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(RESEARCH ARTICLE)

## 1 Effects of 0.2 mT magnetic field exposure on the growth of red chili (*Capsicum annuum* L.) infected with pathogenic fungus *Fusarium oxysporum*

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### 1 Abstract

*Fusarium* pathogens not only affect growth but also cause death of chili plants. Our previous research using magnetic fields revealed that in tomatoes, inhibition of seed germination and growth by *Fusarium* infection can be restored by a 0.2 mT magnetic field. In current study we investigated whether the same 0.2 mT magnetic field exhibit the same effects on red chili plant which has infected with *Fusarium oxysporum*. The experiment was arranged using a randomized block design (RCBD) with two factors. The first factor is the exposure time of 0.2 mT magnetic field namely 0 min ( $M_0$ ), 7 min 48 sec ( $M_7$ ) and 15 min 36 sec. The second factor is the soaking time of chilli seeds in *F.oxysporum* suspension namely 0 min ( $F_0$ ) and 60 min ( $F_{60}$ ). All treatment repeated 5 times. The results revealed that the inducement of 0.2 mT magnetic field for 15 min 36 sec significantly increase leaf area, total chlorophyll content and dry weight of the plant at 28 days after planting (DAP). Seeds infection with *F. oxysporum* decreased leaf area (except for  $M_7F_{60}$ ), total chlorophyll content (except for  $M_0F_{60}$ ), carbohydrate content (except for  $M_7F_{60}$ ) and increased dry weight of the plant (except for  $M_0F_{60}$ ). These data suggest that the magnetic field at the density of 0.2 mT is able to maintain the growth of chilli plants despite being infected by pathogenic *Fusarium* fungi.

**Keywords:** Red chili; *Capsicum annuum*; *Fusarium oxysporum*; Magnetic field

### 1. Introduction

As a horticulture commodity that is very popular with people in many countries because of its taste and nutritional content, the red chili (*Capsicum annum* L.) has good development prospects [1]. Market demand for red chili continues to increase from year to year, however the request is not always fulfilled because of cultivation constraints such as pests and plant diseases that cause a decrease in production [2].

*Fusarium oxysporum* is a soil borne pathogen that causes *Fusarium* wilt in chili. In the soil, this pathogenic fungus can survive up to 10 years in the form of kladospores [3]. Efforts to overcome *Fusarium* infection is still done conventionally, using fungicides or superior seeds resistant to disease. The use of fungicides for *Fusarium* wilt has not satisfactory and additionally their inappropriate use can causes pathogenic organisms to become resistant [4].

There have been many studies on the use of magnetic fields to increase plant growth and agriculture production [5]. The results of these studies reveal that the magnetic field can give rise to a promising growth response in plants and can be developed as a technique to improve the quality of plant growth and production. Magnetic fields can increase the rate of germination of tomatoes [6], *Pinus tropicalis* M. [7], propagation of onion buds [8], even the production of plants including strawberries [9] and chickpeas [10]. Magnetic fields have also been shown to increase membrane permeability and increase nutrient concentrations of N, K, Ca, Mg, Cu, Fe, Mn, Na and Zn strawberry leaves [9], soybean

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lipid content [11], and accelerate the growth of roots, shoots, and increase the activity of germination enzymes and growth [12].

The magnetic field treatment on *Cuminum cyminum* [13], *Satureia hortensis* L. [14] and *Piper Nigrum* [15] found to have increased peroxidase activity. Peroxidase has an important role in the plant's defense system because it can recognize pathogens [16] and acts as a catalyst for lignin formation [17]. Lignin deposit into plant cell walls that are affected by disease is one of the plant's defense systems [18].

As an effort to overcome the obstacles of cultivating red chillies due to *F. oxysporum* infection amid difficulties of getting quality chili seeds, while on the other hand the magnetic field was proven to be able to encourage growth and yield of crop production so we tried to use a 0.2 mT magnetic field on red chili seeds infected with *F.oxysporum*.

## 2. Material and methods

### 2.1. Plant and fungus

The chili seeds used are the red chili cultivars which are obtained from an agricultural shop in Pasar Tengah, Bandar Lampung, Indonesia. The fungi isolate of *Fusariumoxysporum* obtained from SEAMEO BIOTROP Bogor, Indonesia.

### 2.2. Experimental design

The study was conducted in January to April 2019 at the Botany Laboratory and Microbiology Laboratory, Department of Biology, Faculty of Mathematics and Sciences, University of Lampung, Indonesia. The research was arranged in factorial using a randomized block design (RCBD) with two factors. The first factor is the exposure time of red chili seeds with a magnetic field of 0.2 mT for 0 min (M0), 7 min 48 sec (M7), and 15 min 36 sec (M15). The second factor is the soaking time of *F.oxysporum* suspension for 0 min (F0) and 60 min (F60). All trial applied with 5 replications.

### 2.3. Study parameters

The parameters measured were leaf area, total chlorophyll content, carbohydrate content, plant dry weight at 28 days after planting (DAP) and the correlation between the parameters above.

#### 2.3.1. Leaf area

Leaf area was measured when the plants were 21 and 28 days after planting (DAP). Leaf area was measured using the gravimetric method. The leaves of the chili plant that are picked are the 5th leaf from the shoots because it is the widest leaf.

#### 2.3.2. Total chlorophil content

To analyze total chlorophil content of the plant, the chili leaves were blended using acetone as the solvent. Chlorophil content were measured using uv-vis spectrophotometer Shimadzu 1800 at the wavelength of 646 nm dan 663 nm. Total chlorophil content in one gram of chili leaves calculated using formula belows.

$$\text{Total Chlorophil} = (17.3 \times \lambda_{646}) + (7.18 \times \lambda_{663})$$

Where:

$\lambda_{646}$  = absorbance value at the wavelength of 646 nm

$\lambda_{663}$  = absorbance value at the wavelength of 663 nm.

#### 2.3.3. Carbohydrate content

The chili carbohydrate content is measured 21 days after planting (DAP). The chilli leaf extract (1 ml) is put into a test tube, diluted with 2 ml of distilled water, 2 ml of concentrated H<sub>2</sub>SO<sub>4</sub>, and 1 ml of Phenol 5%, shaken and allowed to stand for several minutes. Carbohydrate content was measured using a Shimadzu UV-Vis 1800 spectrophotometer at a wavelength of 490 nm.

### 2.3.4. Dry weight

Whole plant dry weight of the chili was measured 7, 14, 21 and 28 DAP using analytical balance.

## 2.4. Statistical analysis

The data obtained were analyzed for the variant (ANOVA) and Fisher Pairwise test for post hoc tests using Minitab 17 program. In order to find out the degree of relationship between parameters and interaction effects between factor, Correlation analysis is also applied. All analysis and post hoc test results are set at 5% significance level.

## 3. Results and discussion

Effects of treatment units on leaf area, total chlorophyll content, carbohydrate content and whole plant dry weight of red chili are shown in Table 1.

**Table 1** Effects of treatment on the leaf area, total chlorophyll content, carbohydrate content, and plant dry weight of red chili plant

Treatment	Parameters							
	Leaf area (cm <sup>2</sup> )		Total chlorophyll (µg/ml)	Carbohydrate content (mg/g)	Whole plant dry weight (g)			
	21 DAP	28 DAP	28 DAP	28 DAP	7 DAP	14 DAP	21 DAP	28 DAP
M <sub>0</sub> F <sub>0</sub>	22.91 <sup>a</sup>	111.01 <sup>ab</sup>	1.902 <sup>c</sup>	0.2444 <sup>a</sup>	0.0041 <sup>b</sup>	0.0146 <sup>b</sup>	0.125 <sup>b</sup>	0.5355 <sup>ab</sup>
M <sub>0</sub> F <sub>60</sub>	21.09 <sup>a</sup>	82.3 <sup>bcd</sup>	2.855 <sup>abc</sup>	0.2058 <sup>a</sup>	0.0051 <sup>ab</sup>	0.0307 <sup>a</sup>	0.127 <sup>b</sup>	0.455 <sup>b</sup>
M <sub>7</sub> F <sub>0</sub>	22.86 <sup>a</sup>	65.17 <sup>cd</sup>	3.22 <sup>ab</sup>	0.2012 <sup>a</sup>	0.0049 <sup>ab</sup>	0.0175 <sup>b</sup>	0.0977 <sup>b</sup>	0.3682 <sup>b</sup>
M <sub>7</sub> F <sub>60</sub>	19.94 <sup>a</sup>	91 <sup>bc</sup>	1.98 <sup>bc</sup>	0.2332 <sup>a</sup>	0.0054 <sup>ab</sup>	0.0177 <sup>b</sup>	0.1022 <sup>b</sup>	0.4536 <sup>b</sup>
M <sub>15</sub> F <sub>0</sub>	35.7 <sup>a</sup>	132 <sup>a</sup>	3.67 <sup>a</sup>	0.2056 <sup>a</sup>	0.0046 <sup>ab</sup>	0.0222 <sup>b</sup>	0.1834 <sup>a</sup>	0.692 <sup>a</sup>
M <sub>15</sub> F <sub>60</sub>	28.35 <sup>a</sup>	54.1 <sup>d</sup>	2.70 <sup>abc</sup>	0.2006 <sup>a</sup>	0.0067 <sup>a</sup>	0.0199 <sup>b</sup>	0.1286 <sup>ab</sup>	0.3617 <sup>b</sup>

M = magnetic field of 0.2 mT; F = *F. oxysporum* infection; 0 = 0 min (as control). 7 = 7 min 48 sec; 15 = 15 min 36 sec. 60 = 60 min; DAP=days after planting. Values followed by the same superscript in the same column means no statistical difference at  $\alpha = 5\%$ .

The data in Table 1 confirm the results of previous researches that magnetic fields can increase the number and size of leaves in tomatoes [19], leaf area and number of leaves in soybeans [11], increasing the percentage of germination in *Salvia officinalis* L. and *Calendula officinalis* L.[20], wheat, corn and beets [21]. Increased leaf area by magnetic fields is thought to be due to magnetic fields playing a role in cytokinin synthesis [22].

The increase in chlorophyll content due to exposure to magnetic fields in this study is in line with previous studies on tomato plants [23], dates palm [24], and soybeans [25]. The high content of chlorophyll a, chlorophyll b and total chlorophyll is thought to be related to micro and macro nutrients during plant germination.

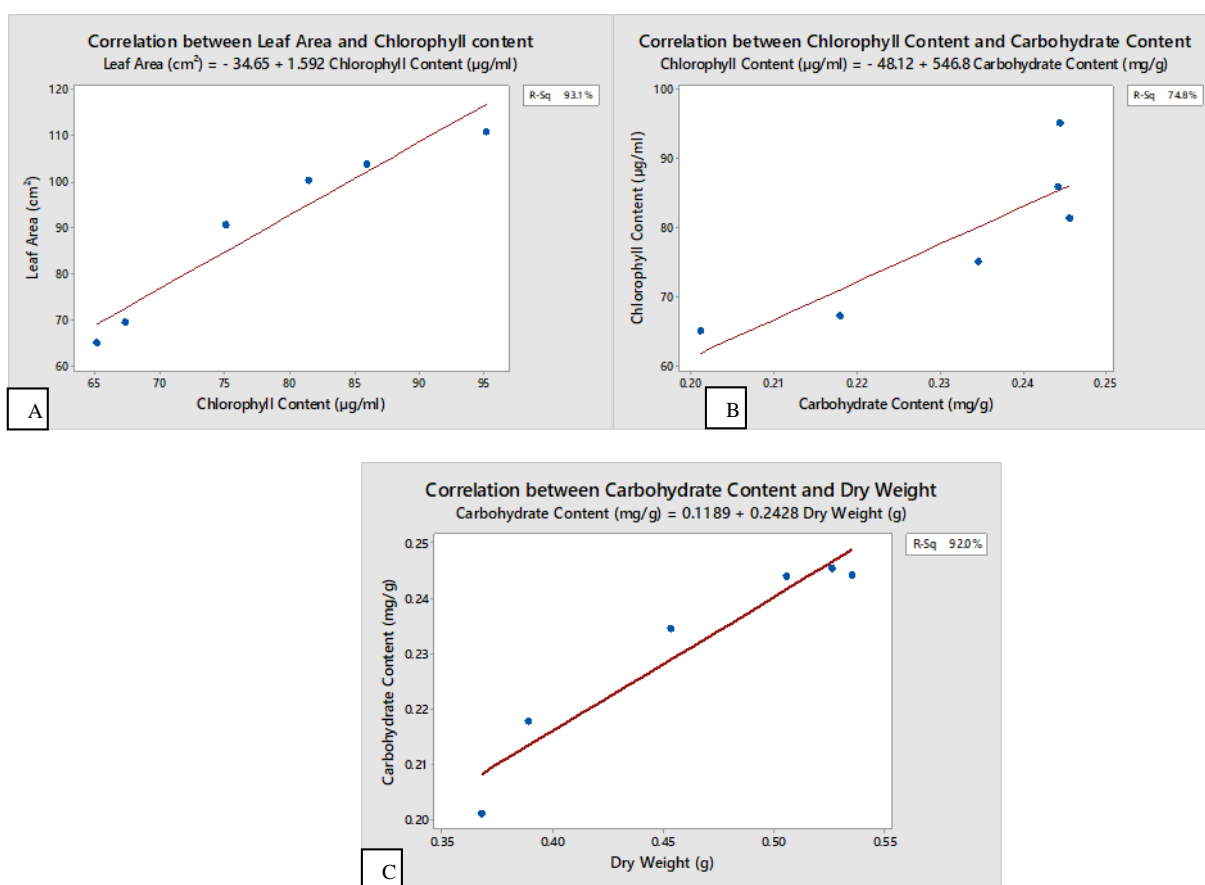
Micro and macro nutrients play an important role in supporting plant growth and production. The elements N and Fe are very important for the synthesis of chlorophyll and chloroplast protein, as well as stimulating vegetative growth. The availability of optimal N elements will increase the chlorophyll content thereby increasing the rate of photosynthesis and assimilates which are important to support the growth of more produced so that plants can grow better [26]. Another element that is no less important is P which is quite important in the formation of new cells in growing tissue, thereby increasing plant growth [27]. Other previous research findings prove that magnetic fields can increase membrane permeability and nutrient concentrations of N, K, Ca, Mg, Cu, Fe, Mn, Na and Zn in strawberry leaves [9].

*F. oxysporum* infections in seeds exposed to a magnetic field of 0.2 mT also resulted in a decrease in chlorophyll content although not significant (M<sub>7</sub>F<sub>0</sub> v.s M<sub>7</sub>F<sub>60</sub> and M<sub>15</sub>F<sub>0</sub> v.s M<sub>15</sub>F<sub>60</sub>). Low total chlorophyll content in a plant can be used as an indication that the plant has a disease. *Fusarium* infection can be through a wound in the root, into the xylem tissue following water transport, resulting in inhibition of water and nutrients to all parts of the plant especially the leaves. Inhibition of water and nutrient transport causes plants to show symptoms of wilting, leaf turn yellow, and eventually

die [28]. *Fusarium solani*, for example, is able to produce various cytotoxin compounds that are translocated to the leaves resulting in chlorosis [29]. In maize plants, low content of chlorophyll and chlorosis are the main symptoms of disease attack [30].

An insignificant reduction in chlorophyll content in plants grown from seeds that have been exposed to magnetic fields shows that an increase in plant metabolism as a result of exposure to a magnetic field of 0.2 mT is able to maintain chlorophyll content. Despite the decline, but statistically it was insignificant, even the M<sub>15</sub>F<sub>60</sub> treatment produced plants with higher chlorophyll content than control. The higher chlorophyll content in seeds that are not exposed to a magnetic field of 0.2 mT and infected with *Fusarium* (M<sub>0</sub>F<sub>60</sub>) than those who are not infected cannot be explained. Although the chlorophyll response to the treatment interaction of magnetic field exposure of 0.2 mT and *Fusarium* infection differed from the leaf area response, the results of the regression analysis between the two parameters showed a positive correlation, R<sup>2</sup> = 93% (Figure 1). This figure shows that the higher the leaf area, the higher the chlorophyll content.

This finding is in line with the results of other research studies that an increase in chlorophyll content is positively correlated with the increase in leaf area. Leaf area is an important parameter indicating close relationship between leaves and various metabolic processes including photosynthesis [31]. Chlorophyll content can be used in determining the rate of photosynthesis and is also related to carbohydrate and biomass content [32]. Chlorophyll content can also be used as an indicator of environmental stress that causes metabolic imbalances in plants [33]. Healthy plants have high chlorophyll content so that plants are able to grow maximum and are resistant to disease [23].



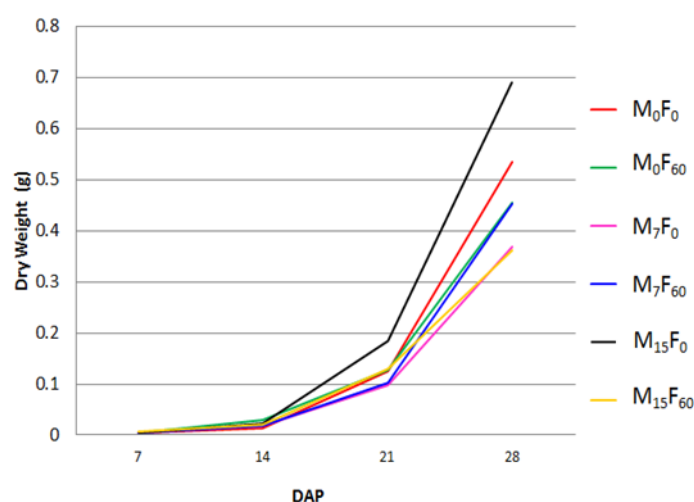
**Figure 1** Correlational analysis between parameters: (A) leaf area and chlorophyll content; (B) chlorophyll content and carbohydrate content; (C) carbohydrate content and dry weight of plant at 28 DAP.

It is suspected that there is a relationship between the high accumulation of photosynthate and the role of carbohydrates such as sucrose, fructose, and glucose as osmoprotectants in plants infected with the disease. Therefore, plants that live in stressful environments such as by pathogens will show a tolerant mechanism response to these conditions, by increasing the accumulation of photosynthate and osmoprotectant compound products [34].

Pathogen attack results in a decrease in the rate of photosynthesis due to damage to photosynthetic tissue (chlorophyll a and chlorophyll b) thus inhibiting the ability of plants to capture light [35]. Pathogen infections can also reduce the absorption of mineral nutrients for chlorophyll synthesis so that it will indirectly reduce the chlorophyll content in plants [36].

<sup>7</sup> Thus, the results of this study indicate that the 0.2 mT magnetic field in chili seeds is able to maintain carbohydrate content against *Fusarium* infection even though it causes a decrease in leaf area and chlorophyll content. Photosynthesis is a biochemical process that takes place in the leaves and with the role of chlorophyll the process of using solar energy to trigger fixation of CO<sub>2</sub> into carbohydrates as the main source for plants to increase resistance to disease [37]. Thus, plants that are resistant to disease will have a higher chlorophyll content than that of not.

Anova results on the dry weight values (Table 1) showed that the interaction between magnetic field exposure of 0.2 mT with *Fusarium* infection was significantly different in each measurement, but based on the Fishers Test at  $\alpha = 5\%$  showed treatments that produced the highest dry weight differed according to measurement time (Table 1 and Figure 2).



**Figure 2** The interaction effect between 0.2 mT magnetic field exposure and *Fusarium* infection on dry weight of chili plant at 7, 14, 21 and 28 DAP. M = magnetic field of 0,2 mT; F = *Fusarium* infection; 0 = 0 min (as control); 7 = 7 min 48 sec; 15 = 15 min 36 sec; 60 = 60 min.

In Figure 2 it is clear that the best dry weight gain was obtained from plants exposed to a 0.2 mT magnetic field for 15 minutes 36 seconds and not infected with *Fusarium* sp. (M<sub>15</sub>F<sub>0</sub>) when compared to the control (M<sub>0</sub>F<sub>0</sub>).

<sup>2</sup> These results confirm the results of previous studies that magnetic fields in tomatoes [38], wheat seeds [39], and in lettuce seeds [40] increase dry weight. Regarding data in Tabel 1 and correlational curve in Figure 1C, it is suggested that the dry weight response of the chili at 21 and 28 DAP are in correlation with the carbohydrate response against magnetic field of 0.2 mT and infection of *Fusarium* with an R<sup>2</sup>=92%. It was confirmed asumption that plant dry weight is the accumulation of photosynthate [41].

Furthermore, the data in Table 1 can also be interpreted that although *Fusarium* infection in seeds exposed to a magnetic field of 0.2 mT results in plant dry weight which is not important compared to that of the control. These results support previous research results on tomatoes which showed that exposure to a 0.2 mT magnetic field was able to produce high vigor as indicated by an increase in plant dry weight compared to control and resistance to *Fusarium* wilt disease [38]. The high dry weight of plants is also thought to be related to the increase in carbohydrate content in response to the treatment of *F.oxysporum*. This is in line with the results of research by Djukri and Purwoko[42] who argue that on taro, the high dry weight of plants correlates with an increase in carbohydrate content in response to the treatment of *Fusariumfungi* [41].



## 4. Conclusion

Exposure to a magnetic field of 0.2 mT for 15 minutes 36 seconds increased leaf area, total chlorophyll content, and dry weight of chili plants at 28 DAP but did not increase carbohydrate content. *Fusarium* infection in chili seeds tends to reduce leaf area, chlorophyll content, carbohydrate content, and increase plant dry weight, but the decrease is not significant unless the seeds are given a magnetic field exposure of 0.2 mT for 15 minutes 36 seconds (M15F0 vs M15F60), there is even *Fusarium* infection which still gives a higher yield than control (M0F0). Thus it can be concluded that the exposure of the 0.2 mT magnetic field is able to maintain the growth of chilli plants despite being infected with *Fusarium* pathogenic fungi.

## Compliance with ethical standards

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### Disclosure of conflict of interest

No conflict of interest among co-authors and between any kinds of product.

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