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THE EFFECT OF MAGNETIC FIELD ON GROWTH AND ACTIVITY OFAUXINOXIDASE ENZYME AND THE QUANTITY AND QUALITY OF GARLIC ESSENCE *(ALLIUM SATIVUM L.)*

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ABSTRACT

Magnetic fields are the environmental factors affecting biological phenomena of living organisms, including plants. In this study, the effect of magnetic fields on growth factors and activity of auxin oxidase enzyme and the quality and quantity of garlic essence (*Allium sativum* L.) was studied. The cloves were exposed to magnetic field intensity of 1, 2 and 4 mTdaily for 15 minutes on 3 consecutive days. Each treatment was repeated at least 10 replications in each 15 samples were performed in the form of completely randomized blocks. Based on the results obtained, the percentage of germination and growth was reduced in the treated samples. Increasing the intensity of the magnetic field significantly increased auxin oxidase enzyme activity. Quantitative assessment of garlic essence showed that increased magnetic field leads to a significant increase in essence amount. Qualitative assessment of garlic essence using GC/MS device showed the presence of 32 compounds in all samples from which 4 compounds were dominant. There was no differences in the chemical composition of the essential in the treated samples. The results indicated that although magnetic field has no positive effects on garlic growth, it can be used as a tool to increase helpful pharmaceutical essences and compounds.

Keywords: magnetic field, phenol, leaf protein, growth percentage.

INTRODUCTION:

Garlic, with scientific name *Allium sativum* L. , is an Asparagales plant, from Alliaceae family and Allium genus (Saeidi, 2012). Garlic has abundant and unique medicinal properties and nutritional values. Garlic is rich in folic acid, vitamin C, calcium, iron, magnesium, potassium and small amounts of zinc and vitamins B1, B2 and B3. Garlic has been traditionally used for blood purification. Garlic contains sulfur compounds that stimulate the immune system and has high potential to destroy cancer tumors. Also, because of its high nutritional value and containing lots of minerals and vitamins, the plant has been always under attention.

Today, the devices producing magnetic and electromagnetic fields in the environment have increased concerns about the effects of these fields on plants, animals and even humans. Therefore, the effect of magnetic fields on living organisms is the subject of many researches. Matsuda et al. in 2007 investigated the effect of magnetic field on the growth and germination of strawberry plant. Carbonel and colleagues in 2010 examined the effect of the magnetic field on the germination of rice seeds treated with magnetic field. Bashisth et al. in 2013 investigated the effect of magnetic field intensity of 50 to 250 mT and duration of 1-4 hours on the germination and growth of sunflower seeds. Majd and Shabrangi in 2008investigated the effect of magnetic field intensity of 1600 and 1800 gauss on lentil plants. Faizi in 2011examined the effect of the magnetic field intensity of 50, 100, 150 mT on the germination and seedling growth of wheat.

It has been tried in this research to investigate the effect of magnetic fields on growth and activity of auxin oxidase enzyme and the quality and quantity of garlic essence(*Allium sativum* L.) in order to find another solution for improving the status of agriculture and cultivation of medicinal plants.

MATERIALS AND METHODS

In this study, two magnets with1 × 2 × 5 cm dimensions were used to create magnetic field intensity of 1, 2 and 4 mT with certain distances from treatment samples. So that, the distance of magnets from both sides of the treated samples for treatments of1, 2 and 4 mT were determined to be 118, 88 and 67 mm, respectively. These distances were measured by Gauss Meter to create magnetic fields with mentioned intensities. The cloves for the culture was prepared from Tehran market. 600 healthy cloves were selected (within 4 treatments of 150). The samples

were divided in certain envelopes to be exposed to the magnetic field in the next phase. Samples were exposed to a magnetic field three times for 15 minutes. 5 samples were exposed in each round and 150 samples were during each replication. It should be noted that at all stages, experimentwas performed at the same temperature and light conditions. Treatment (1) was under a magnetic field intensity of1mT, treatment (2) under a magnetic field intensity of2mTand treatment (4) under a magnetic field intensity of4mTand control treatment was under no magnetic field (0mT). Treatments were exposed to magnetic field for 3 consecutive days, every day for 15 minutes and after that, planting stage was performed in a completely randomized block design in November. After a period of 45 days, simultaneous sampling from germinated samples was carried out in order to extract leaf protein and assess auxin oxidase enzyme activity; overall harvest of the samples was carried out in May.

Growth indicators:

Growth indicators including the average number of leaves, the average percentage of germination, and the average length of aerial organ were measured during time cycles. The average number of cloves, the average wet and dry weight of roots, bulbs, leaf stems, and leaves were also measured.

Measurement of auxin oxidase enzyme activity:

Auxin oxidase enzyme activity was examined using Gordon and Weber method(1951). The mixture contained phosphate buffer, indole acetic acid, manganese chloride, and 2-4-dichlorophenol and immediately after adding the enzyme extract to it, absorption coefficient was recorded using spectrophotometer at a wavelength of 530 nm. Enzyme activity was calculated in terms of changes in absorption unit in a minute per gram of wet weight leaf ($OD.g^{-1}$ FW. Min^{-1}).

Qualitative and quantitative changes in the chemical composition of garlic essential oil:

Water distillation method and Clevenger extraction device were used to extract garlic essence. A small amount of essence was measured by insulin syringe after extraction. After dewatering the essence by anhydrous sodium sulfate (Na2SO4), the above essence was diluted by n-hexane (Merck) solvent (1 to 10) for qualitative analysis. The sample was injected to gaseous chromatography device combined with mass spectrometry (GC / MS).

Statistical analysis:

Experiments were designed according to the randomized complete blocks. Each treatment

had ten replications and each replication 15 samples. To measure the essence, plant extracts of three boxes (as three replications) were used. Three samples were randomly taken and measured from three boxes to examinephenol and enzyme in each experiment. The analysis of all data from different experiments was performed using Anova software and Spss (Version 16). The average of measured indicators was grouped using Duncan test ($p \leq 0.05$).

RESULTS

Growth indicators:

Regarding the effect of magnetic field on the growth indicators, the results show that the magnetic field with applied intensities reduce the growth indices in garlic; so that the results of the average percent of germination and average aerial height during seven note-taking (26, 33, 40, 54, 75, 89 and 107 days after planting) showed that the different intensities of magnetic field decrease the indices of all the treatments compared to the control treatment (Figure 1). On the other hand, in other growth indicators including the average number of leaves, the average percentage of germination, the average number of cloves and average wet and dry weight of roots, bulbs, leaf stems, and leaves, the magnetic field reduces the amount of above indicators in all the treatments compared to the control treatment (Figure 2).

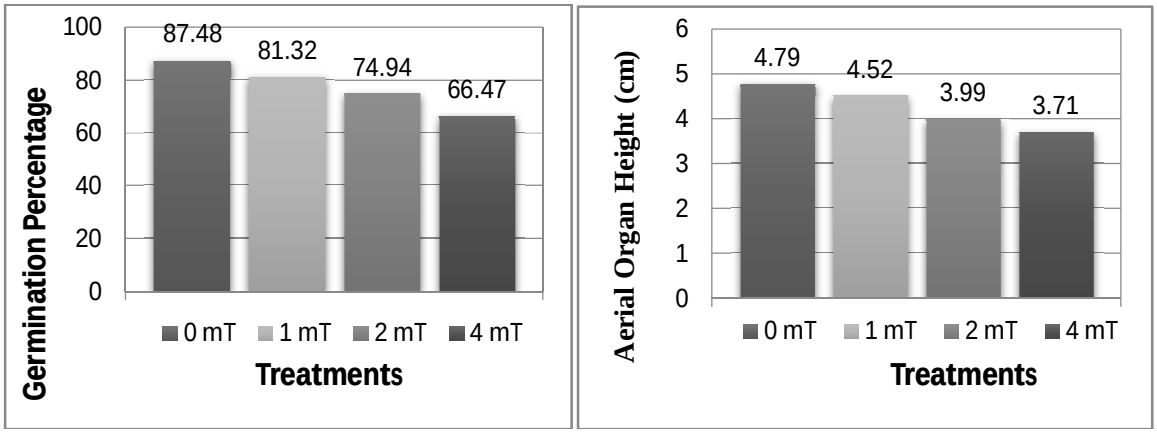

Figure 1. The average height of aerial organ (Right), the average percentage of germination (Left)

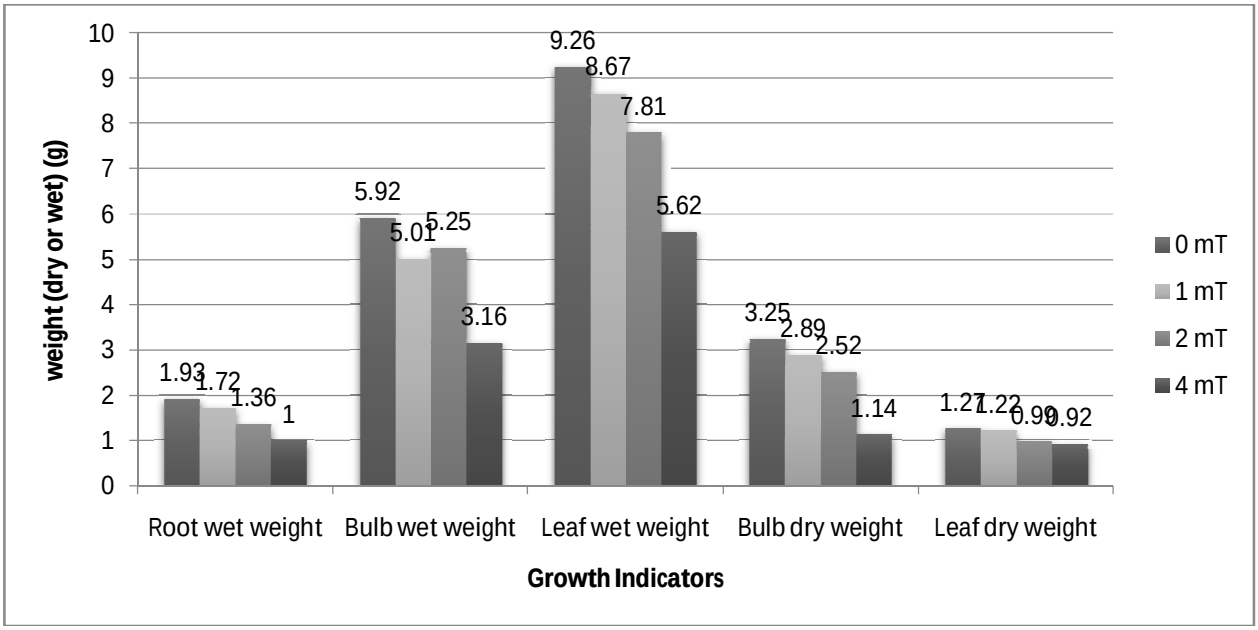

Figure 2: The average of Growth Indicators

Measurement of auxin oxidase enzyme activity:

Regarding the effect of magnetic field on the auxin oxidase enzyme activity, the results show that the average activity of this enzyme in the different treatments is significant ($p \leq 0.05$). So that the highest activity of auxin oxidase enzyme was observed in the treatment of 4 mT ($OD.g^{-1}.Fw.min^{-1}$ 0/48) and lowest one in the control group ($OD.g^{-1}.Fw.min^{-1}$ 0/25)

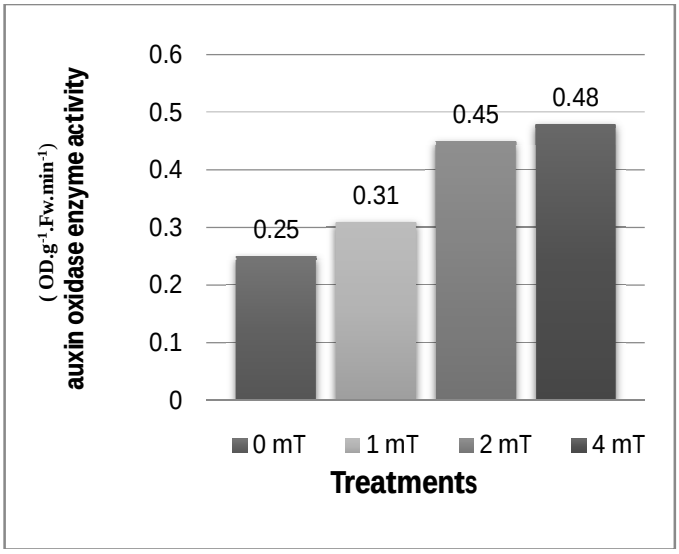

Figure 4: The average level of auxin oxidase enzyme activity

The average of quantitative and qualitative measurement of essence:

The average of quantitative measurement data of garlic in different treatments showed that the difference in the averages is significant ($p \leq 0.05$). Grouping the averages indicated that highest average of quantitative measurement was in the treatment of 4 mT(1.30ml) and the lowest one in the control group (0.70ml) (Figure 6).

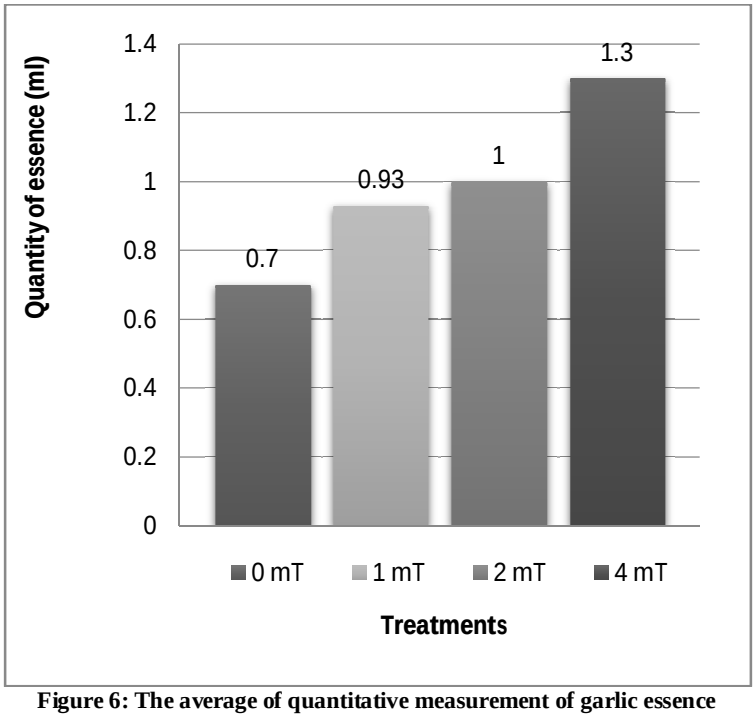

Figure 6: The average of quantitative measurement of garlic essence

Table 3-14. The percentages obtained from garlic compounds in 4 treatments (control 4, 1, 2 and 4 mTesla) using the spectroscopy of garlic essence by GC / MS

No.	Combining Name	Control treatment	Treatment of 1 mT	Treatment of 2 mT	Treatment of 4 mT
1	Allyl sulfide	0.03	0.04	0.04	0.04
2	1-Propene, 1,1'-thiobis-	0.1	0.1	0.1	0.1
3	3,4-Dimethylthiophene	0.02	0.02	0.01	0.02
4	1,3-Dithiane	0.02	0.02	0.02	0.02
5	Disulfide, methyl 1-propenyl	0.02	0.02	0.03	0.03
6	Disulfide, methyl 2- propionyl	2.64	2.95	3.10	3.11
7	Ethanethioamide, N, N-dimethyl-	0.06	0.08	0.07	0.07
8	Dimethyl trisulfide	3.18	2.96	3.14	3.22
9	Allyl disulfide	2.10	2.13	2.11	2.15
10	2-Vinyl-1,3-dithiane	1.4	1.7	1.6	1.8
11	3-(Allylsulfanyl) propanoic acid	1.3	1.2	1.3	1.3
12	2-Ethylidene [1,3] dithiane	0.07	0.08	0.08	0.08
13	Allyl trisulfide	0.08	0.09	0.09	0.09
14	Diallyl tetrasulphide	0.52	0.48	0.53	0.54
15	Dioxane	0.01	0.02	0.02	0.01
16	Diallyl Disulfide	26.13	25.98	26.16	26.17
17	Methyl (allylsulfanyl) acetate	1.20	1.18	1.21	1.25
18	6-(Methylthio)hexa-1,5-dien-3-ol	0.02	0.02	0.01	0.01
19	1,3,5-Trithiane	0.01	0.02	0.02	0.02
20	2-Mercapto-3,4-dimethyl-2,3-dihydrothiophene	0.03	0.03	0.03	0.03
21	3-Vinyl-3,4-dihydro-1,2-dithiane	0.04	0.06	0.05	0.05
22	1-Ethyl-2-methyl-4-pentenyl methyl ether	0.06	0.07	0.06	0.06
23	1-Heptene, 5-methoxy-4-methyl-	0.03	0.03	0.03	0.04
24	2,5-Dimethyl-1,3,4-thiadiazole	0.05	0.04	0.04	0.04
25	Benzothiofuran	0.58	0.59	0.57	0.61
26	Methyl allyl trisulfide	16.34	16.33	16.30	16.35
27	Diallyl trisulfide	18.27	18.39	18.42	18.48
28	Methyl allyl Disulfide	11.85	11.93	11.98	12.10
29	Dimethyl disulfide	2.19	2.12	2.30	2.30
30	Diallyl disulfide	2.40	2.40	2.42	2.42
31	Dithiocyclopenthane	0.39	0.44	0.45	0.48
32	Methyl thio propanol	0.16	0.15	0.14	0.17
33	Methyl propyl disulfide	0.20	0.18	0.17	0.21

DISCUSSION:

According to the results, it can be interpreted that, in general, magnetic field with the intensities used in this study reduced growth indicators in garlic. Consistent with Kordas in 2002 and Dhawi 2009, it can be stated thatmagnetic fields affect both the activity of the ions and polarization of dipole molecules in living cells, leading to the creation of oxidative stress in the plant.What can be interpreted from examining the effect of magnetic field on growth indicators is that, intensities of 1 and 2 mT might have insignificant effect on the plant. However, it can be said that the intensity of 4mT applied to the treatment plants compared to the

control plants is considered as a stress and thusthe intensity of 4 mT on garlic is less able to control the damages caused by stress.

Several studies show that environmental stress affect theauxin oxidase enzyme activity. For example, in 1973, Mills et al reported the reduced activity of auxin oxidase enzyme in wheat under drought stress. The increases activity of this enzyme in onion sprouts treated with electromagnetic field by Xing-fu et al in 2003 reported that the results obtained in this study is consistent. Auxin oxidase enzyme is involved in plant growth through internalauxin regulation (Mills et al, 1973). It indicates that the activity level of this enzyme is inversely related to growth which is consistent with the results obtained in this study; because magnetic field with different intensities used in this study reduces growth and increases the auxin oxidase enzyme activity.

Studies have shown that environmental and stressful factors always affect chemical composition and essence of the plants (Zheljazkov&Warman, 2003). In this study, the compounds found in garlic's essential oil in 4 treatments (control 1, 2 and 4 m T), the dominant compounds of garlic essence used in this study include diallyl disulfide, diallyl sulfide, allyl disulfide and methyl allyl disulfide, respectively. Along with the dominant essence productive compounds, about 30 other compounds were also measured where the change in their relative amounts in the treatment group was not significant compared to the control group. Among dominant compounds, 3compounds of diallyl disulfide, diallyl sulfide, allyl disulfide have medicinal properties. Diallyl disulfide was the most effective compoundin biological and antimicrobial activities of garlic (Ghanbari et al., 2013; Reuter et al, 1996; Ogara et al, 2000 ; Ross et al ,2001; Busquet et al , 2005). Also, allyl disulfide reduces blood glucose and increases insulin concentrations in the blood. Diallyl sulfide is another effective medicinal compound in garlic. Diallyl disulfide in garlic has anti-cancerproperty. So, comparing the percentages obtained in 4 treatments (control, 1, 2 and 4 mT) showed that the greatest amount of this compound is in 4 mT (Shan, 2013; Clemente, 2011).

In the quantitative measurement of garlic essence, the average of data shows that with increasing magnetic field intensity, the amount of extracted essence has also increased. Since the essence compounds are secondary metabolites and they often play important role in plant defensive system against pathogens and environmental stresses

(Alberts, 2010), it can be concluded that the applied magnetic field on garlic creates stress for plants and increased field intensity up to 4 mT intensified the stress and thus the plant was trying to increase resistance to stress by increasing the synthesis of secondary metabolites (essence).

It can be concluded from the research that increased amount of essence in the plants exposed to magnetic field intensity of 4mT indicates the creation of stress conditions with negative effect on growth. While these effects are insignificant in intensities of 1 and 2 mT.Therefore, the plant has become adapted with stress in these intensities and thus magnetic field intensities of 1 and 2 mT have no significant effect on garlic growth. It is recommended to cultivate this plant in places where the magnetic field intensity of the agricultural lands is less than 4 mT.

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