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Effect of Magnetic Field on Growth and Development Parameters of *Rudbeckia hirta* L. Seed in Dry and Humid Conditions

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The magnetic field (MF) is considered as a biophysical treatment to change the growth and development parameters. Current research was carried out to evaluate the effect of different intensities of MF (0, 45, 60, 70 and 75 mT) on seed germination and some morphologic and physiologic parameters of *Rudbeckia hirta* L. in dry and humid conditions for 30 min. Results showed that the MF treatment increased germination percentage of dry seeds more than humid seeds and control. The intensity of 70 mT induced the maximum content of germination of dry seeds (95.40%). The MF decreased the seeds germination rate in both dry and humid conditions, as the control seeds showed the highest germination rate (3.5 days). Investigation of all morphologic traits like plantlet length and vigor index and physiologic (humid and dry weights of plantlet) revealed that the MF caused more induction of growth in these traits than the control. Totally, the use of 70 mT MF on dry seeds for optimum growth and development in *Rudbeckia hirta* L. is recommended.

Keywords: Biophysical treatment, Electromagnetic field, Ornamentals, Seed germination.

Abstract

INTRODUCTION

Rudbeckia hirta L. with Persian name of susan-e-siahcheshm from Asteraceae family is an ornamental-medicinal plant indigenous to East and Central parts of North America. Propagation of this plant is done by seed. Acceleration of the seed germination percentage, suitable establishment of seedling and optimization of quantity and quality characters of plantlets using different physical, mechanical and chemical treatments are important approaches.

Magnetic field (MF) as a biophysical treatment changes the growth and development characteristics of plants without change in their genetic structure (Vasilevski, 2003). These treatments are considered as a safe method to increase the quality and quality of crop plants and by reducing the harmful effects of chemical treatments, they increase the health of food and the environment (Aladjadjiyan, 2007; Dhawi et al., 2009). The use of electromagnetic field as a part of modern technology has been used by some researchers (Bilalis et al., 2013). MF has positive effect on the seed viability, improvement of seed germination, improvement of morphological, physiological and biochemical characters and resistance against many biotic and non-biotic stresses (Ruzic and German, 2002; Esitken, 2003; Rochalska and Orzeszko-Rywka, 2005; De Souza et al., 2006; Cakmak et al., 2010). MF also affects the cell division, protein biosynthesis, some secondary metabolites biosynthesis, cellular respiration, photochemical and enzymatic activities, the content of nucleic acids, oxidative stress, permeability of plasma membrane and other growth and development parameters (Stange et al., 2002; Rakosy-Tican et al., 2005; Celik et al., 2008; Cakmak et al., 2010). The effect of MF may be related to the paramagnetic properties of atoms in plant cells (Aladjadjiyan, 2010). MF increases the activity of auxin and respiration in seeds that is caused to the increase in water absorption by seed (Marinkovic et al., 2008). Several researchers have also reported that the positive effects of MF on seeds are related to improved ion movement across plasma membranes, the increase of amino acid uptake (Stange et al., 2002) and increase of both ion content and biosynthesis of chlorophyll and carotenes useful for seedling nutrition (Dhawi and Al-Khayri, 2009). Increase in the content of some ions like potassium and calcium in the cells causes decreasing water potential followed by greater absorption of water (Garcia and Arza, 2001; Esitken and Turan, 2004; Dhawi et al., 2009).

The effect of the MF treatment on seed germination and growth and development parameters of plantlets was stronger for preliminary soaked seeds (Aladjadjiyan and Ylieva, 2003). The effect of MF on soaked seeds of tobacco (Nicotiana tabacum L.) was greater than that of nonsoaked seeds (Aladjadjiyan and Ylieva, 2003). This observation can be due to the paramagnetic properties of water that absorbs the energy of MF. Intensity and time of the effect of MF affects the germination percentage and rapidity of plantlets (Kalinin and Boshkova, 2003; Aladjadjiyan, 2007; Pietruszewski and Kania, 2010). Most studies regarding to the MF were carried out on agronomic plants such as wheat (Phirke et al., 1996a), rice (Carbonell et al., 2000), soybean (Phirke et al., 1996a), corn (Florez et al., 2007; Garcia et al., 2008), bean (Podlesny et al., 2004), sunflower (Vashisth and Nagarajan, 2010), onion (Alexander and Doijode, 1995) and barely (Martinez et al., 2000) and study on horticultural and medicinal plants is few. One of the obstacles to the use of ornamental-medicinal plants out of natural habitat is limitation in germination percentage and seed dormancy. Therefore, the aim of the present study was to increase the germination percentage, increase the seed vigor and stimulation of some morphological and physiological parameters of Rudbeckia hirta L. (an ornamental-medicinal species) using different MF intensities on humid and dry seeds.

MATERIALS AND METHODS

Experiment condition and plant materials

The investigations were undertaken at the plant physiology laboratory of Islamic Azad Uni-

versity, Lahijan Branch and horticultural sciences laboratory of Islamic Azad University, Rasht Branch. Seeds of *Rudbeckia hirta* L. for the present investigations were purchased from Mansouri Gol Company and used as plant specimens. Investigations were done on 2018. Half of seeds were soaked in distilled water for 24 h. Cultivation beds containing sand, peat and perlite (in ratio of 1:1:1) were prepared.

Experimental treatments

Seeds were divided into two groups including dry seeds and humid seeds. Each of these two groups was treated with different MF intensities. Non-exposed seeds were used as control. The first factor of the experiment included the two levels of dry seeds and humid seeds and the second one included intensity of MF (0, 45, 60, 70 and 75 mT). All seeds were uniformity exposed to the MFs for 30 min. Following the exertion of treatments, seeds were sown on humid double-layer filter paper in Petri dishes and the dishes were sealed with rolled parafilm. Petri dishes containing seeds were kept in a suitable condition with the temperature of $21 \pm 2^{\circ}$ C for germination and growth. After the 14th day of incubation, seedlings were removed from the filter paper and cultivated in the pots containing cultivation beds. The pots were kept in a greenhouse with temperature of $24-26^{\circ}$ C and an air relative humidity of 70% with periodically irrigation.

Measured parameters

Measurements were started after the 2th day of cultivation. Final measurements were done after 60th day of cultivation. When a root tip of 2.0 mm length had emerged from the seed, it was considered germinated. The count of germinated seeds was done every day and continued until there was no increase in germination between two successive counts. Seed germination percentage, seed germination rapidity, stem length, root length, plantlet height, seed vigor index 1 and 2, plantlet fresh weight and plantlet dry weight were measured. Length of the roots and stems were measured by a millimetric ruler. Seed germination percentage and seed germination rapidity were calculated by followings formula:

 $Germination \ percentage = \frac{Total \ number \ of \ germinated \ seeds}{Total \ number \ of \ cultivated \ seeds} \times 100$

$$Vg = \Sigma \frac{Ni}{Di}$$

Where; "Vg" is germination rapidity (seed number per day), "Ni" is the number of germinated seed per day and "Di" is day number.

Seed vigor indexes 1 and 2 were calculated by followings formula:

V.I=Ls×Pg

Where; "V.I." is seed vigor index 1, "Ls" is the mean of plantlet height (cm) and "Pg" is germination percentage.

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Seed vigor index 2 = dry weight of plant × germination percentage
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After measuring fresh weight, plant specimens then were dried in an oven under 70°C temperature for 72 h and their dry weight was measured.

Experimental design and statistical analysis

A factorial experiment in a completely randomized design was employed. Four replicates of five seeds were randomly assigned per plot, thus, a total of 200 seeds were used. The first factor of the experiment was the two levels of dry and humid seeds and the second one was intensity of MF. Statistical analyses were done by using Statistical Package for Social Sciences (SPSS) and MINITAB. The analysis of variance (ANOVA) procedure for a factorial experiment was used to test for significant effect of treatments, followed by LSD test for comparisons of different means of different treatments.

RESULTS AND DISCUSSION Seed germination percentage

Data analysis showed that the effect of different intensities of MF in the two humid and dry conditions were significant on all measured parameters including seed germination percentage (P<0.01) (Tables 1 and 2). MF promoted the germination percentage of seeds under dry condition, so their germination percentage was found more considerable than that of humid seeds and control. It is better to be used the minimum intensity of MF for stimulation of seed germination if using a humid seed. If a comparison is made between all treatments, it is determined that the maximum and minimum germination percentage of seed (95.40 and 39.00%) was observed in dry seeds treated with 70 mT MF and humid seeds treated with 75 mT MF, respectively (Tables 3 and 4). All MF intensities had negative effect on germination percentage of humid seeds and positive effect on dry seeds.

The stimulatory effect of MF on the seed germination reported in this study is in accordance with that obtained by other researchers. Naz et al. (2012) and Kargarshooraki and Majd (2016) reported some positive effects of MF on the germination and emergence of Abelmoschus esculentus and Nigella sativa L. Application of MF improved the germination percentage of Phaseolus vulgaris and Triticum aestivum (Cakmak et al., 2010), Cicer arietinum L. (Podlesny et al., 2005) and Triticum sp. (Pietruszewski and Kania, 2010). Similar findings were reported with 30 and 100 mT on Hordeum vulgare L. (Martinez et al., 2000). De Souza et al. (2010) showed the stimulatory effect of MF on germination rate of Solanum seed in both in vitro and in vivo conditions. Similar results were reported by Florez et al. (2007) on Zea mays. The reason of this effect can be searched in the presence of paramagnetic properties of atoms in cells, which can cause an acceleration of seeds metabolism by magnetic treatment (Aladjadjiyan, 2010). MF treatment causes the physical, biochemical and physiological alterations and increases water absorption, mineral and nutrients uptake, permeability of plasma membrane, photosynthetic activity, proteins and enzymes activity and energy transformation in plants organs particularly seeds (Wadas, 1991; Yinan et al., 2005; Aladjadjiyan, 2010; Selim and El-Nady, 2011). In seeds treated with suitable intensity of MF, seed coat absorbs more water and stimulatory enzymes of germination have more activity (Aksenov et al., 2001). The exact mechanism of the effect of MF on the cell remains unknown, even though some hypotheses have been proposed to confirm the positive effect. One of these assumptions suggests that the MF is likely to improve the respiratory activity and metabolism energy (Rochalska and Orzeszko-Rywka, 2005). Some studies were not shown the positive effect of MF on seed germination percentage (Mahdavi et al., 2008; Majd et al., 2009). MF depending on its application, the conditions of plant matter especially age and severity and environmental conditions such as temperature and humidity can have different effects on plant metabolism (Cakmak et al., 2010). The intensity of the MF and duration of its application varies in different plant species, which comes from the different genetic structure of the species.

Seed germination rapidity

As the fastest seed germination (3.5 days) was observed in control seeds, none of the MF treatments were able to accelerate seed germination rapidity. Among all treatments of MF intensities, the fastest germination time (4.21 days) was observed in humid seeds treated with 45 mT MF. If it is to be selected a treatment that the percentage and rapidity of germination are appropriate and the present study can introduce that, the treatment is dry seeds imposed by the MF intensity of 70 mT. The average rate of seed germination in different treatments does not show a difference between the humid and dry seeds, so that the average rate of germination in humid seeds was 4.71 days and in dried seeds, 4.73 days (Tables 3 and 4).

Contrary to findings of the current study, stimulatory effect of MF on seed germination rapidity was reported in different plants (De Souza *et al.*, 2010; Florez *et al.*, 2007; Kargarshooraki and Majd, 2016). The MF increased the germination rapidity of seeds of some plant species by increasing the permeability of the seed coat to the water and stimulating enzymes involved in the germination (Aksenov *et al.*, 2001; Naz *et al.*, 2012). De Souza *et al.* (2010) showed that the germination rapidity of potato seeds treated with different intensities of MF was higher than that of control. Species difference is the main cause of this difference at the rate of germination of different seeds.

Seed vigor index

MF increased seed vigor index 1 by increasing the germination percentage of dry seeds and optimal stimulation of plantlet height. Therefore, the highest and lowest seed vigor index 1 (1968.00 and 754.00, respectively) was obtained in treatments of dry seeds stimulated with MF intensity of 70 mT and humid seed of control (Tables 3 and 4). On the other hand, the highest and lowest seed vigor index 2 (3.613 and 0.964, respectively) was observed in treatments of dry seeds stimulated with MF intensity of 45 mT and humid seeds stimulated with MF intensity of 60 mT (Tables 3 and 4).

The results obtained in this research are in contrast with some other researches (Mahdavi *et al.*, 2008). These studies showed that MF decreased seed vigor index through decreasing the plantlet height and weight, decreasing the germination rapidity and percentage and increasing the germination time. Reports of some findings on pea and wheat are in accordance with current study (Podlesny *et al.*, 2005; Pietruszewski and Kania, 2010). MF treatment on seed of *Helianthus annuus* caused an increase of 18-74% in seed vigor index (Vashisth and Nagarajan, 2010). Reaction of seed vigor in plants depends on time and intensity of MF, pretreatment method and type of species. The need for further physiology study in this area will be felt completely.

Length of root, stem and plantlet

Dry seeds treated with MF 70 mT by the production of the root to the length of 9.27 cm and the plantlet height to the length of 20.61 cm was the best. Maximum stem length (11.34 cm) was obtained in dry seeds treated with MF 70 mT. Minimum root length (4.26 cm), stem length (6.54 cm) and plantlet height (10.80 cm) was calculated in control seeds (Tables 3 and 4).

Effect of MF on stimulation of growth of vegetative organs in some species was reported (Kargarshooraki and Majd, 2016). The initial stimulation of plantlets follows an increase in the growth and yield of the plant. One of the hypotheses is that the MF increases the absorption of water by seeds and plantlet, which leads to an increase in the root and stem growth (De Souza *et al.,* 2008). Stimulatory effect of MF on the seed may leads to changes in some effective parameters on plant growth such as increasing the uptake of ions and amino acids by the plasma membrane and increasing the chlorophyll and carotene biosynthesis (Stange *et al.,* 2002; Dhawi and Al-Khayri, 2009; De Souza *et al.,* 2010). Report of Kargarshooraki and Majd (2016) is similar to our

S.o.V.	df	Germination Germination Seed vigor percentage rapidity index II	Fermination rapidity	Seed vigor index II	Seed vigor index I	r Fresh weight of plantlet		Dry weight of plantlet	Plantlet height	Stem leng	Stem length Root length
MF intensity	4	550**	1.47**	0.505**	59598**	0.026**	*	0.00021**	49.00**	11.42**	13.20**
Error	15	1.07	0.239	0.016	5311	0.00064235		0.00000577	1.5007115	0.718	0.72
CV (%)	ı	2.01	10.9	9.603	8.595	5.445	Ο.	9.201	7.18	8.88	11.32
** Significant at the 0.01 probability level.	the 0.01 p	probability level.									
Table 2. Anal of <i>Rudbeckia</i>	ysis of va	Table 2. Analysis of variance of the effect of different magnetic field intensities on seed germination and growth and development parameters of <i>Rudbeckia hirta</i> L. in dry condition.	ect of differe	ont magneti	c field inte	nsities on s	eed germ	ination and g	rowth and c	levelopmer	t parameter
Table 2. Anal: of <i>Rudbeckia</i>	hirta L. i	in dry condition. Germination	ect of different	ent magnetic fiel ion Seed vigor index II	c field inter vigor Se	tensities on s Seed vigor index I	eed germinat Fresh weight of plantlet	ination and gro ght Dry weight et plantlet	on and growth and dev Dry weight of Plantlet plantlet height	levelopment let Stem let length	t parameter
Table 2. Anal: of <i>Rudbeckia</i> S.o.V. MF intensity	ysis of va hirta L. df	in dry condition Germinatio	n Germinat n.0.927*	ont magnetic f ion Seed vig / index I	c field inter rigor Se x II i	ensities on s seed vigor index I 727299**	eed germi Fresh weig of plantle	ination and gro ght Dry weight et plantlet 0.00028**	rowth and de ht of Plantle et height 8** 37.28**	levelopment let Stem ht length	t parameter h Root h length ** 4.85**
Table 2. Anal: of <i>Rudbeckia</i> S.o.V. MF intensity Error	hirta L. i df	in dry condition Germinatio percentage 431**	èct of differe n Germinat 0.927*	ont magnetic fie ion Seed vigor index II 2.97**	c field inter rigor Se x II i 7** 7	ensities on s seed vigor index I 727299**	eed germin Fresh weigh 0.028**	ination and grov ght Dry weight et plantlet 0.00028**	rowth and de ht of Plantle et height 8** 37.28**	levelopment let Stem ht length h ^{**} 15.56**	t parameters h Root h length ** 4.85**

Table 1. Analysis of variance of the effect of different magnetic field intensities on seed germination and growth and development parameters of

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MF intensity (mT)	Germination percentage	MF intensity Germination Germination (mT) percentage rapidity (day)	Seed vigor index II	Seed vigor index I	Fresh weight Dry weight of of plantlet (g) plantlet (g)	Dry weight of plantlet (g)	Plantlet height (cm)	Stem length (cm)	Root lengtl (cm)
0	70.10ª	3.545°	1.176°	754 ^{cd}	0.332°	0.0167 ^d	10.80 ^b	6.53 ^b	4.256 ^b
45	53.50^{b}	4.212 ^{bc}	1.889ª	998ª	0.523^{a}	0.0355ª	18.64 ^a	10.43^{a}	8.207 ^a
60	45.05 ^d	4.904^{ab}	0.964^{d}	839 ^{bc}	0.540^{a}	0.0212°	18.56^{a}	10.20^{a}	8.361ª
70	49.45°	4.734 ^{ab}	1.375 ^b	939 ^{ab}	0.464^{b}	0.0280^{b}	19.01 ^a	10.52ª	8.489^{a}
75	39.05°	5.018ª	1.139^{cd}	707 ^d	0.466^{b}	0.0290 ^b	18.16 ^a	9.98^{a}	8.180^{a}

Table 3. Mean comparison of the effect of different magnetic field intensities on seed germination and growth and development parameters

of Rudbeckia hirta L. in dry condition. Table 4. Mean comparison of the effect of different magnetic field intensities on seed germination and growth and development parameters

MF intensity (mT)	MF intensity Germination Germination (mT) percentage rapidity (day)	Germination rapidity (day)	Seed vigor index II	Seed vigor index I	Fresh weight Dry weight of of plantlet (g) plantlet (g)	Dry weight of plantlet (g)	Plantlet height (cm)	Stem length (cm)	Root lengtl (cm)
0	69.8 ^d	3.67 ^b	1.238 ^d	952°	0.3537 ^d	0.0177 ^d	13.65°	7.01 ^d	6.632 ^b
45	88.6°	4.89^{a}	3.613 ^a	1729 ^b	0.5612ª	0.0407^{a}	19.47 ^b	10.50°	8.973ª
60	92.4 ^b	4.71^{a}	2.141°	1878 ^a	0.5410^{a}	0.0232°	20.31^{ab}	11.28 ^b	9.031ª
70	95.4ª	4.59 ^a	2.675 ^b	1968ª	0.4322°	0.0277 ^b	20.61 ^a	11.34^{a}	9.265ª
75	92.9 ^b	4.69^{a}	1.238^{d}	952°	0.3537 ^d	0.0177 ^d	13.65°	7.01^{d}	6.632 ^b

finding. These researchers demonstrated that the length of root and stem in *Nigella sativa* L. treated with MF for 30 min. was higher than those of control. Results of study on seeds of *Lentil lens* Medic. and *Pisum* sp. showed that intensity of 125 mT of MF for 10 min. increased the length of stem and plantlet more than the control (Martinez *et al.*, 2009). Treatment of sunflower seed with MF increased rootlet length for 16-80%, shoot length for 6-41% and plantlet height for 12-57% (Vashisth and Nagarajan, 2010). Similar results were reported in *Zea mays* (Florez *et al.*, 2007; Racuciu *et al.*, 2008). Application of MF increased the growth of seedlings in *Vicia faba, Triticum, Lens esculinaris, Solanum tuberosum* and *Glycine* (Martinez *et al.*, 2002; Kordas, 2002; Esitken, 2003; Peñuelas *et al.*, 2004; De Souza *et al.*, 2010; Cakmak *et al.*, 2010).

Dry and fresh weights of plantlet

Maximum fresh weight (0.56 g) and dry weight (0.041 g) of plantlet was obtained from germination of dried seeds treated with MF intensity of 45 mT. Minimum fresh weight (0.33 g) and dry weight (0.017 g) of plantlet was calculated from germination of humid seeds without treatment with MF. None of MF intensity treatments had inhibitory effect on increasing the fresh and dry weights (Tables 3 and 4).

Results of studies on *Hordeum vulgare*, tomato and *Zea mays* are in accordance with present work (Martinez *et al.*, 2000; Esitken, 2003; De Souza *et al.*, 2006; Florez *et al.*, 2007; Racuciu *et al.*, 2008). Some reports revealed inhibitory effect of stronger MF intensities on fresh and dry weights of plantlets (Racuciu *et al.*, 2008; Cakmak *et al.*, 2010). On the other hand, weak MF had no any effect on dry weight of sunflower seedlings (Fischer *et al.*, 2004). Martinez *et al.* (2000) showed that MF increased the length and weight of barley seeds, and the degree of this effect depended on the duration of exposure.

CONCLUSION

Application of biophysical treatments such as electromagnetic fields as a part of modern technologies, is a safe and without injury to the environment and organisms. This approach causes increasing the quantity and quality of horticultural plants. Therefore, electromagnetic fields treatments are a suitable replacement for chemical treatments. Nowadays, some researchers apply the electromagnetic fields to improve the plant morphological, physiological and biochemical properties and resistance against the types of biologic and non-biologic stresses. Current study showed the positive effect of MF on increasing the germination percentage of *Rudbeckia hirta* L. seed and some morphological and biochemical parameters. As electromagnetic fields cause major changes in plants, it is necessary to pay special attention to factors such as field intensity and treatment duration. The present study proposes the MF intensity of 70 mT as appropriate treatment to optimize the seed germination and establishment of *Rudbeckia hirta* L.

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