Effect of pre-sowing electromagnetic treatment on seed germination and seedling growth in maize (*Zea mays* L.)

Efecto pre-siembra del tratamiento electromagnético sobre la germinación de semillas y el crecimiento de plántulas de maíz (*Zea mays* L.)

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ABSTRACT

The application of magnetic fields appears to produce changes in some physiological processes of plants, including encouraging their development. This study reports the effects of different intensities of electromagnetic fields generated by a solenoid with a variable horizontal magnetic field on the germination and growth of maize seeds. We worked with the experimental seed hybrid San Jose, which was exposed to electromagnetic fields of 0, 2, 4 and 6 mT for 3 min, using a completely randomized design with two seed conditions: imbibed and non-imbibed. Analysis of variance was performed for the variables germination and growth, with a level of significance of 95%; increases were observed in germination, vigor index I, vigor index II and root length, 3, 20, 34 and 23% respectively, compared to the control seeds (untreated) on the eighth day. Similarly, the presence of chlorophyll pigments and carotenoids was assessed and an increase was found in the concentration of chlorophyll a, chlorophyll b and carotene in corn seedlings grown from electromagnetically treated seeds. The response varied depending on the magnetic induction, without any particular trend, the best treatment was 4 μT for 3 min of exposure. The improvement of the evaluated functional variables suggests that the seeds may perform better with an electromagnetic field treatment.

Key words: germination rate, vigor, magnetic induction, physiology.

RESUMEN

La aplicación de campos magnéticos, parece producir modificaciones de algunos procesos fisiológicos de las plantas entre ellos estimular su desarrollo. Este estudio reporta el efecto de diferentes intensidades de campo electromagnético generadas por un solenoide con campo magnético horizontal variable sobre la germinación y el crecimiento de semillas de maíz. Se trabajó con semillas del híbrido experimental San José, las cuales se expusieron a un campo electromagnético de 0, 2, 4 y 6 mT, durante 3 min, bajo un diseño experimental completamente al azar en dos condiciones de semillas: imbibidas y sin embeber. Se realizó el análisis de varianza en las variables de germinación y crecimiento, con un nivel de significancia de 95%, y se observó un incremento en la germinación, el índice de vigor I, el índice de vigor II y la longitud de las raíces, de 3, 20, 34 y 23%, respectivamente, en comparación con las semillas testigo (sin tratar) al octavo día. De igual forma, se evaluó la presencia de pigmentos clorofílicos y carotenos, encontrándose un incremento en la concentración de clorofila a, clorofila b y carotenos en las plántulas de maíz obtenidas a partir de semillas tratadas electromagnéticamente. La respuesta varió en dependencia de la inducción magnética, sin ninguna tendencia particular; el mejor tratamiento fue el de 4μT, durante 3 min de exposición. El mejoramiento de las variables funcionales evaluadas sugiere que las semillas pueden funcionar mejor bajo la acción de un campo electromagnético.

Palabras clave: velocidad de germinación, vigor, inducción magnética, fisiología.

Introduction

Corn is grown in Latin America and constitutes, together with beans, a major staple in Central America. In this region, the FAO has indicated a shortfall in production areas and a need that must be addressed: the increase of maize production, which demands great efforts, but it is one of the few crops with commercial seed production (Laynez-Garsaball *et al*., 2007).

The growing need for organic products, along with the increase of plant materials for food production, has led scientists to search for systemic factors to increase production, considering not only crop development by traditional methods, such as the use of fertilizers and agrochemicals which to some extent cause environmental damage, but also with the use of physical methods, such as the use of lasers, ultraviolet radiation, electromagnetic fields and electricity (Moon and Chung, 2000; Galland and Pazur,

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2005; Hernández *et al*., 2006; Hernández *et al*., 2009; Domínguez *et al*., 2010).

These physical methods are safe for the environment and frequently change the course of some physiological and biochemical processes in the seeds, which translates into increased vigor and improved plant development at later stages (Carbonell *et al*., 2000; García *et al*., 2002).

The effects of low frequency electromagnetic fields on plant growth are not yet clear and may be different depending on magnetic frequency, shape and intensity of the waves or the biological species in question. For example, many studies have explored the influence of exposure to sinusoidal magnetic fields 50/60 Hz on organisms (Smith *et al*., 1993; Yano *et al*., 2004; Dattilo *et al*., 2005). The effect of different exposure times (Podleśny, 2005) and flux densities have also been evaluated (Carbonell *et al*., 2000; Flórez *et al*., 2007; Vashisth and Nagarajan, 2008).

Today, it is known that the application of magnetic fields of extremely low frequencies positively affects some characteristics of plants and processes, such as seed germination, shoot development, plant length, fresh weight, fruit production and mean fruit weight (Danilov *et al*., 1994; Namba *et al*., 1995; Aladjadjıyan, 2002; Esitken, 2003; Rochalska and Orzeszko-Rywka, 2005; De Souza *et al*., 2006; Nimmi and Madhu, 2009; Cakmak *et al*., 2010).

The positive effects of magnetic fields have also been shown in the biosynthesis of proteins, cell production, photochemical activity, respiration rate, enzyme activity and nucleic acid content (Lebedev *et al*., 1975; Phirke *et al*., 1996; Levin and Ernst, 1997; Stange *et al*., 2002).

In maize, a decrease in the physiological potential of seeds has been observed due to natural aging, which gradually decreases the germination capacity and rate of early seedling growth and tolerance to adverse conditions (Gutíerrez-Hernández *et al*., 2007). However, the use of electromagnetic fields can significantly counteract this phenomenon, because its effect on seeds can change the course of some physiological processes and stimulate plant development (Phirke *et al*., 1996; Pietruszewski, 1999; Pittman, 1977; Podleśny *et al*., 2005).

Therefore, when the rational use of agricultural production space is promoted, pre-treatment of seeds with physical factors becomes more important (Phirke *et al*., 1996; Pietruszewski, 1993; Podlesny, 2001).

For these reasons, this study was designed to determine the influence of pre-sowing electromagnetic seed treatment on the corn hybrid "San Jose" considering emergence, growth and development of plants under germination tests.

Materials and methods

Maize seeds of (*Zea mays* L.) the San Jose variety were used, from the Department of Fitotecnia, Universidad Autónoma Chapingo, they were in good phytosanitary conditions, without apparent defects, insect damage or malformations. The seeds were homogenized by size, with an average weight of 0.289 g/seed, as measured with an analytical balance, Adventure (OHAUS).

In March and April 2010, the germination tests were carried out at the Laboratories of CINVESTAV-IPN and ESIME-IPN, Mexico DF. There were two conditions for the seeds before receiving the pre-germination magnetic field treatment: a first group of 100 seeds, with three replicates, was soaked in distilled water at a rate of 1 mL of water / seed for 24 h at a temperature of 23±2°C; a second group of 100 seeds, with three replicates, which did not receive imbibition.

To apply the electromagnetic treatment, the study used a 40 cm long magnetizer solenoid which consisted of a 16 gauge copper wire (1.1 mm), with a total of 363 turns. The intensity of the magnetic field was horizontally variable, changing from the ends toward the center.

The values of the magnetic induction (B) applied to the seeds were B_1 = 2, B_2 = 4 and B_3 = 6 μT, during a fixed exposure time of 3 min (Tab. 1). The samples were placed randomly in a dielectric container and placed within the magnetic field at the ends and center of the solenoid, in a homogeneous space.

TABLE 1. Electromagnetic treatments applied to the corn seed for a constant time.

Imbibition	Control	B_1	B_2	B_3
Seeds without imbition	0	2 μT*	4 μT	6 μT
Seeds with imbition	0	2 μT	4 μT	6 μT

* Magnetic induction (B), t = seed exposure time, 3min

The flux density was measured with a digital gaussmeter (Magnet-Physics, Dutch, model FH 54) in units Tesla (T) ($1T = 1\ V\ s\ m^{-2} = 1\ kg\ s^{-2}\ A^{-1}$).

Germination tests

For germination of the maize seeds, three replicates of 100 seeds each were placed on moistened filter paper in Petri dishes (11.0 x 1.5 cm), then placed in an incubation chamber at 20°C.

After 8 d, the percentage of germination was evaluated. The 25 seeds were randomly selected from each replicate per treatment, to assess the length of the root and coleoptile.

The seed vigor was calculated following the formula of Abdul-Baki and Anderson (1973) (equation 1).

Vigor index I = germination% x shoot length (radicle and coleoptile) (1)

To calculate the speed of germination (X), the number of germinated seeds was counted every day until day 8, when there were no more germinated seeds (equation 2).

X = No. Germinated seeds/1st day of the count + ... + No. Germinated seeds/last day of count (2)

Growth tests

Two replicates were set up with 30 plants for each treatment, which were germinated on paper and subsequently planted in plastic containers with a depth of 9 cm containing 100 g of compost (mixture 1: 1: 1 v/v organic soil, humus, perlite sand, and agricultural lime).

At 15 d, 10 seedlings from each replicate were dried for 4 h in an oven at 104°C, and the dry weight of the shoots was determined. With these values, we calculated the vigor index II, according to equation 3.

Vigor Index II = germination% x shoot dry weight (root and stem) (3)

Determination of chlorophyll pigments concentration

All the leaves were taken from 10 seedlings to obtain chlorophyll extract in each of the treatments, using 70% alcohol, the absorbance of the extracts was measured at 663, 645 and 472 nm with a UV-Vis spectrophotometer (Perkin Elmer, mod. Lambda 25), according to Meyer-Berthenrath's modified method (equation 4, 5 and 6).

$$chlA - \frac{12.3E(663) - 0.86E(645)}{1000dw} v \tag{4}$$

$$chlB - \frac{19.3E(645) - 3.6E(663)}{1000dw} v \tag{5}$$

$$t.c - \frac{10E(472)}{2485dw} v \tag{6}$$

where:

- chlA / B, content of chlorophyll A and B (mg g^{-1});
- t.c, total content of carotenoids (mg g^{-1});
- E (l), wavelength; d, cell width (1 cm);
- V, volume 70% ethanol extract (mL); w, tissue weight (g)

Statistical Analysis

The study used a completely randomized design and the results were analyzed with SPSS 13.0, comparing the values obtained for each of the treatments. A simple classification ANOVA was conducted (One-Way ANOVA), considering results obtained with confidence levels of 95% as significant, corresponding to significant differences ($P \leq 0.05$) and the differences between treatments that were significant were calculated using the DMS test (Least Significant Difference).

Results and discussion

Analysis of variance of the results showed that for seeds without imbibition there were no significant differences between treatments and the control for the variables tested, except for the 2 μT treatment, with inferior results compared to the control, while for the seeds with imbibition, inferior results were obtained with significant differences compared to the control, for treatments 4 and 6 μT concerning germination speed.

However, there was an improvement in the seeds with imbibition over the control treatment of 3% in germination percentage for the inductions 2 μT and 4 μT, without significant differences compared to the control (Tab. 2), with a more uniform growth after sowing in the seeds treated with the magnetic field in comparison with the control.

By analyzing the variables for the growth of maize seedlings, in seeds generated with imbibition, an increase was observed in the length of the roots, the total length of the shoots and vigor indexes I and II, with significant differences from the control for treatments 4 and 6 μT.

Similarly, the shoot dry weight increased in the 4 μT treatment, with respect to the control, there was an increase from 23 to 30% in the length of roots and in the vigor indexes I and II, 20 and 34%, respectively.

The increase in physiological activity due to increased moisture absorption in the treated seeds could be responsible for a greater length of shoots and vigor indexes.

TABLE 2. Effects of pre-germination exposure to different flux densities of a low frequency electromagnetic field on corn seed.

Treatment	Germination (%)	Speed of germination	Dry weight (g)	Shoot length			Vigor Index	
				Coleoptile	Radicle	Total	I	II
SSI								
Control	78	38.17	0.96	4.00	5.76	9.77	762.21	61.53
2 μT	56*	29.02*	0.62*	4.13 NS	4.16*	8.29*	464.35*	14.84*
4 μT	69 NS	34.43 NS	1.38 NS	4.08 NS	4.12*	8.20*	566.90*	77.33 NS
6 μT	72 NS	38.12 NS	0.38*	4.05 NS	5.06 NS	9.10 NS	655.48*	11.85*
ES (5%)	3.04	2.07	0.65	0.23	0.88	0.86	11.26	5.74
SEI								
Control	72	41.13	0.41	4.26	6.28	10.54	759.17	12.80
2 μT	74 NS	40.97 NS	0.39 NS	4.52 NS	6.97 NS	10.88 NS	805.71 NS	11.64 NS
4 μT	75 NS	36.42*	0.58*	4.47 NS	7.76*	12.22*	916.80*	17.26*
6 μT	66*	34.05*	0.33 NS	4.86*	8.22*	13.07*	863.02*	7.80*
ES (5%)	2.00	1.86	0.32	0.49	0.99	1.08	8.27	1.97

SSI, seeds without imbibition, SEI, seeds with imbibition.
* Significant differences $P \leq 0.05$. NS, not significant.

The seeds without imbibition yielded only an improvement in vigor index II, otherwise there were no significant differences with respect to the control.

These results agree with those shown by Penuelas *et al.* (2004), who examined diamagnetic susceptibility such as the response of root growth when exposed to magnetic fields of two legumes (*Lens culnaris* and *Glycine soja*) and one cereal (*Triticum aestivum*), and observed a reduction in root growth.

Results similar to those found in this investigation were reported by Fischer *et al.* (2004), who showed that high intensity magnetic fields had no effect on the germination of wheat seeds, but increased the fresh weight of roots and shoots.

This may be related to the issues raised by Rajendra *et al.* (2005), who observed a significant increase in the mitotic index, and the incorporation of ^{3}H-tymidina within the DNA in seeds of *Vicia faba* that were exposed to a 100 μT electromagnetic field.

The results also coincide with those reported by Vashisth and Nagarajan (2008), for chickpea seeds (*Cicer arietnum*) treated with an intensity of 50 mT.

Water uptake by the seed is directly influenced by the presence of the seed coat and the permeability thereof. The reserve tissue absorbs water at a medium speed, until complete hydration (Moreno *et al.*, 2006).

Oguntunde and Adebawo (1989) determined the individual patterns of water uptake of maize seeds, soaked for 0 and 72 h. The results obtained showed that most water absorption of the seeds occurred within the first 24 h.

Similarly, Paiva *et al.* (2006) indicated that during imbibition of *Swietenia macrophylla* seeds, the movement of water within the seed is due to the action of diffusion and capillarity.

There are no reliable hypotheses about the behavior mechanisms of living things subjected to magnetic fields. Magnetic fields penetrate tissues, organelles and cell membranes, which have different electromagnetic properties, and act on biochemical reactions and more than a couple of unpaired electrons (Reiter 1993; Atak *et al.*, 2007).

However, Reina *et al.* (2001) reported in experiments in lettuce seeds, that exposure to static magnetic fields alters water absorption in seeds and explained the changes in germination and growth rates, and that there was a close relationship between the theoretical calculations and the experimental results.

These authors state that the magnetic field interacts with the ionic currents in the cell membrane by changing the conductivity of the membrane and thus the concentration and osmotic pressure on both sides of the membrane and that these changes alter the mechanisms of imbibition of seeds.

Furthermore, Podlesny *et al.* (2005) reported a positive effect of magnetic fields (30-100 mT) on germination and emergence in two cultivars of the pea (*Pisum sativum*), while Martinez *et al.* (2002) observed that the magnetic field increased the length and weight of barley seeds (*Hordeum vulgare*) and the extent of this effect is a function of the duration of exposure.

Flórez *et al.* (2007) found that the accumulation of the dry weight of 10-day-old corn shoots exposed to 125 mT increased logarithmically with the field strength. In their studies, the seeds had imbibition in water before exposure to magnetic fields.

Others have suggested that magnetic treatment does not affect the percentage of emergence in corn seed, even though there is an increased rate of emergence of the seeds (Carbonell *et al*., 2000; Flórez *et al*., 2007; Vashisth and Nagarajan, 2008).

In analyzing the results of the concentration of chlorophylls and carotenoids, significant differences were found for the concentration of chlorophyll a and b, with respect to the control, in the case of seeds without imbibition, the highest concentration occurred in the 4 μT treatment, followed by treatments 6 and 2 μT. In the case of seeds with imbibition, significant differences with respect to the control were found, with the best treatment being 6 μT, followed by 4 and 2μT (Fig. 1).

By analyzing the concentration of carotenoids, no significant differences were found with respect to the control in the seeds with imbibition, but for the other treatments there were, although in both cases the results were higher in the treated seeds (Fig. 2).

Ursache *et al*. (2009) obtained an increase in the concentration of chlorophyll pigments in corn plants treated with

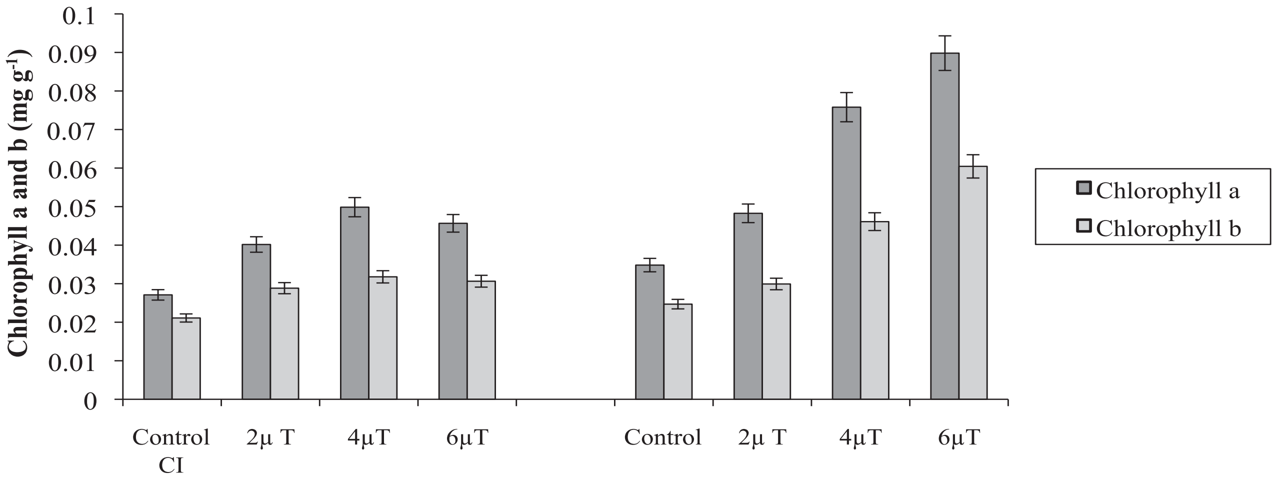

FIGURE 1. Effect of electromagnetic field on the concentration of chlorophyll a and b, in seedlings grown from maize seeds with imbibition (CI) and without imbibition (SI) (95% significance level). The bars on the columns indicate standard error.

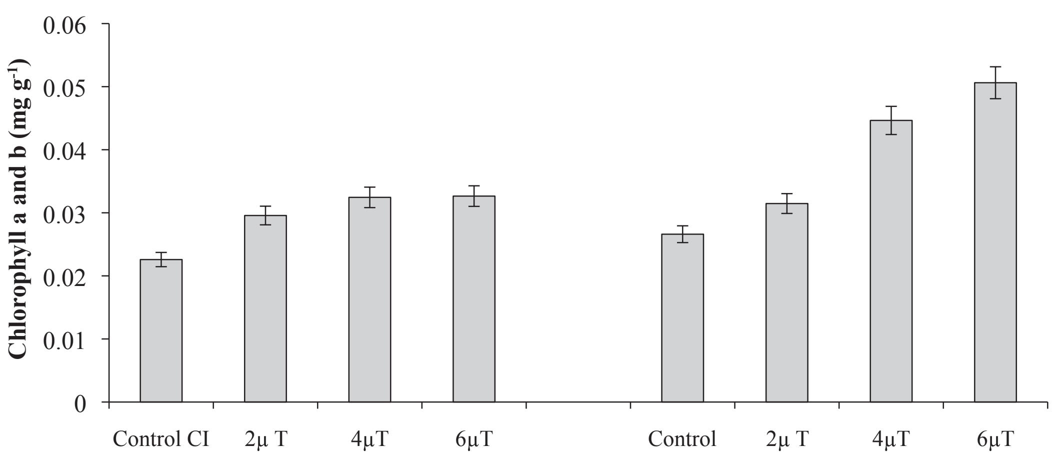

FIGURE 2. Effect of electromagnetic field on the concentration of carotenoids in seedlings grown from maize seeds with imbibition (CI) and without imbibition (SI). The bars on the columns indicate standard error.

high-frequency electromagnetic waves without significant differences with respect to the control, which is consistent with the results obtained in this study.

There have been numerous hypotheses to explain, from the physical standpoint, the cellular changes of biological organisms caused by exposure to magnetic fields, however, one should consider that the cyclotron resonance and paramagnetic resonance, which are the main effects of electromagnetic fields, are probably due to the alteration of the membrane associated with the transport of the calcium ion (Galland and Pazur, 2005).

The search for other methods, cleaning techniques, and non-contaminates to help increase the survival rate of seedlings, seed quality and yield of crops, has established relationships with other sciences, as in the case of physics, within which electromagnetics has played an important role. This has become one of the current alternatives to stimulate plant growth and increase the physiological and sanitary quality associated with better agronomic performance. The data from this research shows that electromagnetic fields could become a useful tool for better plant growth in the early stages of development.

Conclusions

• In the germination process of *Zea mays,* the best results were obtained from electromagnetic induction at the 4 μT level at 3 min of exposure.

• Seeds with imbibition exposed to magnetic treatment, showed an improvement of 2-3% in the germination percentage and a 23-30% increase in root length and increases in vigor indexes I and II of 20 and 34%, respectively.

• In maize seeds that only received pre-germination magnetic treatment, there was an increase of fresh weight and dry weight of shoots of 39 and 43% respectively, as compared to the control.

• An increase in the concentration of chlorophyll a, chlorophyll b and carotene in corn seedlings obtained from seeds treated electromagnetically was recorded.

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