Original

Effect of Non-Ionizing Radiations on Liver and Kidney Function Tests in an Animal Model

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ABSTRACT

Background: Nowadays, the growing use of non-ionizing electromagnetic radiation has raised concerns about its potential health effects.

Objective: In this work, an animal model exposed to Wi-Fi and jammer signals was used to examine the effects of non-ionizing electromagnetic radiation on kidney and liver function.

Material and Methods: In this experimental study, twenty-one male Wistar Albino rats were separated into three groups: Wi-Fi, jammer, and sham groups. The animals were exposed to electromagnetic radiation for two hours per day for two weeks. Blood samples and kidney and liver tissues were collected and analyzed for various biochemical parameters.

Results: The findings of this study showed a mild inflammatory response in both tissues after exposure to the fields. However, no notable or serious alterations were noted in the groups under study. The Wi-Fi and jammer signals had no significant impact on creatinine, albumin, blood urea nitrogen, cholesterol, high-density lipoprotein, triglycerides, albumin/globulin ratio, total bilirubin, direct bilirubin, and alkaline phosphatase levels. However, the jammer group revealed a notable decline in low-density lipoprotein compared to the sham group. Significant differences were observed in the levels of aspartate aminotransferase and alanine aminotransferase between the Wi-Fi and sham groups but not between the other groups.

Conclusion: This work emphasizes the importance of considering individual organ characteristics in response to electromagnetic radiation exposure. Prolonged or closer exposure to the radiation source may significantly affect the organ function.

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Keywords

Radiation; Non-ionizing; Kidney Function Tests; Liver Function Tests; Wi-Fi Signals; Jammer Signals

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Introduction

Wireless Local Area Network (WLAN) systems operating at a frequency of 2450 MHz are now widely used in educational, commercial, and residential settings for broadband multimedia communications. However, extensive use of WLAN and mobile phones has highlighted important concerns regarding the potential health risks associated with exposure to Electromagnetic Radiation (EMR) [1]. The use of jammer routers, which emit noise with an identical frequency signal to mobile phones, can disable cell phones in the area where the jammer is placed [2]. EMR is classified into two categories: ionizing and non-ionizing radiation. While ionizing radiation is known to cause significant damage to biological tissues, non-ionizing radiation is generally considered less harmful due to its lower energy. However, it retains the ability to change the rotational, vibrational, or electronic configurations of both molecules and atoms, making it a powerful tool in molecular manipulation [3]. However, epidemiological studies and laboratory investigations have reported various potential health risks associated with EMR exposure, including carcinogenic, reproductive, neurological, and genotoxic effects, among others [3]. The International Commission on Non-Ionizing Radiation Protection [4] and the European Commission [4] have issued suggestions to reduce the potential health risks associated with EMR exposure.

The health effects of non-ionizing EMR contribute to various benefits, including cancer treatment that combines chemotherapy with radiation, as well as its adverse effects. Adebayo et al. conducted an experiment, in which male rats were exposed to electromagnetic radiation, resulting in histopathological changes in their liver and kidneys [5]. Similarly, Li et al. found that exposure to Wi-Fi and mobile phone radiation induced oxidative stress and inflammation in rats, indicating potential harm to liver and kidney function [6]. Fahmy et al. reported negative impacts on liver and kidney function in rats due to chronic exposure to 1800-MHz radiofrequency radiation, as evidenced by biochemical parameter alterations [7]. In another study, Özorak et al. [8] exposed the rats to 2.45 GHz EMR for an hour each day over three months, resulting in increased levels of liver enzymes, suggesting potential liver damage. Finally, Ragy et al. [9] found that exposing rats to 900 MHz EMR for two hours per day for two weeks increased oxidative stress and decreased kidney function.

Nevertheless, studies have produced conflicting experimental evidence leading to uncertainties regarding the potential health effects of exposure to EMR. For instance, Jeong et al. [10] found that long-term exposure to RF-EMF did not affect age-related oxidative stress or neuroinflammation in C57BL/6 mice. The oxidative stress level, DNA damage, apoptosis, astrocyte, or microglia markers in the brain, moreover locomotor activity, did not alter by long-term RF-EMF exposure in aged mice [10]. Sundaram et al. [11] showed That 150 kHz intermediate frequency electromagnetic radiation exposure for two months leads to changes in the liver and lungs, but it is not sufficient to cause clinical or functional manifestations. In another study, Esmaili et al. explained short short-time mobile phone radiation did not affect anxiety-like behaviors and serum enzyme activity in male rats [12]. Likewise, Owjfard et al. [13] exposed rats to 900 MHz EMR for one hour per day for four weeks and observed no significant effects on liver and kidney function. The objective of this study was to examine the impact of Wi-Fi and jammer signals on liver and kidney function in an animal model, using biochemical parameters as indicators of organ damage. In this work, we used an animal model to conduct an investigation of the effects of non-ionizing EMR on hepatic and renal functions. Rats were exposed to EMR emitted by cell phones, WLAN, and jammer routers for 14 days, and their hepatic and renal functions were evaluated through assessments analyzing various biochemical parameters. The findings of this study can help contribute to our understanding of the potential health effects of EMR exposure on liver and kidney function, and the findings may have implications for human health.

Material and Methods

This research study is an experimental type of intervention conducted on rats.

Subjects and study design

Twenty-one male Wistar Albino rats, three months old with a mean weight of 225±25 g, were acquired from the animal center of the Research Institute of Shiraz University of Medical Sciences (Shiraz, Iran). The rats were housed in standard cages and maintained under controlled conditions of 25±3 °C and relative humidity of 24±6% with a 12-hour light/dark cycle. The animals were given standard chow for the duration of the study.

The rats were randomly divided into three groups, each consisting of seven, as follows:

Wi-Fi group: Every rat was placed into a separate cage. The shelves were located on the circumference of the circle with a radius of one hundred centimeters from the center of the circle. The Wi-Fi modem was in the center of the circle. Wi-Fi routers typically use 2.4 GHz EMF, commonly for two hours per day for fourteen days. The diagram below helps to clarify the content (Figure 1).

The Wi-Fi device used in this study was a Wi-Fi D-Link router (DLinkDIR-600L, D-Link Corporation, Taiwan). The Wi-Fi router functioned efficiently at a powerful level of 1 W, ensuring reliable connectivity.

Jammer group: All animals had exactly the same pattern as the "Wi-Fi-100 cm group", except that the rats were within a 100 cm radius of the jammer router. The diagram below displays the content (Figure 1).

A commercially available mobile jammer (MB06- Mobile Blocker) was used that emitted radiation at four different frequencies (Global System for Mobile Communications [GSM], Digital Cellular Service, Code Division Multiple Access, Third Generation); all frequencies were used in this study. The effective jamming distance for this signal jammer has been announced to be 10-40 m as presented by Mortazavi et al., [14].

Sham group: Experimental conditions like those of the treated groups were applied to the animals, except that the device was turned off during the treatment period. The following diagram shows the contents (Figure 1).

It should be noted that the Animal Experimentation Ethics Committee of Shiraz University of Medical Sciences approved all animal procedures. All experiments received approval from the local Institutional Review Board, following the Helsinki recommendations and adhering to the National Institutes of Health guidelines for animal treatment.

When the jammer device is turned on, it disables the wireless signal transmission. Therefore, all communication through the signal jammer is blocked. The jamming range from the jammer router is up to 40 meters.

Electromagnetic field measurements were conducted using an Aaronia Spectran HF-4060 device (Euscheid, Germany), which operates within a frequency range of 100 MHz to 6 GHz (Table 1).

Preparing blood samples and tissue slides

Rats were euthanized in accordance with international guidelines for animal care and ethical standards. Kidney and liver tissues, as well as blood samples, were collected. The right kidney and liver were then extracted, fixed

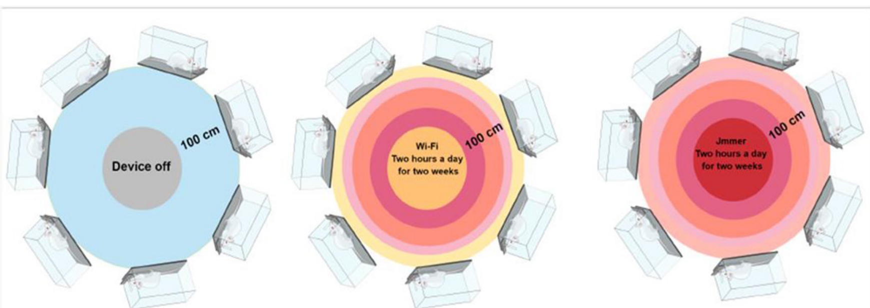

Figure 1: This diagram displays the matter.

Table 1: The frequency band, power output, and power density of Wi-Fi and Jammer electromagnetic fields were measured at about 100 cm from the animals' heads.

Radiation source	Frequency band (MHz)	Power output (dB mW)	Power density (μw/m²)	*ICNIRP (%)
Wi-Fi	2422-2461	-81±3	00.00215	0.00
Jammer	1079-1763	-36±3	24.79000	0.22

*Determining the limits of exposure to non-ionizing radiation, especially in the frequency range of radio and microwave radiation. The standard has also been endorsed by competent authorities such as the Atomic Energy Organization of Iran and the Ministry of Health; this standard aligns completely with the recommendations of the International Commission on Non-Ionizing Radiation Protection (ICNIRP).

in 10% formalin, and embedded in paraffin. Hematoxylin and eosin (H&E) staining was performed on the samples, and the histopathological changes in the renal and liver tissues were assessed using the method described by Bancroft et al., [15].

Statistical analysis

Results are presented as the mean ± standard error of the mean (SEM). Statistical significance was assessed using the Kruskal-Wallis test and one-way ANOVA, followed by Tukey's post hoc test. The experimental data were analyzed using SPSS statistical software (version 16.0; SPSS Inc., Chicago, IL, USA). A significance level of $P<0.05$ was used.

Results

Effect of the Wi-Fi & Jammer radiation on blood parameters associated with kidney and liver functions

The study investigated the effects of Wi-Fi and Jammer radiation on blood parameters related to kidney and liver functions. The results showed a significant difference in total protein between the sham group and Wi-Fi at about 100 cm (*P*-value=0.001), while no significant differences were observed in other groups (Figure 2a). There were no significant differences in creatinine (*P*-value=0.374) (Figure 2b), albumin (*P*-value=0.169) (Figure 2c), blood urine nitrogen (Figure 2d), cholesterol (*P*-value=0.194) (Figure 2e), and HDL (*P*-value=0.837) (Figure 2f) between the groups. However, the Jammer group showed a significant decrease in LDL compared to the sham group (*P*-value=0.032), while there was no significant difference in the other groups (Figure 3a). The average triglyceride level did not show any significant differences between the groups studied (*P*-value=0.850) (Figure 3b).

Our findings revealed no significant differences in glucose levels between the investigated groups (*P*-value=0.876) (Figure 3c). However, the globulin level of the rats in the Wi-Fi group was significantly decreased compared to the sham group (*P*-value=0.001) (Figure 3d), while the albumin/globulin ratio (Figure 3e), total bilirubin (Figure 3a), direct bilirubin (Figure 4b), and Alkaline phosphatase (ALP) levels (Figure 4c) showed no significant difference between the studied groups. Alanine aminotransferase (ALT) levels showed a significant difference between the sham and Wi-Fi groups at about 100 cm (Figure 4d), and Aspartate aminotransferase levels were significantly different between the sham group and Wi-Fi groups (*P*-value=0.015) (Figure 4e), but not in the other groups.

Histopathological Evaluation of Renal and Liver Tissues

Histopathological evaluation of kidney and liver tissues before and after irradiation was conducted (Figure 5(a-f)). The results showed that there was mild portal inflammation in the

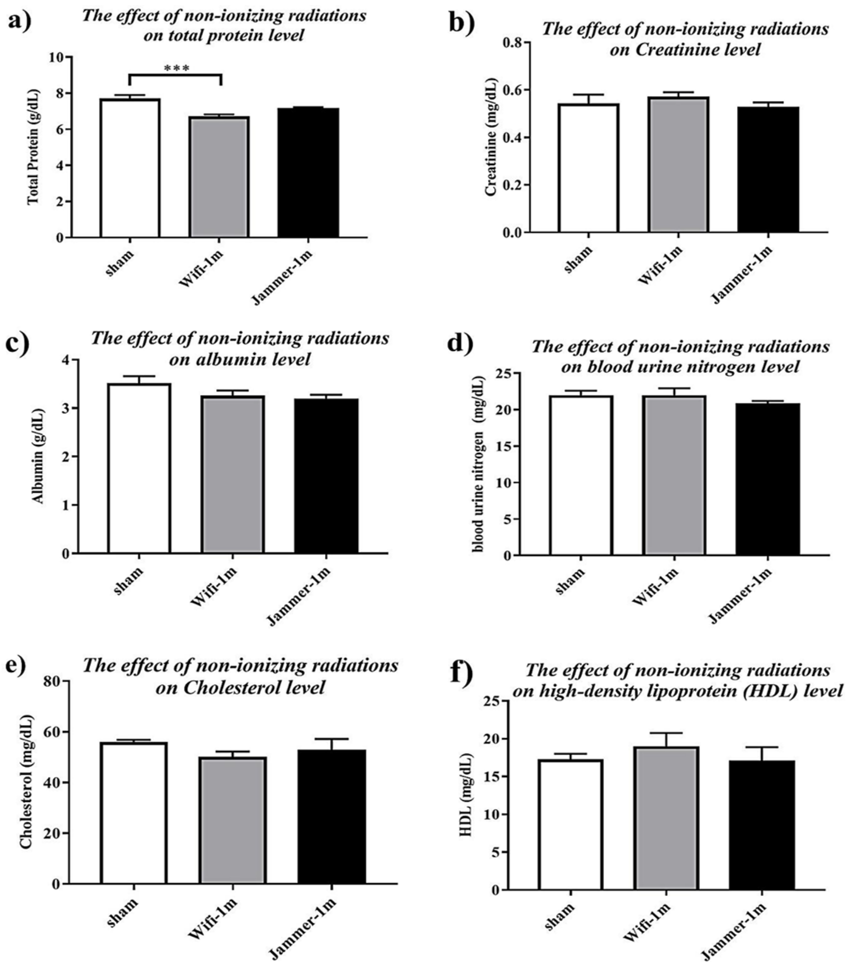

Figure 2: The impact of Wi-Fi and Jammer radiation on blood parameters related to kidney and liver functions. (**a**) A significant difference in total protein levels was observed between the sham group and the Wi-Fi group at a distance of 100 cm. (**b-f**) No significant differences were found in creatinine, albumin, blood urea nitrogen, cholesterol, and HDL levels between the groups. Note: HDL stands for high-density lipoprotein. Statistical significance is indicated as ***$P<0.001$.

liver and peritubular inflammation in the kidney for the Wi-Fi group compared to the Sham group. On the other hand, the Jammer group exhibited chronic perivascular inflammation in the kidney tissue and portal inflammation with piecemeal necrosis in the liver tissue, which is higher than the Sham group. The study highlights mild portal and peritubular inflammation in the liver and kidney, respectively, due to Wi-Fi and Jammer radiation exposure.

Discussion

The effects of non-ionizing electromagnetic radiation on human health remain a highly debated topic despite an increasing amount of literature on the subject [16-18]. The goal of this study was to fill this gap by examining the effects of radiation emitted from Wi-Fi and Jammer devices on the kidney and liver organs, which are critical for regulating metabolic activities. Given the limitations of human investigations, the use of animals under ethical protocols is an essential tool in understanding the pathophysiology of diseases, as it allows for faster research and lower costs [19-21]. Therefore, we conducted an animal model study based on the aforementioned method.

Our results regarding the normal range of total protein in rats are in the same line with previous research, which reports it to be between 5.6-7.6 g/dl [22]. Notably, we observed a significant difference in total protein and globulin levels between the sham group and the Wi-Fi groups (*P*<0.05), but not between the sham and jammer groups (Figures 2a and 3d). However, there was no significant difference in albumin protein or the albumin/globulin ratio between the sham group and the Wi-Fi or jammer groups (Figures 2c and 3e). Our study found that the concentrations of globulin and albumin proteins in rats following radiation exposure were within the normal range reported by Borzoueisileh et al., [23]. These protein concentrations may provide essential information for both prognosis and diagnosis [22].

Previous studies have shown a significant link between inflammation and the serum albumin-to-globulin (AG) ratio in chronic kidney disease [24]. Our findings indicate

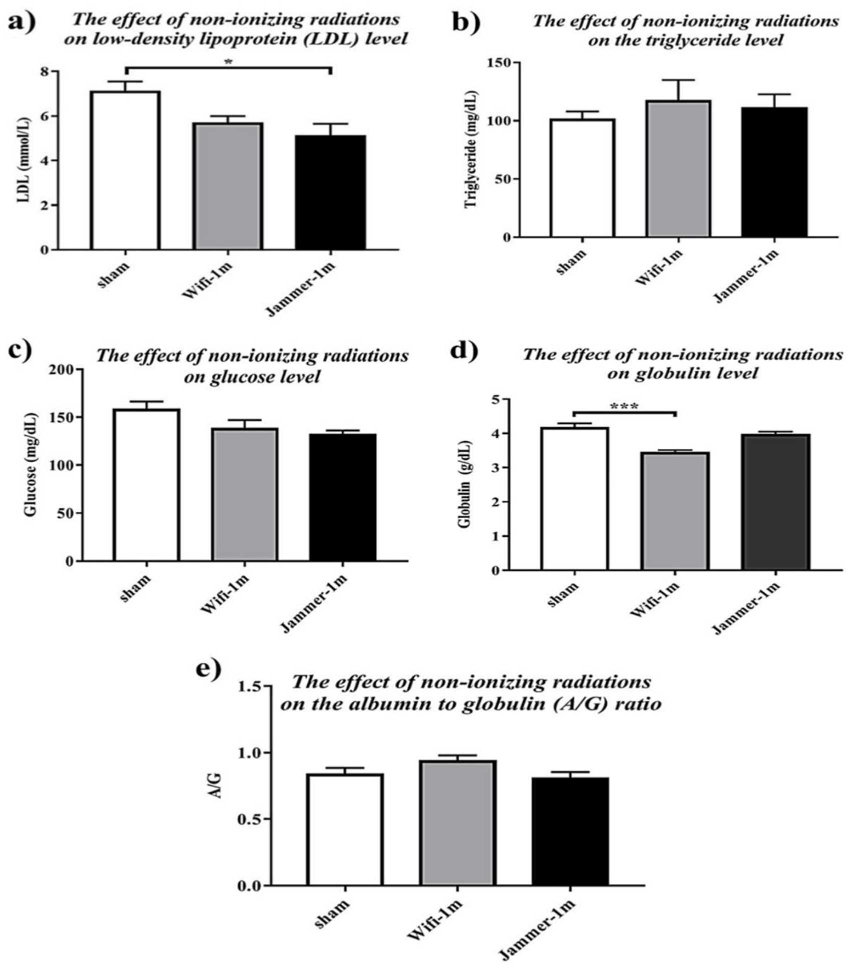

Figure 3: The impact of Wi-Fi and Jammer radiation on blood parameters related to kidney and liver functions. (**a**) The Jammer group exhibited a significant decrease in low-density lipoprotein (LDL) levels compared to the sham group. (**b**) There was no significant difference in mean triglyceride levels between the studied groups. (**c**) The glucose levels did not show any significant variation between the groups. (**d**) The globulin level in the Wi-Fi group was significantly lower compared to the sham group. (**e**) The albumin/globulin ratio showed no significant difference among the studied groups. Statistical significance is indicated as **P*<0.05 and ***P*<0.005.

that exposing animals to Wi-Fi and jammer radiation for two hours a day over two weeks did not lead to any significant changes in kidney function (such as removing waste products from the blood and urineas well as blood pressure control). This outcome is consistent with previous studies reporting normal Blood Urea Nitrogen (BUN) levels in rats (8.5–22.7 mg/dl) [11] (Figure 2d). Our results are consistent with those of Borzoi and colleagues [23], Stender and colleagues [25], and Fahmy and colleagues [26], but in contrast to those of Aweda and colleagues [27].

In our previous study [28], exposure to radiation from a jammer led to a substantial decrease in plasma glucose levels. Although we observed a decrease in blood sugar concentration, our findings did not reach statistical significance (Figure 3c). Notably, our study found that exposure to Wi-Fi and jammer radiation did not affect BUN levels, which are commonly used as an indicator of kidney

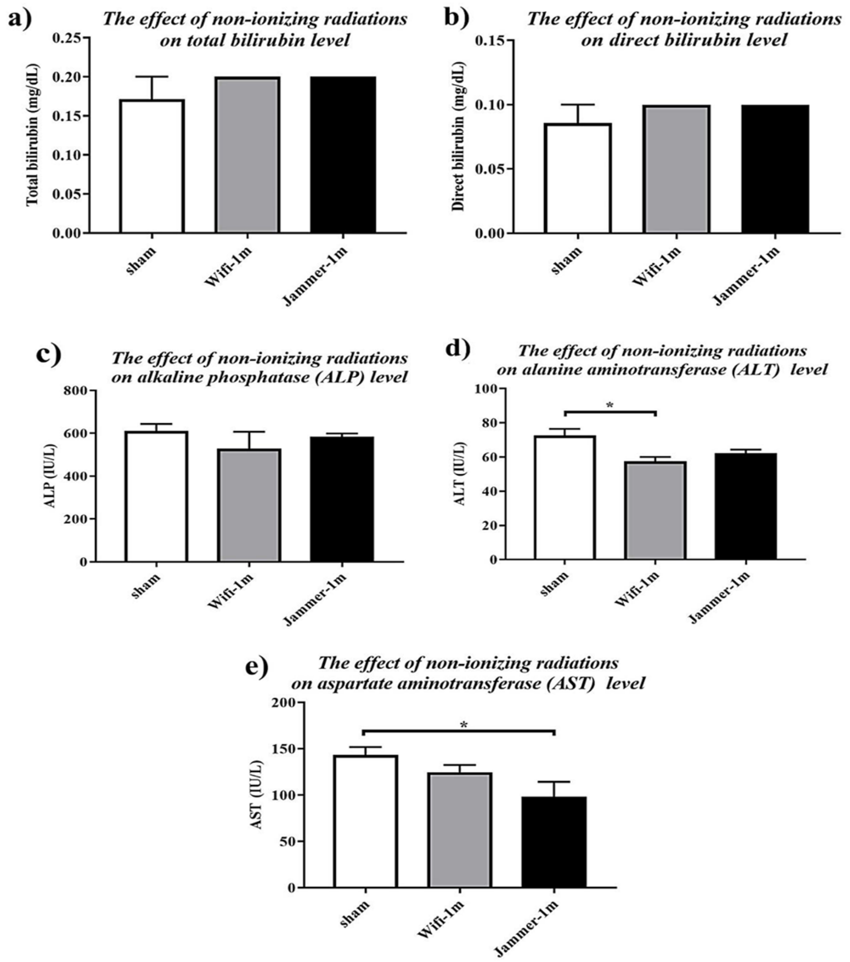

Figure 4: Illustration of the impact of Wi-Fi and jammer radiation on blood parameters related to liver function. (**a**) Total bilirubin levels did not show a significant difference between the studied groups. (**b**) Direct bilirubin levels also did not exhibit a significant difference among the groups. (**c**) Alkaline phosphatase (ALP) levels were not significantly different between the studied groups. (**d**) However, alanine aminotransferase (ALT) levels displayed a significant difference between the sham and Wi-Fi groups at a distance of 100 cm. (**e**) Aspartate aminotransferase (AST) levels showed significant differences between the sham group and the Wi-Fi groups, but not in the other groups. Statistical significance is indicated by *P<0.05.

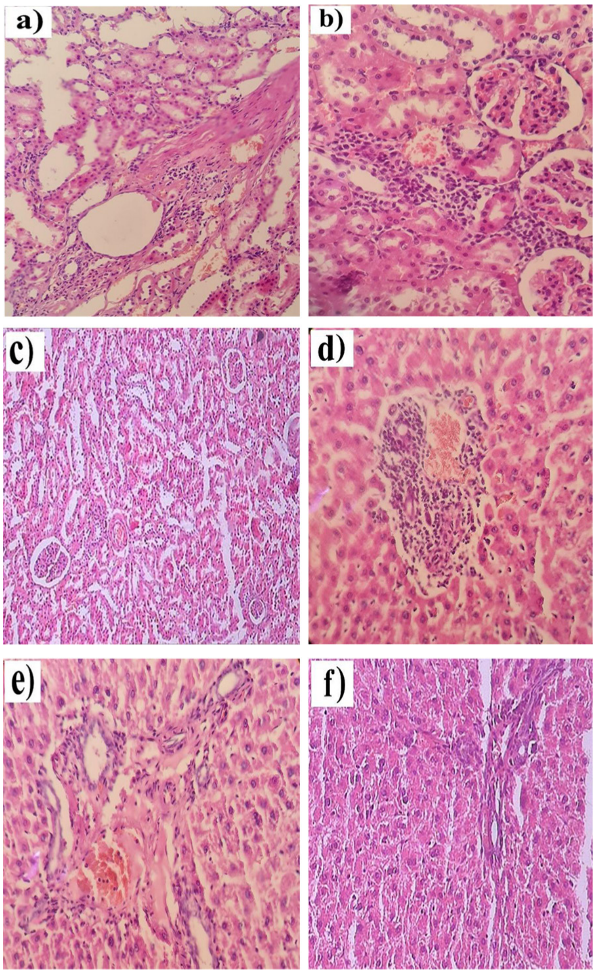

Figure 5: Illustration of the effects of Wi-Fi and jammer radiation on kidney and liver tissues, as assessed through histopathological analysis. (**a**) In the kidney of the jammer group, there is evidence of perivascular chronic inflammation. (**b**) The kidney of the Wi-Fi group shows peritubular chronic inflammation. (**c**) The sham group (without irradiation) displays normal renal tissue. (**d**) The liver of the jammer group exhibits portal chronic inflammation and piecemeal necrosis. (**e**) The liver of the Wi-Fi group shows portal inflammation. (**f**) The sham group (without irradiation) presents normal liver tissue. The study reveals mild portal and peritubular inflammation in the liver and kidney, respectively, resulting from exposure to Wi-Fi and jammer radiation.

disease [29].

For assessing renal function more accurately, creatinine is considered a more suitable marker than BUN because it is less influenced by nutrition and hydration factors [30,31]. Our results showed that serum creatinine levels in animals exposed to Wi-Fi and jammer radiation for two weeks remained within the normal range and did not significantly differ between the groups (Figure 2b). These results are consistent with the studies performed by Borzoueisileh et al. and Castro et al. [23,31], but in contrast with the findings from Imam Hasan et al. who reported an increase in serum creatinine levels after 60 days of daily radiation exposure [32].

Due to their high blood perfusion and aerobic mechanisms, kidneys are susceptible to radiation, producing significant levels of reactive oxygen species (ROS) and a limited antioxidant defense system [33]. Despite progress, the cellular and molecular mechanisms that underlie the effects of radiation on the kidneys remain poorly understood [33]. To improve the accuracy of future studies, it is suggested that longer radiation durations and closer distances between animals and radiation sources should be considered.

The liver is an organ with high metabolic activity, responsible for processing nutrients and converting them into usable and storable materials [34]. Additionally, it synthesizes bile and plasma proteins, while detoxifying harmful substances such as drugs into safe compounds [35,36]. To assess liver function, medical professionals utilize a series of tests, which includes AST, ALT, ALP, total protein, globulin, albumin, and total and direct bilirubin. These tests assist in evaluating the extent of liver damage [37].

Aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase levels are commonly analyzed to assess the liver's enzymatic defense mechanism through serum [38].

Our data analysis indicates that the concentration of alanine aminotransferase in the Wi-Fi group significantly decreased compared to the sham group. Additionally, the levels of aspartate aminotransferase and low-density lipoprotein (LDL) in the Jammer group significantly decreased when compared to the sham group. However, the LDL levels and aspartate aminotransferase in the Wi-Fi group did not show significant changes compared to the sham group (see Figures 3a and 4e for reference).

AST, an enzyme found in various tissues, including the liver, kidney, pancreas, heart, and red blood cells, is used as a biomarker for liver health in conjunction with ALT. Although ALT is more specific to liver health, AST and ALT levels, as well as their ratio (AST/ALT), are measured in the serum to assess liver function. Elevated level of these enzymes in the bloodstream indicates liver damage due to cellular injury [39].

Our study aimed to investigate whether exposure to radiation from Wi-Fi and jammer devices for two hours a day over two weeks had any impact on liver or kidney tissue damage and the concentration of these enzymes. Our findings suggest that such exposure did not induce cellular damage in the liver or kidney tissues and thus did not affect the concentration of these enzymes. This indicates that the increase in the enzymes observed in liver disease may have resulted from oxidative stress. This stress may stimulate the body's enzymatic defense system in response.

We also measured other factors including cholesterol, HDL, TG, bilirubin, and alkaline phosphatase concentrations in the study. Our results revealed no significant changes ($P>0.05$) in these factors among the different groups (Figures 2e, f, 3b, 4a, b, c). Taken together, our findings suggest that exposure to radiation from Wi-Fi and jammer devices does not affect liver or kidney function or the concentrations of other measured factors.

Our analysis of histological slides revealed mild portal inflammation in the liver and peritubular inflammation in the kidneys of the Wi-Fi group compared to the sham group. In the jammer group, the histopathology of the rat liver also exhibited portal inflammation, while chronic perivascular inflammation was observed in the kidney tissue when compared to the sham group (Figures 5a-f).

To assess the intensity of inflammatory activities, we found that enzyme activity measurements were the most useful method. These few alterations observed are likely associated with functional changes in the renal nephrons and hepatocytes due to the exposure to radiation effect for 2 hours per day for 2 weeks. However, these results are not consistent with the findings of Parul Chauhan and colleagues' project, who observed oxidative and histopathological changes in animals after 35 days of radiation exposure. The longer duration of exposure time in their study may have caused such changes [40].

Conclusion

In conclusion, our study highlights the effects of Wi-Fi and jammer radiation on liver and kidney tissues of rats. It underscores the necessity for additional research to fully comprehend the potential health impacts of these devices on humans. Future studies should explore the effects of prolonged radiation exposure and the consequences of being closer to the radiation source.

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Authors' Contribution

S. Farahanee contributed to the implementation and practical work of the plan. In addition, she was involved in data collection and statistical analysis of data, as well as in drafting articles. F. Kadivar was involved in the implementation and practical work of the plan. In addition, she was involved in data collection. F. Khajeh collaborated on taking photos of tissue slides and interpreting them. MB. Shojaeifard participated in all steps of study design, proofread, and supervised the research, and edited the manuscript. All the authors read, modified, and approved the final version of the manuscript.

Ethical Approval

The ethics committee at Shiraz University of Medical Sciences in Shiraz, Iran, approved the study. The guidelines set by the Iran National Committee for Ethics in Biomedical Research regarding the welfare of laboratory animals were followed (IR.SUMS.REC.1397.30).

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Conflict of Interest

None

Data Availability Statement

The data supporting this study's findings can be obtained from the corresponding author upon request.

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