**ORIGINAL ARTICLE** 



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Antioxidant enzyme response of medical plant Persian *Fenugreek (Trigonella foenum-graecum L.)* to irrigation with microwaves treated water

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Article History:	ABSTRACT (Deck for updates
Received on: 04 Jul 2020 Revised on: 04 Aug 2020 Accepted on: 25 Aug 2020 <i>Keywords:</i>	The research, influences of different energy microwaves treated water on the antioxidant enzyme of <i>Trigonella foenum-graecum</i> (T. foenum ) were studied. The analysis was carried out in 2020 in the laboratories of Ecology Department, College of Science under stable variables such as temperature, humidity, sunlight. The antioxidant enzyme. The catalase (CAT), ascorbate peroxi-
T. foenum, irrigation, microwaves, Antioxidant enzyme, germination	dase (APX), superoxide dismutase (SOD) and peroxidase (POD) activity in T. foenum (hypocotyl and radical) were measured after twenty days of experiments. Results showed that germination was increased with water energy, and all enzymes activity were increased in low energy treated water gradually but decreased rapidly in high energy treated water. hypocotyl showed a higher response than Radical. (CAT) and (SOD) enzymes activity rises slowly in 30w* until it reaches a peak in 50 w * 120 s then decreases rapidly in 70 w * 150 s and whenever the time of treatments increased the enzyme becomes less effective. Both (APX) and (POD) had the same response but showed an increase in activities at 30W. The results improve that microwaves treated water-induced antioxidant enzyme response in <i>T. foenum</i> . While the specified energy level and duration showed a better growth rate compared to regular watering water. The results of this research serve those interested in developing cultivation and water treatment methods and improving our understanding of T. foenum irrigation behaviour using microwave treated water.

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# INTRODUCTION

Microwaves are electromagnetic waves with power between 300MHz and 300GHz (Ishii, 1995). They are not heating energy, but heat produces through their interaction with molecules (Benvenuti *et al.*, 2001). The use of microwave technology is increasing in the industry, especially for food warmers and cooking appliances. Several studies on changes in water exposed to microwaves include a change in pH and water molecules (Wong *et al.*, 2009).

In a survey of (Brodie *et al.*, 2012) on the use of microwave in the Lucerne treatment. The total content and loss of dry matter and ratios of complete protein were measured in digest residue compared

to untreated samples during cellulose digestion in laboratory pepsin.

Microwave treatment proved beneficial, as it led to significant animal growth rather than maintaining possible weight, which is in the untreated Lucerne group. Likewise, In a study of (Abdelmoumen and Idrissi, 2009) on the use of microwave in the treatment of oats in the laboratory to digest starch, which is useful in making feed monogastric animals such as horses. The seeds and germination mechanism is an essential role in the development of agriculture and effective management programs. Also increased interest in medicinal plants that have industrial and medicinal value (Kevseroglu et al., 2000). T foenum belongs to dicotyledonous plant Leguminacae family (the Fabaceae). Used as food, medicine, even as animal feed (Acharya et al., 2008). There are no published on the microwaves treated water effects on T. foenum seeds germination requirements or its impact on their antioxidant enzyme response. This study on the possibility of improving the germination and growth characteristics of the T. foenum plant may help in developing an ideal cultivation program. The main purposes of this research are to determine the effects of microwave treated water on seed germination and the antioxidant enzyme response of T. foenum to improve the cultivation strategy.



Figure 1: CAT enzyme activity of T. foenum (Hypocotyl and Radical) irrigation with different Microwaves treated water

#### **MATERIALS AND METHODS**

T. foenum (TF-925) seeds were obtained from the seed bank in Iraq, and all studies were conducted at laboratories of Ecology Department, College of Science, Kufa University. CRD (completely randomized design) with four replications was used in all experiments. The seeds were sterilized according to the method (Benvenuti *et al.*, 2001)

with a 3% solution of calcium hypochlorite for 5 minutes, and placed in Petri dishes after rinsing



Figure 2: APX enzyme activity of T. foenum (Hypocotyl and Radical) irrigation with different Microwave streated water

them with sterile distilled water. Four replications were applied to each treatment of 50 seeds on filter paper in each experiment and moisturized with 9 ml distilled water or a test solution. Germination, The antioxidant enzymes (CAT), (APX), (SOD) and (POD) activity in (hypocotyl and radical) were recorded in 20 days after the experiment in controlled environmental conditions. Germination rates were measured. (Muthik et al., 2019). All samples were homogenized (0.5 g) in the 0.1 m cold phosphate ice solution (pH 7.5) containing 0.5 mL molar of EDTA with pre-formed grout and mortar. The homogeneity was transferred to centrifuge tubes and centrifuged at 4C in the Beckman refrigerated centrifuge at 15000 r.p.m. For 15 minutes. The floating material was transferred to 30 ml tubes, and reference was made to the enzyme extract, then



Figure 3: SOD enzyme activity of T. foenum (Hypocotyl and Radical) irrigation with different Microwaves treated water

enzymes activity measured according to (Guda *et al.*, 2018).

#### Water treated by microwave

The household type microwave (650 w) was used to prepare 16 samples of water according to the following energy levels: 30 w, 50 w, 70 w, 90 w, exposure time is 60 s, 90 s, 120 s, 150 s. It is provided

Power(w)	(30w) %	(50w) %	(70w) %	(90w) %
Time (s)				
The 60s	93.75c	94.71c	94.87c	95.76a
The 90s	95.53a	95.35a	96.31a	96.51a
120s	95.76a	95.43a	97.12 ac	98.43ac
150s	96.32a	92.54c	85.50ab	88.3ab

Table 1: Germination of the seeds after twenty days of irrigation with different Microwaves treated water

Different letters in each column mean significant differences at P>0.05 level

by a magnetron operating at 2450MHz in continuous mode. Use 100 ml of distilled water in Borosil glass beaker for each treatment at an initial temperature of 25 ° C. The energy provided for the water is calculated from the following equation, according to the method of (Mehrafarin *et al.*, 2010).

 $P = C_p K \Delta T (m/t)$ 

where *P* is the apparent power absorbed by water sample (J s<sup>-1</sup>) = 640 w, *C<sub>p</sub>* is the heat capacity of water (J ml<sup>-1o</sup>K<sup>-1</sup>), *K* = 4.184 is a factor to convert thermal, chemical cal ml<sup>-1o</sup>K<sup>-1</sup> to watts (J s<sup>-1</sup>),  $\Delta$ T (°C) is the difference between initial temperature and final temperature of the water, m is the mass of water (g), and t is the duration of microwave energy application.



Figure 4: POD enzyme activity of T. foenum (Hypocotyl and Radical) irrigation with different Microwaves treated water

Statistical analysis

Data were analyzed by analysis of variance (ANOVA). Significant differences between treatments were considered by SPSS (Ver.17) and (LSD) Test.

# **RESULTS AND DISCUSSION**

The mean germination and anti enzyme activity of T. foenum irrigation with different Microwaves treated water are given in Table 1 and Figures 1, 2, 3 and 4.

The change in the germination of the seeds after

seven days were tabulated in Table 1. The results showed the highest value in (90w) \* (120s) equal 98.43 and the lowers value in (70w) \*(150s) equal 85.50. The highest values, in general, were in the treatment of (90w), and the lowest values were in the treatment of (30w). This indicates that the germination rate increased with increasing the water energy, or that irrigation with microwave treated water in a higher power. This may be because the excess kinetic energy of the treated water increased the conversion of seed starch into sugars that agree with (Pahlevani *et al.*, 2008).

The change in antioxidant enzymes of seedlings after 20 days was values of CAT, APX, SOD and POD activity in T. foenum radical and hypocotyl were plotted in Figures 1, 2, 3 and 4. Results showed that the CAT enzyme activity shows the highest value in 50 w \* 120 s was 27 in hypocotyl, while the lowest value in 70 w \* 150 s was 3 in Hypocotyl Figure 1. The APX enzyme activity shows the highest value in 30 w \* 60 s was 3.7, while the lowest value in 90 w \* 150 s was 0.4 in Hypocotyl Figure 2. The SOD enzyme activity shows the highest value in 50 w \* 120 s was 15.83, while the lowest value in 90 w \* 150 s was 1.52 in Radical Figure 3. The POD enzyme activity shows the highest value in 30 w \* 120 s was 0.28 in hypocotyl, while the lowest value in 90 w \* 150 s was 0.04 in Hypocotyl Figure 4. Antioxidant enzymes response of plants depends on both power level and exposure duration. Around 90 s exposure duration at 30 w and 50w power were observed as the best value for seedling enzymes. The analysis of the results shows that high power levels and exposure durations are always harmful to the growth trails and enzymes response. This is maybe because microwave energy increases the kinetic energy of water molecules. Increasing the kinetic energy of the water can dissolve some substances or free radicals that are harmful to growth (Mehrafarin et al., 2011), or maybe the microwave treated water affected three parts of a seed (embryo, endosperm, and seed coat) that are sensitive to microwaves (Muthik et al., 2016)

#### and (Guda et al., 2017).

# CONCLUSIONS

The current study shows that the effect of irrigation with microwave-treated water with different energy levels and exposure times on fenugreek seeds increased germination levels with increased water energy. Increase water energy cussed rapidly decreased in antioxidant enzymes activity. Low energy levels and exposure time (30 w with an exposure time of about 90 s) showed an increase on antioxidant enzyme response of the plant. However, the more energy of water increased, the more effect on antioxidant enzyme response of the plant. Antioxidant enzyme response abnormalities were noted. The results are beneficial in biotechnology to increase production.

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## **Conflict of Interest**

The authors declare that they have no conflict of interest for this study.

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