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Magnetic field pre-sowing treatment as an organic friendly technique to promote plant growth and chemical elements accumulation in early stages of cotton

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Abstract

The enhancement of plant growth using an environmental friendly technique is an absolutely desirable step for modern agriculture. This study reports the positive effect of pulsed electromagnetic field in early growth characteristics of cotton plants, as well as the percentage content of some important chemical elements. Pulsed electromagnetic field was used for 0, 15 and 30 minutes as a presowing treatment in cotton seeds in a pot experiment under field conditions. The experiment followed a completely randomized design, with 3 main treatments and 60 replications for each treatment in a one year study. The use of magnetic field as a pre-sowing treatment is becoming more familiar among researchers, as it is closer to the agricultural practices. In all measurements (transpiration rate, photosynthetic rate, stomatal conductance, root growth, shoot growth and N, P, K, Ca and Mg percentage), plants derived from treated seeds, performed better than control plants with statistically significant differences at the 5% level of significance. The exposure of seeds in magnetic field for 15 and 30 minutes gave statistically significant better results than control in most measurements. In chemical analysis, seeds treated for 30 minutes (MF-30) gave the highest values for all measurements, except K where seeds treated for 15 minutes (MF-15) gave the higher values in all three measurements. The investigation of plant physiology and chemical elements accumulation revealed an alternative approach for the mechanism of magnetic field enhancement.

Keywords: Magnetic field, organic, cotton, pre-sowing, physiology, roots, chemical elements. **Abbreviations:** MF-Magnetic Field; PEMF-Pulsed Electromagnetic Field; DAS-Days After Sowing.

Introduction

The new trend of the modern agriculture to more sustainable ways of agriculture, has led to the investigation of some more environmental friendly techniques. These techniques should have a low environmental impact, and the same time to contribute for the increase of yields in crops. Magnetic fields are widely used by many researchers, as they fulfill the requirements of organic agriculture. Different types of magnetic fields (Vashisth and Nagarajan, 2010; Aladjadjiyan, 2003) have been used in plant experiments, giving some interesting results (Esitken and Turan, 2004). Magnetic fields promoted the germination ratios of bean and wheat seeds and moreover the treated plants grew faster than control (Cakmak et al., 2010). Pulsed electromagnetic fields showed that could replace hormones in vegetative propagation of oregano, stimulating rooting process in stem cuttings (Bilalis et al., 2012). The influence of magnetic treatment in two pea varieties proved favorable on the emergence, growth, development and the final seed yield (Podlesny et al., 2005). Furthermore, the electromagnetic stimulation of amaranth seeds resulted in the increase of essential fatty acids and the decrease in most of the saturated fatty acids (Sujak and Dziwulska-Hunek, 2010). Influence of magnetic field in cottonseeds has not been investigated analytically. Recent studies have found that pre-sowing exposure of cotton seeds to weak sinusoidal magnetic fields led to yields as much as double those of unexposed control seeds (Leelapriya et al.,

2003). Furthermore, pulsed electromagnetic fields have been found to promote germination and improve early growth characteristics of cotton seedlings (Bilalis et al., 2012). The use of magnetic field as a pre-sowing treatment is becoming more familiar between researchers, as it is closer to the agricultural practices. In recent studies, a pre-sowing magnetic treatment of tomato seeds enhanced the growth and yield of tomatoes (De Souza et al., 2006) and germination and early growth characteristics in chickpea (Vashisth and Nagarajan, 2008). Similar are the results in beans and wheat (Cakmak et al., 2010). Pulsed magnetic field has been found to increase the plant height, fresh and dry weight, and protein content in soybeans. In addition, activity of enzymes such as b-amylase, acid phosphatase, polyphenol oxidase and catalase was enhanced (Radhakrishnan and Kumari, 2012). Pulsed electromagnetic fields have been found to have positive effects in medical science (Giannakopoulos et al., in press), as well as in agriculture (Bilalis et al., 2012). The influence of magnetic field on chemical elements has not been investigated thoroughly. Strawberry plants that were treated with magnetic field strengths of 0.096, 0.192 and 0.384 Tesla (T) in heated greenhouse conditions, showed that increasing MF strength from control to 0.384 T increased contents of N, K, Ca, Mg, Cu, Fe, Mn, Na and Zn, but reduced P and S content (Esitken and Turan, 2004). Many researchers approach the mechanism of plant growth

Table 1. Effect of magnetic field on early stages of cotton physiology. Means followed by the same letter for treatments are not significant different.

| | Transpiration Rate | | | Photosynthetic Rate | | | Stomatal Conductance | | |
|----------------|--------------------|-----------------|-----------------|---------------------|----------------|-------------|----------------------|-----------------|---------------|
| Treatment | 25 DAS | 35 DAS | 45 DAS | 25 DAS | 35 DAS | 45 DAS | 25DAS | 35DAS | 45 DAS |
| MF-0 | 3.11b | 6.17b | 5.18b | 7.95b | 10.68b | 10.72a | 0.16a | 0.18b | 0.15a |
| MF-15 | 3.78a | 8.42a | 6.83a | 9.32a | 11.97a | 11.47a | 0.17a | 0.31a | 0.19a |
| MF-30 | 3.45ab | 8.36a | 6.31ab | 9.34a | 11.71a | 11.83a | 0.16a | 0.33a | 0.17a |
| ME 0: Untroate | d soods ME 15 | · coode troated | with pulsed ala | etromagnetic fi | ald for 15 min | ME 30 soads | treated with pul | ad electromagne | tic field for |

MF-0: Untreated seeds, MF-15: seeds treated with pulsed electromagnetic field for 15 min, MF-30: seeds treated with pulsed electromagnetic field for 30 min, DAS: Days After Sowing.

Table 2. Effect of magnetic fields on early stages of cotton shoot growth. Means followed by the same letter for treatments are not significant different.

| | Shoot Fresh Weight | | | Shoot Dry Weight | | | Leaf Area | | |
|----------------|--------------------|-----------------|-----------------|------------------|----------------|------------|---------------------|-----------------|----------------|
| Treatment | 25 DAS | 35 DAS | 45 DAS | 25 DAS | 35 DAS | 45 DAS | 25 DAS | 35 DAS | 45 DAS |
| MF-0 | 1.04b | 1.74b | 2.67b | 0.21b | 0.41b | 0.76a | 1548a | 3153b | 4421b |
| MF-15 | 1.26a | 2.38a | 3.69a | 0.26a | 0.57a | 0.86a | 1699a | 4465a | 5783a |
| MF-30 | 1.25a | 2.32a | 3.58a | 0.26a | 0.58a | 0.84a | 1688a | 4431a | 5739a |
| ME O. Untracto | d soads ME 15 | . coods treated | with muland ala | atnoma anatia fi | ald for 15 min | ME 20. and | two at a d with myl | and algotromagn | atia field for |

MF-0: Untreated seeds, MF-15: seeds treated with pulsed electromagnetic field for 15 min, MF-30: seeds treated with pulsed electromagnetic field for 30 min, DAS: Days After Sowing.

enhancement using different methods. The results show a variety of magnetic field influences in many plant measurements. The aim of this study was to investigate the effect of pulsed electromagnetic fields in cotton concerning plant physiology, shoot and root growth and chemical elements in outdoor conditions.

Results

The use of pulsed electromagnetic field as a pre-sowing treatment was found to enhance cotton plants in early stages of growth. In all measurements, plants derived from treated seeds, performed better than control plants. Both duration times of exposure gave statistically significant better results than control in most measurements.

Physiological measurements

Pulsed Electromagnetic Field (PEMF) improved main physiology measurements such transpiration rate. photosynthetic rate and stomatal conductance (Table 1). The highest values in transpiration rate were measured in MF-15 treatment (3.78, 8.42 and 6.83 mmol $H_2O \text{ m}^{-2} \text{ s}^{-1}$ for 25, 35 and 45 DAS, respectively). In all cases MF-15 gave statistically significant differences compared to control plants. MF-30 gave better values than control, however differences between them were not statistically significant in all measurements. Differences between the two magnetic field treatments (MF-15, MF-30) were not statistically significant. The highest values in photosynthetic rate were measured in MF-15 and MF-30 treatments. MF-30 gave the higher values (9.34 and 11.83 $\mu mol~CO_2~m^{-2}~s^{-1},$ for 25 and 45 DAS, respectively). MF-15 gave higher values (11.97 µmol $CO_2 \text{ m}^{-2} \text{ s}^{-1}$ for 35 DAS). Differences between magnetic field treatments and control were statistically significant in the first two measurements. In stomatal conductance, magnetic field treatments gave higher values than control plants in all measurements. The differences were statistically significant only 35 DAS, where MF-15 gave 0.31 mol $m^{-2} s^{-1}$ and MF-30 gave 0.33 mol $m^{-2} s^{-1}$ (control gave 0.18 mol $m^{-2} s^{-1}$).

Shoot growth

Shoot growth characteristics gave statistically significant differences in all measurements (Table 2). The highest values in plant fresh weight were measured in MF-15 treatment (1.26, 2.38 and 3.69 g for 25, 35 and 45 DAS, respectively). MF-30 treatment gave also statistically significant higher

values than control in all measurements. Differences between MF-15 and MF-30 were not statistically significant. Plant dry weight was statistically significant higher for magnetic field treatments 25 and 35 DAS. In the first measurement, values of MF-15 and MF-30 were the same (0.26 g per plant), but in the second measurement MF-30 was slightly higher than MF-30 (0.58 g per plant). In the third measurement (45 DAS), values of magnetic field treatment were higher than control, although differences were not statistically significant. Leaf area measurements showed some statistically significant differences in the second and the third measurement. In the first measurement magnetic field treatments gave higher values, although differences were not statistically significant. The highest values in leaf area were measured in MF-15 treatment (1699, 4465 and 5783 mm² for 25, 35 and 45 DAS. respectively).

Root growth

Root growth measurements showed some statistically significant differences among treatments (Table 3). The highest values in root length were measured in MF-30 treatment (5794 and 6183 mm for 35 and 45 DAS, respectively). Both MF-30 and MF-15 gave statistically significant differences from control values in the second and the third measurement. In the first measurement magnetic field gave better values than control plants, although differences were not statistically significant. Root dry weight was higher in magnetic field treatments, although differences were statistically significant only in the second measurement. In this measurement MF-15 gave the higher value (0.44 g) followed by MF-30 (0.41 g) and MF-0 (0.21 g). In root surface measurement, a slight negative effect of magnetic field was recorded. MF-15 value (499 mm²) was lower than control value (513 mm²) only in the first measurement. However differences were not statistically significant. In the next two measurements MF-30 gave the highest values (2582 and 3246 mm² for 35 and 45 DAS, respectively).

Chemical elements accumulation in plants dry matter

Nitrogen content has been found to increase by the use of magnetic field treatment. The highest values of nitrogen percentage were measured in *MF-30* treatment (1.29, 1.44 and 1.91 % for 25, 35 and 45 DAS, respectively). In the first two measurements differences were not statistically

| | Root Length | | | Root Dry Weight | | | Root Surface | | |
|-----------|-------------|--------|--------|-----------------|--------|--------|--------------|-------|--------|
| Treatment | 25 DAS | 35 DAS | 45 DAS | 25 DAS | 35 DAS | 45 DAS | 25 | 35 | 45 DAS |
| Heatment | 25 DAS | 35 DAS | 45 DAS | 25 DAS | 33 DAS | 43 DAS | DAS | DAS | 45 DAS |
| MF-0 | 1873a | 3153b | 4427b | 0.11a | 0.21b | 0.38a | 513a | 1436b | 1785b |
| MF-15 | 2814a | 5648a | 6138a | 0.12a | 0.44a | 0.49a | 499a | 2561a | 3238a |
| MF-30 | 2631a | 5794a | 6183a | 0.13a | 0.41a | 0.53a | 554a | 2582a | 3246a |

Table 3. Effect of magnetic fields on early stages of cotton root growth. Means followed by the same letter for treatments are not significant different.

MF-0: Untreated seeds, MF-15: seeds treated with pulsed electromagnetic field for 15 min, MF-30: seeds treated with pulsed electromagnetic field for 30 min, DAS: Days After Sowing.

Table 4. Effect of magnetic field on chemical elements accumulation. Means followed by the same letter for treatments are not significant different.

| | | Nitrogen | Phosphorus | Potassium | Calcium | Magnesium |
|-------|-------|----------|------------|-----------|---------|-----------|
| 25DAS | MF-0 | 1.15a | 0.076c | 0.38b | 0.24b | 0.168b |
| | MF-15 | 1.22a | 0.095b | 0.65a | 0.27a | 0.169b |
| | MF-30 | 1.29a | 0.114a | 0.63a | 0.3a | 0.189a |
| 35DAS | MF-0 | 1.31a | 0.083b | 0.49b | 0.29a | 0.173a |
| | MF-15 | 1.32a | 0.099b | 0.71a | 0.36a | 0.186a |
| | MF-30 | 1.44a | 0.116a | 0.66a | 0.36a | 0.189a |
| 45DAS | MF-0 | 1.72b | 0.115a | 0.83a | 0.31b | 0.195b |
| | MF-15 | 1.81ab | 0.116a | 0.92a | 0.38a | 0.195b |
| | MF-30 | 1.91a | 0.125a | 0.91a | 0.39a | 0.251a |

MF-0: Untreated seeds, MF-15: seeds treated with pulsed electromagnetic field for 15 min, MF-30: seeds treated with pulsed electromagnetic field for 30 min, DAS: Days After Sowing

significant. In the third measurement nitrogen content in MF-30 treatment was statistically significant higher than control (Table 4). In phosphorus, differences between treatments were statistically significant only in the first two measurements, while in third the percentage content was slight higher in MF-30 treatment. The highest values of phosphorus percentage were measured in MF-30 treatment (0.114, 0.116 and 0.125 % for 25, 35 and 45 DAS, respectively). In the first measurement MF-30 gave the higher value (0.114%) followed by MF-15 (0.095%) and MF-0 (0.076 %) and all differences were statistically significant. In the second measurement, MF-30 value was statistically significant higher than MF-15 and MF-0 (Table 4). In potassium, MF-15 treatment gave the higher values in contrast to all other chemical measurements (0.65, 0.71 and 0.92 % for 25, 35 and 45 DAS, respectively). In the first and the second measurement, MF-15 and MF-30 values were statistically significant higher than MF-0 treatment, while in the third there were no statistically significant differences (Table 4). Calcium content was higher in MF-30 treatment (0.30, 0.36 and 0.39 % for 25, 35 and 45 DAS, respectively). Differences were statistically significant in the first and the third measurement. Differences between the two magnetic field treatments (MF-15, MF-30) were not statistically significant (Table 4). MF-30 treatment increased 28.7% magnesium content in cotton plants 45 DAS. In this measurement MF-30 gave the higher value (0.251 %) followed by MF-15 (0.195 %) and MF-0 (0.195 %). In all cases differences between control and MF-15 were not statistically significant. In the second measurement no statistically significant measurements observed (Table 4).

Discussion

The results obtained in this pot experiment showed a positive impact of pulsed electromagnetic field in plant physiology, shoot growth, root growth and chemical elements accumulation. The combination of all these measurements suggest that the influence of magnetic field can lead to a better establishment of cotton and even to induce precocity in plant development. Early growth characteristics, such as shoot fresh and dry weight, and leaf area, have been found to be promoted by electromagnetic fields. Magnetic field of 0.15 T strength on maize samples led to an increase of the shoot fresh weight by 72 % compared to the control (Aladjadjiyan, 2002) and EMF treatment by 10 mT intensity of wet treated seeds caused increase in fresh and dry biomass weight of maize (Shabrangi et al., 2010). Root growth has been positively influenced by magnetic field pre-sowing treatment in the first 45 days of plant growth. Recently Vashisth and Nagarajan (2010) found that root length and root surface area showed significant increases in sunflower seedlings exposed to static magnetic fields of strength from 0 to 250 mT. Similar results have been recorded by Muraji et al. (1998) where in corn seedlings alternating magnetic field of 10 and 20 Hz resulted in 20% greater root growth than control plants. Moreover, root length, and root surface are used as important physiological parameters for evaluation of chemical elements uptake (Wang et al., 2006). Chemical analysis showed that magnetic field has improved elements percentage. Nitrogen, phosphorus, potassium, calcium and magnesium were higher in MF treatments. According to Esitken and Turan (2004), MF has an effect on plant nutrient element uptake from growth media. Increase in the MF strength from control to 0.384 T increased concentration of N, K, Ca, Mg, Fe, Mn and Zn of strawberry plant leaves, but at the 0.384 T strength concentration of P and S decreased compared with control. The higher nutrient content of cotton plants treated with magnetic field resulted to higher rates of photosynthetic machinery. Nutrients like P, K, and Mg improve the root growth which in turn increases the intake of water which helps in stomatal regulation. Efthimiadou et al., (2010) also observed that the photosynthetic rate and stomatal conductance of the corn plants fertilized was higher than control. There is little published research on the effect of magnetic fields on photosynthesis. Only recently, Javed et al., (2011) found that pretreated corn seeds with different electromagnetic treatments particularly 100 and 150 mT for 10 min significantly alleviated the drought-induced adverse effects on growth by improving photosynthesis, transpiration rate and stomatal conductance. In a similar experiment under greenhouse conditions, it was found that MF treatment (100 mT for 2 h and 200 for 1 h) of maize seeds enhanced the photosynthesis rate (Anand et al., 2012).

Materials and methods

Plant materials

A pot experiment took place in Agricultural University of Athens, in summer 2009, under field conditions. Cottonseeds (*Gossypium hirsutum* L. cv. Campo) of mean seed moisture 8% were used. The experiment followed a completely randomized design, with 3 main treatments (*MF-0, MF-15* and *MF-30*) and 60 replications for each treatment.

Device of exposure

Cotton seeds were treated by Papimi electromagnetic field generator for 15 or 30 minutes before planting. Papimi device is a pulsed EMF generator (PAPIMI model 600, Pulse Dynamics, Athens, Greece. Manufacturer characteristics: 35-80 J/pulse energy, 1 X 10^{-6} s wave duration, 35-80 X 10^{6} W wave power, amplitude on the order of 12.5 mT, rise time 0.1 ms, fall time 10 ms, repetitive frequency of 3 Hz.). The same device has been used in medical and agricultural studies (Giannakopoulos et al., in press; Athanasiou et al., 2007; Bilalis et al., 2012; Milgram et al., 2004). *Measurements*

All measurements took place 25, 35 and 45 days after sowing (DAS) and were destructive, except physiological measurements that were non destructive. Shoot fresh weight was measured by a precision balance and then the samples were oven dried at 70° C for three days in order to measure the dry weight in grams per plant. Leaf Area was measured by using an automatic leaf area meter (Delta-T Devices Ltd). Before measuring root characteristics, each root sample was separated from the soil by washing the samples over a 5 mm mesh sieve, as regard pot experiment. A formalin/acetic acid/alcohol (FAA) staining solution was used. The determination of root length and root surface was made by a high-resolution scanner, using DT software (Delta-T Scan version 2.04; Delta-T Devices Ltd, Burrwell, Cambridge, UK) (Kokko et al. 1993). Root dry weight was measured using the same procedure like plant samples. Measurements of photosynthetic rate (μ mol $CO_2m^{-2} s^{-1}$), transpiration rate (mmol H₂O m⁻² s⁻¹) and stomatal conductance (mol m⁻² s⁻¹) were undertaken between the hours of 10.30 and 14.30 on fully expanded leaves, with five measurements per treatment. Measurements were made using an LCi Leaf Chamber Analysis System (ADC, Bioscientific, Hoddesdon, UK).Samples were oven dried at 70° C for three days and analyzed for N, P, K, Ca and Mg using Association of Official Analytical Chemists method (AOAC, 1995). Nitrogen was determined by the Kjeldahl method. Phosphorus was determined by a spectrophotometer. Potassium was determined by a flame spectrophotometer. Calcium and Magnesium were determined by atomic absorption spectrometry.

Statistical analysis

The experimental data were analyzed using the software Statistica (StatSoft, 1996), according to the completely randomized design. Analysis of variance (ANOVA) and comparisons of means were calculated using the least significant difference (*LSD*) test, at the 5% level of significance.

Conclusion

Our results indicate that the application of magnetic field as a pre-sowing treatment can improve plant growth measurements and the accumulation of the main chemical elements. Moreover, this technique is organic friendly, an attribute highly desirable in modern agriculture. Magnetic field treatment enhanced all measurements (transpiration rate, photosynthetic rate, stomatal conductance, root growth, shoot growth and N, P, K, Ca and Mg percentage) in early stages of cotton plants.

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