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ENHANCEMENT OF GERMINATION IN SOYBEAN (*Glycine max* L. Merrill) SEEDS BY PRE-TREATMENT OF SEEDS WITH MAGNETIC FIELD

Gizem Oren Cibik[1], and Fehime Sevil Yalçın[2*]

[1] Çanakkale Onsekiz Mart University, Graduate School of Natural and Applied Sciences, Biology; Canakkale, Turkey
https://orcid.org/0000-0002-3308-6125
[2*] Çanakkale Onsekiz Mart University, Faculty of Education, Science Education, Biology Education; Canakkale, Turkey
https://orcid.org/0000-0003-0661-6431
* Corresponding author: sevilyalcin@comu.edu.tr

ABSTRACT

The effects of magnetic field (MF) on the germination percentage of soybean seeds and the development of seedlings were investigated in this study. Static MFs of 0, 0.5, 1.0 and 5.0 mT, generated by a coil, were applied to seeds. Sterilized seeds in plastic bags were exposed to the MF flux densities for one hour per day over four days. After MF application, seedlings were potted in soil and germinated under greenhouse conditions. The germination experiment was conducted over a 72 hour-period. Germination percentages increased in the 0.5, 1.0, and 5.0 mT treatments, compared to the control group. The length and fresh weight of seedlings exposed to MF were greater than those of the control group (p<0.005), but there was no significant difference in mean dry weights between the experimental and control groups (p>0.005).

Keywords: Soybean, germination percentage, magnetic field, seedling height, dry weight, fresh weight.

INTRODUCTION

Soybean (*Glycine max* L. Merrill) is an annual plant belonging to the Fabales order and Fabaceae family. Soybean seeds contain 18-24% oil, 35-40% proteins, 30% carbohydrates, and 5% minerals. In addition to its use in dietetics and medicine fields, soybean is one of few plants that are also used in the production of over 400 industrial products, including glue, ink, soap, gasoline, insecticide, alcohol, plastics, and rubber (Rahman, 2021; Ahmed et al., 2023; Mishra et al., 2024).

The Earth's magnetic field (MF) is a global vector field, also called the geomagnetic field. Its strength varies naturally, ranging from about 25 and 65 μT (microTesla). All living systems live under the Earth's magnetic influence (Erdman et al., 2021). The MF has a non-ionizing effect at the molecular level (Dhawi, 2014). Studies at the cellular level have shown that RNA and protein synthesis on G_1 phase are influenced by changes in MF flux density and increases in cell division rate for cells exposed to the MF (Dhawi, 2014; Gorobets et al., 2023). The biological effects of MFs depend on energy level, exposure time, distance of target from source and structure of organism (Krylov and Osipova, 2023). However, it is difficult to evaluate the effects on regular functions when plant biological systems are exposed to MFs, primarily due to the complexity of these systems. In this sense, the biological effects of MFs have been the subject of more extensive studies, as they can penetrate deeper into tissues.

The mechanism of the MF used treats the crop

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seeds through a relatively complicated process that is not entirely understood. Many studies have described the positive impact of MF on the internal enzyme activity of seeds and nutrient utilization inside plant seeds (Dhawi, 2014; Bai et al., 2015; Sarraf et al., 2020; Hafeez et al., 2023). In recent years, many researchers around the world have begun investigating the positive effects of MFs on living organisms, as well as their negative effects (Yang et al., 2020; Levitt et al. 2022). Magnetic stimulation is an interesting alternative to chemical methods for increasing the performance and capability of ecological and economical plants. These biophysical methods are not only more profitable but also environmentally friendly for the future of agriculture (Konefał-Janocha et al., 2019).

Early research on the MF treatment of seeds reports that MFs lead to an acceleration of plant growth, protein biosynthesis, and root development (Maffei, 2014). Dziergowska et al. (2021) observed that an application of an MF of 250 mT for 3 min positively affected the germination speed of soybean seeds, as well as seedling length and fresh weight. Bai et al. (2015) reported that mung bean seeds treated by an electric field of 25 kV and an MF of 0.6 T increased germination percentages. Other studies have reported on the positive effects of MF treatments on plant characteristics such as seed germination (Konefał-Janocha et al., 2019), root length (Muhova et al, 2016), seedling growth (Singh and Agrawal, 2020), fresh weight, and dry weight (Podleśna et al., 2019).

When a plant interacts with an MF, its growth and metabolism may be affected. However, the factors of the application process, such as MF flux density and duration, are key factors in observing any impact on the development of plants. Therefore, the experimental designs presented in this study will help support the understanding of the impact of MF. In this regard, the present study aims to understand how MF treatment influences seed germination, seedling growth, fresh weight, and dry weight of the Arısoy variety of soybean (*G. max* L. Merrill) seeds, and to provide an overview of the potential basic biological reasons for the observed growth patterns.

MATERIAL AND METHODS

Preparation of plant material

Soybean (*G. max* L. Merril) seeds of the Arısoy variety were used as plant material. The seed were obtained from the Black Sea Agricultural Research Institute (Black Sea, Turkey).

Seeds were placed in an oven at 130 °C for 1 hour (Ashtiani et al., 2014) and weighed before and after the drying process. Moisture content was measured using the following formula:

$$\text{Moisture content} = W_1\text{-}W_2/W_2\text{x}100\%$$

where W_1 is seed weight before drying and W_2 is seed weight after drying (Cetin and Sevik, 2016). The moisture content of the seeds was determined as 13.1% at the beginning of the experiment.

Magnetic field generation and seed treatment

Magnetic field system

An MF system consists of a coil forming an electromagnet mounted on a glass frame. To create a uniform and constant MF, a glass cylinder with a diameter of 19 cm was used. On this cylinder, 196 rings of copper wire with a diameter of 1.2 mm were rolled, creating a resistance of about 2 ohms. Bases with a height of 2 cm were used for the coil to allow air transport and enable the temperature inside the chamber to reach equilibrium with the environment (Fig. 1).

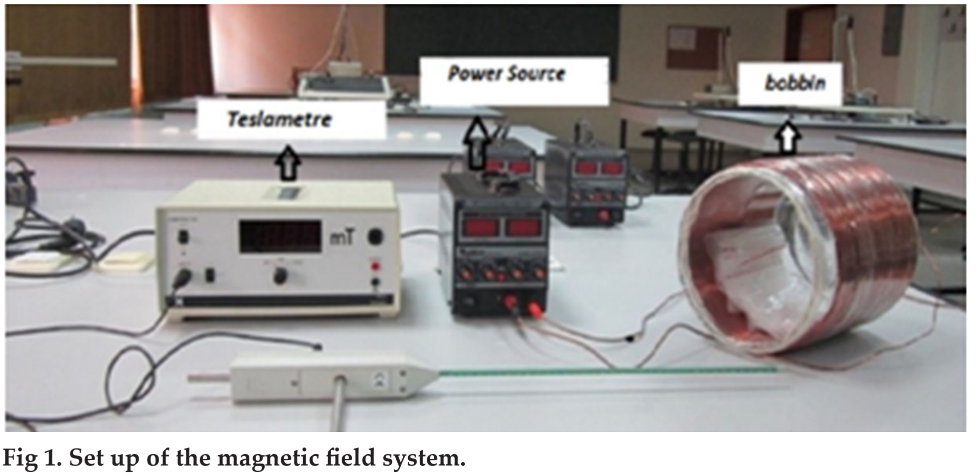

Fig 1. Set up of the magnetic field system.

In this system, it is assumed that the flux density, occurring at a suitable distance within the coil source (parallel to each other), can vary slightly, making the flux density homogeneous. The coil had a hole in its geometrical center in order to gauge flux density levels using a Teslameter. The mean static MF in the center of the coil ranged from 0.5 to 5.0 mT. The coil, which was placed horizontally, was connected to a power supply (DC 0-30 volts 0-6 A, CY-306). Furthermore, flux density through the coils was measured by an ampermeter. The accuracy and uniformity of the magnetic flux density were detected by a digital Teslameter. During the study, the ambient temperature remained the same for both the control and exposed samples (24±0.5 °C).

Seed treatment

The coil was placed horizontally and connected to a power source. Calibration of the fixed-range MF flux density in the coil was determined by a Teslameter in a centroid area (0.5-5.0 mT), approximately 2 cm above the ground. Soybean seeds in plastic bags were placed in this area on a block. An MF generated by a coil of three flux density levels (0.5, 1.0 and 5.0 mT) was applied to the seeds. Table 1 indicates the generated flux density level, current, and voltage range in the coil system.

After being treated with 0.5% hypochlorite for 20 min at the end of the first MF application, the seeds were washed with distilled water three times, and seed surface was sterilized (Ranjan et al., 2017). The main purpose of treating seeds with a 0.5% hypochlorite solution is to perform surface sterilization by disinfecting the seed surface and eliminating microorganisms and pathogens. This process ensures that the seeds germinate in a sterile environment, particularly in laboratory germination experiments or cultivation of sensitive plant species (Quynh et al., 2022).

In the present study, 100 seeds were used for each MF application. MF flux densities (0, 0.5, 1, 5 mT) were applied to the seeds 1 hour per day for 4 days. After the treatment, the seeds were germinated in a drying oven at 25°C for 4 days in plastic bags on moist filter paper. At the end of four days, the germinated seeds, exposed to different MF flux densities, were sown in four replications according to a randomized block experimental design in test boxes containing 30% peat, 30% leaf rot, 30% pine needles, and 10% perlite soil. The seeds were irrigated with equal volumes of water when needed.

Oren (2016) found that for *G. max* (L.) Merrill, MF application with 0.5 – 5.0 mT magnetic flux for 60 min resulted in the highest percentage of seed germination and root length. Therefore, magnetic flux densities of 0.5, 1.0 and 5.0 mT and 60 min were preferred in this study. All treatments were carried out in the Magnetic Field Laboratory, Faculty of Science and Arts, Department of Physics, Canakkale Onsekiz Mart University. Temperature and humidity were controlled inside the laboratory.

The germination percentage of seeds were determined at 24, 48, and 72 hours after MF application. The germination percentage was calculated using the following formula:

$$GP = SNG/SNO \times 100$$

where GP is the germination percentage, SNG is the number of germinated seeds, and SNO is the number of experimental seeds with viability (Nafees, 2019).

After four days, the germinated seeds were planted in pots containing experimental soil. The experiments were conducted in a randomized design, and each treatment was replicated six times.

Seedlings were grown under controlled greenhouse conditions. After 14 days, the seedlings were uprooted and fresh weight (g) was measured. The uprooted seedlings were then placed into paper bags and subsequently put into an oven at 60 °C for 72 h to determine dry weight (g) (Hiama et al., 2019). Measurements of seedling length (SL) in ± 0.01 cm, fresh weight (FW), and dry weight (WD) in ± 0.01 mg were taken in the control and MF-treated groups at 14 days (Kataria et al., 2017).

Statistical analysis

The germination percentages of seedlings under the three MF conditions in the control group were compared using chi-square tests. The

Table 1. Current and voltage range applied from the power supply.

Flux density (mT)	Voltage (V), volt	Current (A), ampere
0.5	0.70	0.33
1.0	1.30	0.63
5.0	6.90	3.36

effects of the different MF treatments within the control group were tested using the general linear model. Differences between mean values of each measurement (SL, FW, and WD) were tested post hoc using Bonferroni's modified LSD multiple range test (p<0.05). The R in RStudio (4.0) was used in all of the statistical analysis and visual plots. All the analyses were conducted in R (R Core Team, 2020) and RStudio (RStudio Team, 2020).

RESULTS

A remarkable difference was found at 24 hours after MF application between the treated group (seeds exposed to 1.0 mT and 5.0 mT MF) and the control group (Table 2).

The germination of soybean seeds in the 1.0 and 5.0 mT treated groups started earlier compared to non-treated control seeds. At 48 hours, the results showed that the magnetic flux density of 5.0 mT caused an increase in germination percentage with respect to the control group. At 72 hours, all the magnetic flux densities (0.5 mT, 1.0 mT, and 5.0 mT) resulted in higher germination percentages compared to the control. However, these increases were not statistically significant (Fig. 2).

The applied MF densities of 0.5, 1.0, and 5.0 mT significantly increased seedling length by 15.2, 15.7%, and 16.7%; as well as seedling fresh weight by 18.5, 14.5, and 15.8% , respectively, compared to control groups (Table 3; Fig. 3).

As seen in Fig. 4, a wide range of the seedling length (9.5–39.0 cm) was found in the control group when compared to the groups exposed to 0.5, 1.0, and 5.0 mT, with values of 15.5-40.0, 14.0-41.2, and 14.0-40.0 cm, respectively. Similar distribution patterns were obtained for FWs, with values of 2.4 in the control group, and 2.1, 2.2, and 2.2 in the groups exposed to 0.5, 1.0, and 5.0 mT, respectively.

The mean SLs and FWs of the three treatments and control groups were different and these differences were statistically significant (F=8.91; df=3; p<0.001). The mean FWs of the three treatments and control groups were also differ and these differences were statistically significant (F = 4.85; df = 3; p<0.05). However, the differences among mean DW of four groups (three treatment and one control) were not statistically significant (F = 0.29, df= 3; p>0.05).

The 95% family-wise confidence levels indicated that the higher differences were between the control and 5 mT treatment group in SL (-5.43) and FW (-0.29) (Fig. 5).

DISCUSSION

Plant development in terms of germination percentage is affected by MF due to physiological and biological processes (Sarraf et al., 2020). For instance, a study conducted by Jecli̇čka et al. (2015) revealed that germination percentages of tomatoes (*Solanum lycopersicum* L.) increased when seeds were exposed to MF flux densities of 20, 40, and 60 mT. However, other studies have not confirmed this effect (Ibrahim, 2015; Górski

Table 2. Effect of magnetic field (MF) treatments on germination percentages of soybean (Arısoy variety) at 24, 48 and 72 hours after MF application.

		Percentage of germinated seeds (%)		
Variety	MF Treatment (mT)	24 hours	48 hours	72 hours
Arısoy	Control	40	83	83
	0.5	35	71	94
	1.0	48	72	91
	5.0	61	86	92
		X^2=8.4, p<0.05	X^2=2.2, p>0.05	X^2=0.8, p>0.05

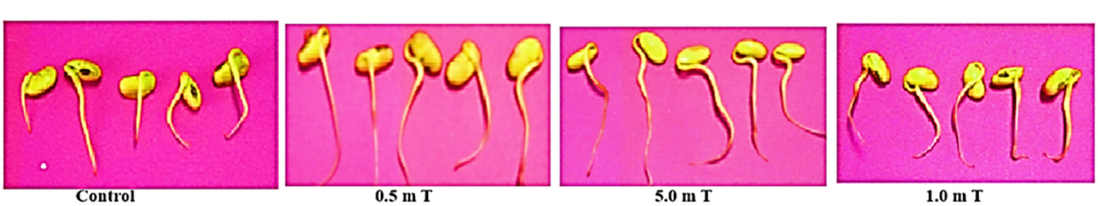

Fig 2. Germination of soybean seeds exposed to different magnetic field flux densities at 72 hours.

Table 3. Seedling length (SL), fresh weight (FW), and dry weight (DW) in MF treatments of 0.5 mT, 1.0 mT and 5.0 mT applied to soybean seeds (Arısoy variety).

Treatment (mT)	N	SL (cm)±sd	FW (g)±sd	DW (g)±sd
Control	61	27.54±8.26b	1.51±0.57b	0.14±0.04a
0.5	60	31.73±5.44a	1.79±0.51a	0.14±0.03a
1.0	54	32.52±5.72a	1.77±0.42a	0.14±0.03a
5.0	58	32.98±5.79a	1.79±0.43a	0.14±0.04a

Different letters in the same column indicate significant differences ($p < .05$) according to LSD Multiple Comparison Test. sd: standard deviation.

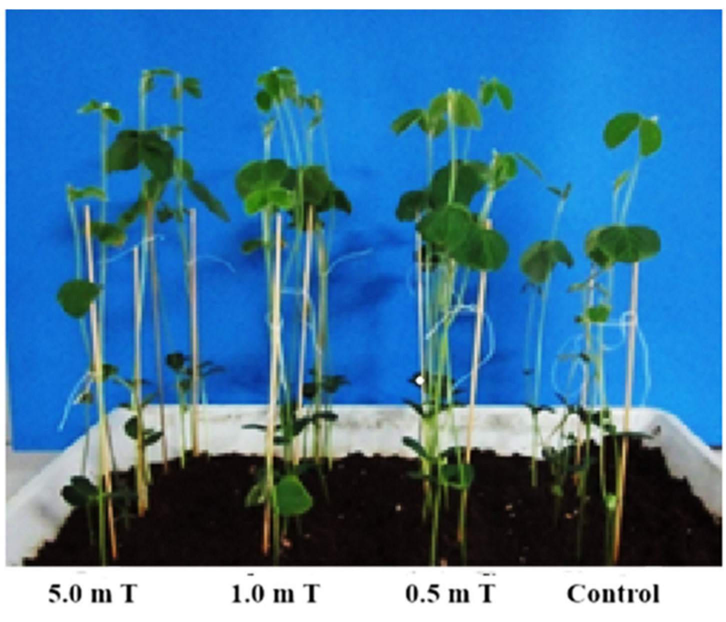

Fig. 3. Seedling height response of soybean seedlings at 72 hours after magnetic field application of different magnetic field flux densities.

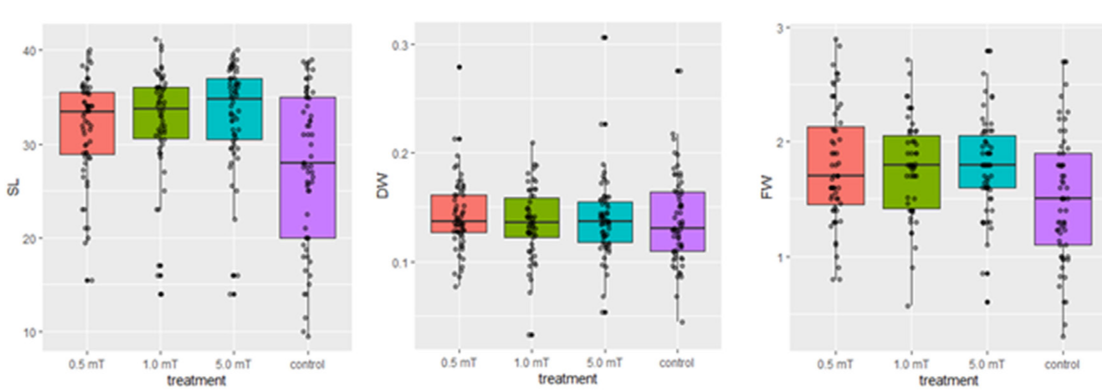

Fig. 4. Boxplot with dots of data (SL: seedling length in cm, FW: fresh weight in g and WD: dry weight in g) measured in seedlings exposed to three MF flux densities and control groups.

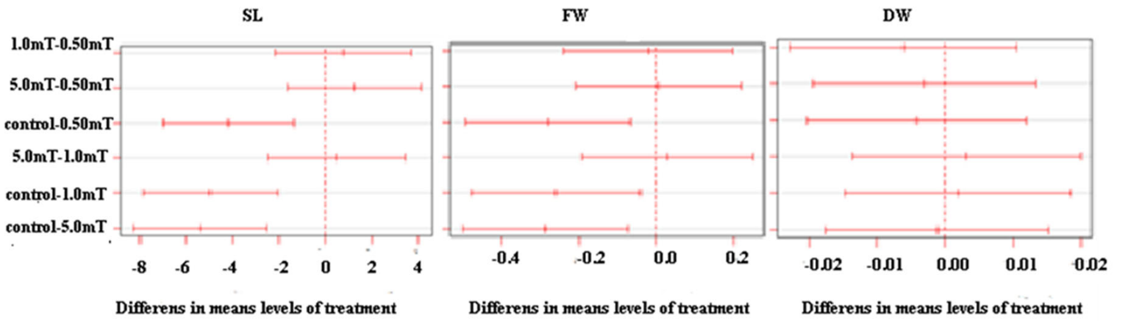

Fig. 5. 95% family-wise confidence level of the data set (SL: seedling length in cm, FW: fresh weight in g, WD: dry weight in g) indicates differences in mean level of treatment.

et al., 2019; Nassisi et al., 2020). Besides the type of plant species, MF flux density and duration of exposure of seeds or seedlings to MF are two key factors that determine whether the effect is positive or negative. The direction of this impact mechanism is important, particularly for seeds with low germination capacity. Đukić et al. (2017) reported that low-frequency (16, 24, 30 and 72 Hz) MF exposure in soybeans resulted in high percentages of germination. In the present study, the application of extremely low frequencies of MF (0.5-5.0 mT) for a 1-hour period positively affected the germination percentage of soybean plants.

Seeds stimulated by MFs not only germinate faster but also exhibit more uniform growth. They start vegetation earlier and show enhanced enzyme activity due to a higher yield, which accelerates metabolism. The effects of MFs on plant seeds has been studied across a wide range of magnetic induction values, varying from just over 10 μT to as high as 10 T (Pietruszewski and Martínez, 2015), since differences in plant genotypes can lead to varied responses to MFs. Therefore, detecting optimum MF frequencies that contribute to the development of all plant types is important. In soybeans, Payez and Ghanati (2018) determined that an MF of 30 mT at 10 kHz MFs increases productivity, with positive impacts on soybean growth. Another study conducted by De Souza-Torres et al. (2020) showed that different MF frequencies, such as 120 mT (10 min) and 80 mT (5 min), caused an increase in soybean root length, plant height, root and shoot dry mass, leaf area per plant, root and shoot relative growth rates and net photosynthesis rate. Furthermore, Ramesh et al. (2021) indicated that the application of shock waves to seeds increased the germination rate and physiological growth of *Catharanthus roseus* plants. Similarly, shock wave irradiation showed positive effects on the agronomic parameters of *Vigna mungo* seeds sown in red soil. In this sense, the results of the present study are consistent with previous research, showing that MFs have positive impacts on soybeans in terms of seedling length, seedling weight and germination percentage.

The increase in growth parameters may be related to the physiological process observed in plants. Cytokinins (plant growth regulators in cell division), auxins, and gibberellins are also the most important parameters (biomolecules) to increase plant elongation (Baltazar et al., 2021). Previous studies have revealed that MFs have an impact on plant growth regulators, especially the metabolism of auxin, cytokinin, and gibberellin during seedling and root elongation (Hafeez et al., 2023; Erez and Özbek ,2024). In addition to protein synthesis mechanisms, MF exposure causes an alteration by influencing ionic current density, permeability, ionic concentration in each side of cell membrane, osmotic pressure, and water intake rate of cells (Jones, 2016; Sarraf et al., 2020; Zadeh-Haghighi and Simon, 2022).

In the present experiment, the treated seedlings did not show any significant increase in terms of dry weight. However, MF application resulted in increased seedling length and fresh weight, but had no impact on dry weight, which may be related to the water retention capacity of the plant.

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CONCLUSIONS

Whether living systems are affected by electric, magnetic, and electromagnetic fields depends on their response system. The effects of magnetic exposure on plant growth still require more accurate explanations, as current knowledge is based on preliminary characteristics and needs to be validated through field experiments. The results obtained in the present study indicate that magnetically treated seeds showed higher seed germination percentage, seedling growth, plant height, and shoot fresh weight compared with non-magnetically treated seeds. The treatment of 5 mT with a magnetic exposure of 1 hour yielded the best results.

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Conflicts of Interest: The authors declare no conflict of interest.

Author Contributions

Conceptualization: Gizem Ören Cıbık and F. Sevil Yalcin; Methodology: F. Sevil Yalcin; Discussion: Gizem Ören Cıbık and F. Sevil Yalcin; Writing- review and editing: Gizem Ören Cıbık and F. Sevil Yalcin. The authors approved the final version of this manuscript.

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