EFFECTS OF RADIO FREQUENCY / MICROWAVE ON THE CHEMICAL COMPOSITION AND ANTIOXIDANT POTENTIAL OF *LYCOPERSICON ESCULENTUM* L.

Rammal Marwa¹, Rammal Hassan^{1,2*}, Jebai Fatima¹, Hijazi Akram¹, Badran Bassam¹

¹ Doctoral School of Science and Technology, Research Platform for Environmental Science (PRASE), Lebanese University, Lebanon

² Lebanese Universities, Faculty of Agronomy and Veterinary Sciences, Lebanon

*Corresponding Author Email: hasanrammal@hotmail.com

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ABSTRACT

In our work, the effects of radio frequency / microwave on the morphology, chemical composition and the antioxidant power of the plant *Lycopersicon esculentum* Mill. grown in our laboratory have been accomplished. The obtained results showed that after 10 days of exposure of this plant to electromagnetic fields with high frequency (1250 MHz) the stems were long with less leaves than the non exposed plant. However, after 20 days of exposure to this same frequency the stems were long and contain more leaves than the non exposed plant. These leaves were larger and thicker in comparison with those of the non exposed plant. On the other side, the phytochemical screening of the ethanolic extract revealed the presence of flavonoids in the exposed and non exposed plant. Alkaloids, phenols and saponins were only present in non exposed plant. The tannins were absent in the exposed and non exposed plant. Therefore, resins were highly expressed in the exposed plant. On the other side, the x-ray fluorescence indicated the presence of various trace elements more particularly niobium and molybdenum. After exposure, the amount of these elements varies. On the other hand, the DPPH and H₂O₂ tests showed an important decrease in the antioxidant potential after exposure to studied frequency. This decrease was from 42 % to 18 % at the concentration 0.5 mg/ml. All of these results show that the high frequency emitted by the electromagnetic fields exert a strong effect on the plant and by consequence on human health.

Keywords: *Lycopersicon esculentum*, electromagnetic fields, phytochemical screening, morphology, antioxidant activity.

INTRODUCTION

Due to the wide spread use of cellular phones the portion of the population exposed to high frequency electromagnetic fields, such as radiofrequency (RF) and microwaves, has increased rapidly. Also, with the increased application of electrical systems in household the environment of humans has changed and by consequence possible effects on human health may result from the interaction between all of these equipments and the consumption of various types of vegetables and fruits exposed to the radio frequency / microwave radiations. *Lycopersicon esculentum* Mill. belongs to the family Solanaceae. Tomato fruits are consumed fresh in salads or cooked in sauces and used as a flavoring in soups and meat or fish dishes. They are made into sweetened candies, dried fruits, and even into wine. Economically equally important are the processed forms such as purée, juice, ketchup, canned whole and diced fruits.

The effects of the EMR on human health may be direct and indirect. In our present study we aimed to investigate the effects of radio frequency / microwave radiation on the morphology, chemical composition and on the antioxidant potential of *Lycopersicon esculentum* Mill. grown in our laboratory.

MATERIALS AND METHODS

Plant Materials

Seeds of *Lycopersicon esculentum* have been sown in 6 pot, 3 of them were exposed the radio frequency / microwave radiation (1250 MHz) and the other 3 pot were placed in another room without exposure to radiation. After the growth of the plant, it was collected and left on air at room temperature for two weeks to be very well dried. After that, it was crushed up and ground to get homogeneous fine powder by a grinder and then kept in a dark place at room temperature till its use in the different studies.

Ultrasonic assisted extraction (UAE)

UAE involves the application of sound waves of high intensity and high frequency and their interaction with matter. UAE is a potentially useful technology because it does not require complex instruments and is relatively inexpensive. Under the action of ultrasonic solid and liquid particles are vibrated and accelerated, and thus the solutes diffuse rapidly from the solid phase to the solvent. If the ultrasonic intensity is increased in a liquid, then it reaches a point at which the intra molecular forces are not able to maintain intact the molecular structure, so that it breaks down and the bubbles are created, this process is called cavitation. The collapse of bubbles can produce physical, chemical and mechanical effects that lead to the disruption of biological membranes to facilitate the release of extractable compounds, increase the penetration of the solvent into the cells and improve mass transfer¹.

Ethanolic crude extracts

1 g of powder was added to 100 ml of ethanol in a flask, and the mixture was dipped in an ultrasonic bath for one hour at a temperature just below the boiling point of the ethanol. The extract was then filtered using Buchner funnel. Finally, the removing of ethanol was done under reduced pressure using a rotating evaporator at 40 °C.

Phytochemical screening

The preliminary phytochemical screening of various active compounds of *Lycopersicon esculentum* was accomplished according to the method of Muanda *et al.*².

Evaluation of the antioxidant activity

DPPH radical scavenging activity

The method of Farhan *et al.*³ has been used. 1 mL of different concentrations (100, 200, 300, 400 and 500 µg/ml) of diluted extracts of the plant in ethanol was added to 1 mL of DPPH

(0.15 mM in ethanol) and at the same time, a control consisting of 1 mL DPPH with 1 mL ethanol was prepared. The reaction mixtures were mixed very well by hand and then incubated in the dark at room temperature for 30 minutes and the absorbance was measured at 517 nm by a Gene Quant 1300 UV-Vis spectrophotometer. The ascorbic acid was used as a positive control and the ethanol was used as blank. The DPPH scavenging ability of plant extracts was calculated using the following equation:

$$\% \text{ scavenging activity} = [(Abs \text{ control} - Abs \text{ sample}) / (Abs \text{ control})] \times 100$$

The Abs control is the absorbance of DPPH + ethanol; Abs sample is the absorbance of DPPH radical + sample. Also, three controls have been prepared.

H₂O₂ radical scavenging activity

The method of Farhan *et al.*⁴ has been used. A solution of H₂O₂ (40 mM) was prepared in PBS (pH 7.4) and concentrations were determined spectrophotometrically at 230 nm. Different concentrations (100, 200, 300, 400 and 500 µg/mL) of the two extracts were added to a H₂O₂ solution (0.6 mL, 40 mM) and the absorbance of H₂O₂ was determined at 230 nm after 10 minutes against a blank solution containing the plant extracts without H₂O₂. Ascorbic acid was used as standard reference. The percentage scavenging of H₂O₂ was calculated using the following equation:

$$\% \text{ scavenged } [H_2O_2] = [(Abs \text{ control} - Abs \text{ sample}) / Abs \text{ control}] \times 100.$$

X-ray fluorescence spectrometry (XRF)

The analysis of major and trace elements in geological materials by x-ray fluorescence is made possible by the behavior of atoms when they interact with radiation. When materials are excited with high-energy, short wavelength radiation (e.g., X-rays), they can become ionized. If the energy of the radiation is sufficient to dislodge a tightly-held inner electron, the atom becomes unstable and an outer electron replaces the missing inner electron. When this happens, energy is released due to the decreased binding energy of the inner electron orbital compared with an outer one. The emitted radiation is of lower energy than the primary incident X-rays and is termed fluorescent radiation. Because the energy of the emitted photon is characteristic of a transition between specific electron orbitals in a particular element, the resulting fluorescent X-rays can be used to detect the abundances of elements that are present in the sample.

Microwave exposure

Microwave exposure of plants has been made to create real conditions of low level and permanent electromagnetic field exposure. We used a microwave source at the frequency of 1250 MHz and patch antennas to illuminate the plants. The power has been regulated to create a field of 6 V/m. The experiment has been carried out for 20 days permanently.

Statistical Analysis

All analyses were carried out in triplicates. The results of scavenger activity, were performed from the averages of all samples reading Mean $\pm$ SD used Excel 2003.

RESULTS AND DISCUSSION

Germination and growth of seeds

The obtained results showed that after 10 days of exposure of *Lycopersicon esculentum* to radio frequency / microwave radiation (1250 MHz) the stems were long and contain less leaves than the non exposed plant. However, after 20 days of exposure to this same frequency the stems were long and contain more leaves than the non exposed plant. These leaves were larger and thicker in comparison with those of the non exposed plant.

Phytochemical screening

In order to study the chemical composition of the ethanolic extract prepared from *Lycopersicon esculentum*, a phytochemical screening was performed allowing to observe the effects of the exposure to the chemical ingredient of the studied plant and by consequence consider the possible medical uses that may have this plant as several studies have demonstrated the positive correlation between the phytochemical composition of plants and their medicinal uses³⁻⁷.

Table 1: Phytochemical screening of the ethanolic extract of *Lycopersicon esculentum*

	Exposed	Non exposed
Tannins	-	-
Resins	+++	+
Saponins	-	+
Alkaloids	-	+
Flavonoids	+	+
Phenols	-	+

The results obtained from the phytochemical screening presented in Table 1 show that *Lycopersicon esculentum* is rich in various secondary metabolites at different concentrations. Indeed, we note the presence of saponins, alkaloids, flavonoids, phenols and resins in the non exposed plant. However, saponins, tannins, alkaloids and phenols were disappeared after exposure. Also, the phytochemical screening revealed an increase in the amount of resins after exposure. These results show that exposure to radio frequency / microwave radiation changes the chemical composition of the plant and by consequence it may decrease the possible biological properties of this plant.

Trace elements

The results of the analysis of 1 g of powdered tomato by XRF (Figure 1) show that the tomato contains various forms of metals particularly niobium and molybdenum. These two metals are of great importance in medicine. In humans, four enzymes depend on molybdenum: sulfite oxidase, xanthine oxidoreductase, aldehyde oxidase, and mitochondrial amidoxime reductase⁸. People severely deficient in molybdenum have poorly functioning sulfite oxidase and are prone to toxic reactions to sulfites in foods⁹ and some niobium alloys are physiologically inert and thus hypoallergenic. For this reason, niobium is found in many medical devices such as pacemakers¹⁰. After exposure of tomato to radio frequency / microwave radiation the metal composition varies. The potassium disappears while nickel appears. In addition, exposure to microwave radiation affected the content of certain metals. In fact, the content of niobium and molybdenum decreased (Figure 2), this can reduce the antioxidant activity since molybdenum is an antioxidant factor.

Figure 1: X ray fluorescence spectrometry of tomato non exposed to radio frequency / microwave radiation

Figure 2: X-ray fluorescence spectrometry of tomato after exposure to radio frequency / microwave radiation

Antioxidant activity

A number of studies on flavonoids, polyphenols have indicated that they have antioxidant potential and the ability to react as scavengers of free radicals. These phytoconstituents can exert multiple biological effects against tumors, heart disease and various diseases due to their radical activity. In our study, the antioxidant potential of exposed and non exposed plant to radio frequency / microwave radiation was compared *in vitro* using two different methods: DPPH and H_2O_2 . The Figure 3 shows an important decrease in the antioxidant potential of different concentrations of *Lycopersicon esculentum*. This decrease was concentration-dependent. As shown in this figure, the antioxidant potential of *Lycopersicon esculentum* decreases from 42 % to 18 % for the concentration 0.5 mg/ml after exposure to the radio frequency / microwave radiation.

Figure 3: Antioxidant activity of exposed and non exposed plant using the DPPH test

To confirm these results, another *in vitro* test was then applied, the H_2O_2 test. H_2O_2 is an important reactive oxygen species because of its ability to penetrate biological membranes. However, it may be toxic if converted to hydroxyl radical in the cell¹¹. Scavenging of H_2O_2 by the plant extracts may be attributed to their phenolics, which donate electron to H_2O_2 , thus reducing it to water. Figure 4 shows an important decrease in the antioxidant potential at the concentration 0.5 mg/ml 49 to 18 % after exposure to EMF.

Figure 4 : Antioxidant activity of exposed and non exposed plant using the H_2O_2 test

CONCLUSION

The results of our work demonstrated the effects of radio frequency / microwave radiation on the morphology, chemical composition and antioxidant potential of *Lycopersicon esculentum*. This microwave has led to a decrease or sometimes to the disappearance of some secondary metabolites responsible for some biological

property such as antioxidant activity. Also, our results showed the disappearance of some trace elements involved in different metabolic processes. At the same time, radio frequency / microwave radiation leads to a significant decrease in the antioxidant activity of this plant and by consequence it reduces its nutritional value and its ability to intervene in the prevention of certain diseases related to oxidative stress.

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