studies have shown that Troponin I and CK-MB Iso-enzyme levels elevate during a heart attack. Elevation in CK-MB levels following cardiac damage specifically occurs in 3-8 hours and reaches its maximum levels in 9-26 hours and finally, at 48-72 hours returns to its initial stage. CK-MB is considered as a specific cardiac biomarker since its levels are not significant in non-cardiac tissues. It is worth mentioning that CK-MB levels may elevate in other injuries rather than heart attack [11-13].

LDH is also elevated during cardiac injury and returns to its initial level in 3-6 days. LDH is produced by all cell types in the body which makes it a less important biomarker compared to Troponin I and CK-MB in the diagnosis of heart attack or as a risk factor of the disease [14,15].

The Short-range WiFi network operates in short wavelength ranges of the electromagnetic spectrum [16]. There are different sources of electromagnetic waves in our environment, including wireless communications, radars, satellites, and TV and radio antennas. WiFi is a cheap common technology which exposes its users to a spectrum of waves. Modems are among the devices that generate 2.45 Gigahertz (GHz) microwave waves for wireless networks. Without any doubt, close and continuous exposure to weak electromagnetic fields will have undeniable effects on human body [17-19].

Various studies have been conducted to examine the effects of WiFi on human health, while the risk level of high risk patients has remained unclear. This study investigated the effects of WiFi network (2.45 GHz) on the occurrence of heart attack in rats, suffering from stroke. For this purpose, rats with stroke were exposed to WiFi signals and alterations in the levels of three main markers of heart attack (LDH, CK-MB and Troponin I) were analyzed.

2. Materials and Methods
2.1 Experimental Groups

Four groups of 10 male Wistar rats [with similar conditions: gender (male) and age (three months old)] with body weight ranging from 200 to 250 g were chosen randomly and without any tension, from the Laboratory Animal Center of Tehran University. The animals had free access to food and water, maintained on a 12 h light/12 h dark cycle in a temperature-controlled environment (22°C). The studies were performed in accordance with guidelines established by the university research center (Department of Biology, School of Basic Sciences, Science and Research Branch, Islamic Azad University, Tehran, Iran) [17] and all animals received human care according to the criteria outlined in the “Guide for the Care and Use of Laboratory Animals” prepared by the National Academy of Sciences and published by the National Institute of Health (NIH publication 86-23 revised 1985) [16]. Usage of animals was in accordance with the ethical committee of the Science and Research Branch, Islamic Azad University, Tehran, Iran. Ethics approval was gained for this research.

Experimental group 1 (C): the control group, consisted of rats exposed to no electromagnetic field.

Experimental group 2 (T+WiFi2.45): consisted of healthy rats exposed for three consecutive days (2 hours daily) to WiFi waves (2.45 GHz).

Experimental group 3 (S): consisted of rats with induced stroke with no exposure to WiFi waves (same as the control group).

Experimental group 4 (S+WiFi2.45): consisted of rats with induced stroke exposed for three consecutive days (2 hours daily) to WiFi waves (2.45 GHz).
2.2 Induction of Stroke in Rats

Stroke induction by occlusion of the middle cerebral artery was performed by nylon string (Ehicon, Germany). In this method, middle cerebral artery was blocked temporarily (for a short time) and the blood flow was dropped to less than 20%. After 90 minutes (the certain amount of time needed for induction of brain injury), normal blood flow was provided. At the time of stroke induction, rat’s body temperature was maintained at 37°C. Neurological movement disorders were evaluated to confirm stroke induction.

2.3 WiFi2.45 Exposure System

A room (2m×2m×2m) was prepared and WiFi exposure was provided through 4 antennas (NanoStation Loco M2, 2.45 GHz, 8.5 dBi, Ubiquiti Networks, Inc. USA), placed on each side of the room. Rat’s Plexiglas cages (40cm×40cm×20cm) were placed in the middle of the room while the animals had movement freedom. The safe level of Specific Absorption Rate (SAR) and surface power density for two experimental groups (T+WiFi2.45 and S+WiFi2.45) were calculated according to regulations published by the Atomic Energy Organization of Iran and the International Commission of Non-Ionizing Radiation Protection (ICNIRP). The following equation was used for SAR calculations

\[
SAR = \sigma \cdot E^2 / \rho
\]

in which \( E \) is the electric field (v/m), \( \sigma \) is conductivity (s/m) and \( \rho \) is tissue’s mass density (Kg/m3). This equation shows that SAR value can be determined by knowing the magnitude of the electric field and that SAR has a direct relationship with electric conductivity. Surface power density and electric field magnitude was determined using a field meter (NARDA, N-550, USA). Our evaluations showed that during WiFi exposure the values of surface power density and SAR in rat’s brain was 0.8 mW/cm2 and 5.174406171 W/Kg, respectively.

2.4 Experimental Procedure

In this study, blood samples were collected after three days from all animals in four experimental groups. Blood samples (1 CC) were taken from the lateral tail vein of the rats following general anesthesia. All samples were collected in the morning after the third 2-hour exposure to WiFi waves. Blood samples were then centrifuged (in 4°C, 3000 g for 5 minutes) and the serum was collected.

In the present study, LDH level was determined through biochemical methods (DGKC opt, 1970, 37°c), using an auto-analyzer. CK-MB levels were determined immunologically (Mak, immunological 37°c), using an EISA device; Troponin I was assessed by Chemiluminescence (LIASOIN® DianSorin Italy) with an auto-analyzer.

2.5 Statistical Analysis

The t-test and one-way ANOVA was performed for statistical analysis of data with SPSS software (version 19) and the P value<0.05 was considered significant.

3. Results

In this study, levels of three enzymes were evaluated in four test groups. The results showed that T+WiFi2.45, S and S+WiFi2.45 groups had elevated levels of CK-MB compared to control group [57% and 15% respectively, (2.35 fold increase)]. The S+WiFi2.45 group showed 135% and 99% increase compared to control and S group, respectively (Figure 1 and table 1).

Troponin I studies showed no significant difference between T+WiFi2.45 and S groups with a control group (36% and 21% respectively). However, S+WiFi2.45 group showed a 6.56 fold increase compared to control group and a 5.4 fold increase compared to S group which is 556% and 440% increase compared to the control and S group, respectively (figure 2 and table 1). Studies on LDH levels showed an 8% and 13% increase in T+WiFi2.45 and S groups compared to control group, respectively.
This increase was much higher for the S+WiFi2.45 group compared to the control group (2.2 fold) which was also 1.95 fold higher than S group. This elevation was 20% and 90% higher than control and S group, respectively (figure 3 and table 1).

### Table 1. Statistical analysis (t-test and one-way ANOVA; $P < 0.05$) of all three enzymes for Control (C), Test + WiFi$_{2.45}$ (T+WiFi), Stroke (S) and Stroke + WiFi$_{2.45}$ (S+WiFi).

<table>
<thead>
<tr>
<th>Test</th>
<th>Mean</th>
<th>Median</th>
<th>Error Of Mean (SD)</th>
<th>Variance</th>
<th>Min</th>
<th>Max</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>CK-MB U/L</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>992</td>
<td>999.5</td>
<td>18.33</td>
<td>336</td>
<td>951</td>
<td>1017</td>
<td>66</td>
</tr>
<tr>
<td>T+WiFi</td>
<td>1557</td>
<td>1260</td>
<td>33.22</td>
<td>1104</td>
<td>1198</td>
<td>1289</td>
<td>91</td>
</tr>
<tr>
<td>S</td>
<td>1171</td>
<td>1188.5</td>
<td>59.10</td>
<td>3488</td>
<td>1080</td>
<td>1253</td>
<td>173</td>
</tr>
<tr>
<td>S+WiFi</td>
<td>2330</td>
<td>2362</td>
<td>93.88</td>
<td>8813</td>
<td>2154</td>
<td>2501</td>
<td>347</td>
</tr>
<tr>
<td><strong>Troponin I Ng/L</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>0.248</td>
<td>0.27</td>
<td>0.790</td>
<td>0.006</td>
<td>0.09</td>
<td>0.38</td>
<td>0.29</td>
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<tr>
<td>T+WiFi</td>
<td>0.337</td>
<td>0.33</td>
<td>0.122</td>
<td>0.015</td>
<td>0.15</td>
<td>0.62</td>
<td>0.11</td>
</tr>
<tr>
<td>S</td>
<td>0.301</td>
<td>0.30</td>
<td>0.038</td>
<td>0.002</td>
<td>0.21</td>
<td>0.35</td>
<td>0.14</td>
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<tr>
<td>S+WiFi</td>
<td>1.627</td>
<td>1.61</td>
<td>0.091</td>
<td>0.008</td>
<td>1.5</td>
<td>1.79</td>
<td>0.29</td>
</tr>
<tr>
<td><strong>LDH U/L</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>1318</td>
<td>1303.5</td>
<td>48</td>
<td>2339</td>
<td>1241</td>
<td>1422</td>
<td>181</td>
</tr>
<tr>
<td>T+WiFi</td>
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<td>1476.5</td>
<td>64</td>
<td>4069</td>
<td>1309</td>
<td>1512</td>
<td>203</td>
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<tr>
<td>S</td>
<td>1486</td>
<td>1500</td>
<td>78</td>
<td>6147</td>
<td>1312</td>
<td>1602</td>
<td>290</td>
</tr>
<tr>
<td>S+WiFi</td>
<td>2894</td>
<td>2955.5</td>
<td>105</td>
<td>11203</td>
<td>2722</td>
<td>3012</td>
<td>290</td>
</tr>
</tbody>
</table>

**Figure 1.** CK-MB changes for Control (C), Test + WiFi$_{2.45}$ (T+WiFi), Stroke (S), and Stroke + WiFi$_{2.45}$ (S+WiFi) groups
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Figure 2. Troponin I levels for Control (C), Test + WiFi2.45 (T+WiFi), Stroke (S), and Stroke + WiFi2.45 (S+WiFi) groups

Figure 3. LDH levels for Control (C), Test + WiFi2.45 (T+WiFi), Stroke (S), and Stroke + WiFi2.45 (S+WiFi) groups

Statistical analysis was performed and the results are presented in table 2. No significant difference was observed in the level of Troponin I between T+WiFi2.45 and control group; however, LDH and CK-MB levels showed significant increase. In S group, only CK-MB elevation was significant compared to control group and in S+WiFi2.45 group, all three enzymes showed significant elevation compared to control and S groups.

Based on the obtained results, it can be concluded that among all three studied enzymes, CK-MB is more sensitive to WiFi2.45 waves in both healthy rats and the ones suffering from a stroke. Troponin I serum levels was only elevated by WiFi2.45 waves in rats suffered from stroke which indicates that exposure of rats...
suffered from stroke to WiFi2.45 waves can increase their risk of heart attack. Figure 4 represents detailed data about the changes observed in the levels of three enzymes following exposure to WiFi2.45.

Table 2. Statistical analysis to determine the significance level of the results (the \( P < 0.05 \) was considered significant)

<table>
<thead>
<tr>
<th></th>
<th>LDH</th>
<th>CK-MB</th>
<th>Troponin I</th>
</tr>
</thead>
<tbody>
<tr>
<td>C and T+Wi-Fi2.45</td>
<td>*1.32016E-05</td>
<td>*1.03866E-10</td>
<td>0.000433473</td>
</tr>
<tr>
<td>C and S</td>
<td>0.000452971</td>
<td>*1.24036E-06</td>
<td>0.019698358</td>
</tr>
<tr>
<td>C and S+Wi-Fi2.45</td>
<td>*1.811188E-11</td>
<td>*3.71738E-12</td>
<td>*6.55779E-12</td>
</tr>
<tr>
<td>S and S+Wi-Fi2.45</td>
<td>*2.06509E-14</td>
<td>*88.61486E-14</td>
<td>*1.07716E-13</td>
</tr>
</tbody>
</table>

* significant

Figure 4. Diagram scatter, trandline drawings for CK-MB, Troponin I, and LDH based on data from table 1. Comparison graphs for each of three enzymes (CK-MB, Troponin I, and LDH). Four different modes are shown in the following diagram (Control, Test + WiFi2.45, Stroke, and Stroke + WiFi2.45).

4. Discussion

Today, wireless networks are present in our homes, at work and in public places and has become an inseparable part of our lives. There have been reports on effects of WiFi waves on cell biology and human health [18,19]. Most of these reports are related to studying the negative effects of electromagnetic fields on human health, including their effects on the endocrine system. In this respect, the authors of the present study had previously studied the possible effects of WiFi waves on sex hormones and observed significant adverse effects [20,21]. Saili and et al in 2015 showed that WiFi2.45 GHz increased heart frequency (+22%) and arterial blood pressure (+14%). Their results showed that that exposure to WiFi2.45 GHz affects heart rhythm, blood pressure, and catecholamines efficacy on the cardiovascular system,
indicating that radiofrequency can act directly or indirectly on the cardiovascular system [22]. The present study manifests that the Troponin I enzyme increases after being exposed to WiFi2.45 GHz, and will increase the likelihood of a heart attack for high-risk patients.

Some of the WiFi wave’s cellular targets include cell membrane, cellular proteins and enzymes. Although the WiFi role in cancer has not yet been confirmed, these changes could be the beginning of the transformation process. It has been reported that WiFi could increase free radicals, which have an important role in cancer development [23,24]. Recently, Celiko et al conducted a research on the induction of oxidative stress in the brain and liver under WiFi2.45 GHz. Their results showed significant adverse effects.24 There are also reports on lipid peroxidation under WiFi2.45 GHz, which plays an important role in cellular protection mechanisms. WiFi exposure does not always result in enzyme level elevation. In the case of Glutathione-s transferases (GSTs) which are involved in detoxification and metabolism, WiFi exposure lowers GSTs levels significantly while unexpectedly, preventing its elevation in the case of poisoning [25-29].

5. Conclusion
Considering current data on WiFi exposure effects on the brain, heart and related enzymes separately, the present study was conducted to assess all these three topics simultaneously. The results confirm WiFi’s biological effects and the results showed WiFi contribution in stroke. However, the rate and quality of these effects varied. Based on the obtained results, WiFi2.45 GHz exposure affects three cardiac enzyme markers of heart attack including LDH, Troponin I and CKMB. In the case of Troponin I, this effect was significantly additive. Since LDH is produced by all metabolically active cells, it is not the best marker for heart attack. However, the present study did not show any significant changes in LDH levels upon WiFi exposure compared to Troponin I and CKMB in healthy rats. Considering that CKMB iso-enzyme level is relatively low in tissues other than cardiac tissue (only 3% of prostate, diaphragm and skeletal muscles), it is a suitable marker for high risk of heart attack.

Despite all the data and reports on biological effects of electromagnetic fields, the range and rate of these effects has not yet been determined. More research is required in this area to achieve the mentioned objective. Consequently, it is suggested that patients and people at high risk of heart and brain disease live in safe conditions and away from electromagnetic waves.

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The authors declare that they have no conflict of interests, and usage of animals was in accordance with the ethical committee of the Science and Research Branch, Islamic Azad University, Tehran, Iran. Ethics approval was made for this research

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