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Seed Germination Enhancement of Zinnia (*Zinnia elegans*) Using Electromagnetic Field

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The effect of electromagnetic field was examined on Zinnia (Zinnia elegans) seeds to enhance seed germination and early growth. The seeds were exposed to four magnetic strengths of 15, 100, 400 and 800 µT in four durations of 30, 60, 120 and 240 min. The experiments were carried out in two forms of laboratory germination and soil emergence. Results showed that magnetic field application enhanced seed performance in both terms of laboratory germination and soil emergence. The magnetic field significantly (p≤0.05) affected root length, shoot length, fresh and dry root weight, fresh and dry shoot weight, seedling length and weight, germination percentage, speed of germination, vigor index I and II in laboratory germination. Also, the magnetic field exposure significantly affected soil emergence factors of root length, shoot length, dry root weight, dry shoot weight, seedling length, emergence percentage and emergence speed at 0.05 level of probability. The other factors were significantly (p≤0.05) different compared with unexposed control. No significantly differences were found on the interaction of time and exposure field. The best results were found to 400 µT (micro Tesla) on seed germination and seed emergence. The exposure durations of 240 and 30 min showed promising results over all magnetic fields in seed germination and emergence, respectively. However, there was no significant difference among exposure times.

Abstract

Keywords: Germination percentage, Speed of germination, Vigor index, Emergence ratio, Magnetic intensity.

INTRODUCTION

Seed pretreatment including chemical and physical treatments are widely used to improve plant performance. Physical treatments like electrical, microwave and irradiation are known to improve seed performance (Vashisth & Nagarajan, 2008). Since chemical treatments are cost effective and harmful for the environment, physical treatments are able to improve plant performance without adversely affecting the environment. The physical treatments influence the physiological and biochemical process in seeds and thereby contribute in greater vigor and improve crop stand. Magnetic field treatment is one of the physical pre-treatment that have been reported to enhance the performance of various crops. All plants on earth live under an electromagnetic field because the earth is a magnet. The external electromagnetic fields have been reported to influence both the activation of ions and the polarization of dipoles in living cells (Moon and Chung, 2000). The optimal external electromagnetic field could accelerate the activation of seed germination but the mechanism of these actions is still poorly understood. It seems that some specific cells in plants are sensitive to earth magnetic field. These cells include amylloplast which are commonly located at root ends. Their sensitivity to magnetic field has been proved (Piruzian et al., 1980). Alexander and Doijode (1995) reported that onion and rice seeds exposed to very weak magnetic filed for 12 h had better performance in germination, shoot and root length of seedling. Harichand et al. (2002) reported that exposure of magnetic field of 10 mT (mili Tesla) in duration of 10 h increased plant height, seed weight per spike and yield of wheat. Aladjadjian (2002) found that the magnetic field can dramatically enhance the root development of maize. The faster germination and higher root and shoot weight was also reported. Rajendra et al. (2005) observed that Vicia faba seedling was enhanced by alternating magnetic field of 100 µT (micro Tesla). They reported that the reason could be increasing of mitotic index and water uptake. In broad bean and pea cultivars the magnetic stimulation of seeds improved the sprouting and emergence of seed and resulted in higher pod numbers and seed yield (Podlesny et al., 2004, 2005). Carbonell et al. (2000) concluded that a static magnetic field can stimulate the rice germination. Martinez et al. (2000) reported a significantly effect of 25 µT exposure in wheat initial growth. There are several reports on the effects of magnetic field on enhancement of various seeds (Samy, 1998, Garcia et al., 2001). Pittman (1963) observed an increase on rate of germination of cereal seeds exposed to magnetic field. Alexander and Doijode (1995) noted that the application of an external magnetic field as a pre-germination treatment improved the germination and seedling vigor of low viability rice seeds. Further, Podlesny et al. (2004) also published the positive effect of magnetic treatment on the germination and emergence of two broad bean cultivars. Similar effects were observed on cucumber seedlings by Yinan et al. (2005). Soltani et al. (2006 a, b) have also published the positive effect of magnetic field on Asparagus officinalis and Ocimun basilicum seed germination and seedling growth. There are many other research on the effect of magnetic field on various seeds and agricultural products (Aksyonov et al., 2001, Burtebayeva et al., 2003, Dorna et al., 2010, Fisher et al., 2004, Piacentini et al., 2001, Rajendra et al., 2005, Florez et al., 2004).

Zinnia (Zinnia elegance Jacq.) is grown as a bedding plant and summer specially cut flower (Nau, 1991). Many cultivars of this annual plant are sold worldwide. The zinnia tolerates dry and sunny environment make it to be suitable for those places in which growing other types of flower would be difficult. Reduction of germination of zinnia seeds exposed to high humidity (70%) and high temperature (37°C) was reported by Mano et al. (2006). These climatic conditions exist in the most period of year in Jiroft, south east of Iran, where the germination of zinnia seed as a bedding plant has some difficulties. Mano et al. (2006) found that a magnetic field applied to Arabidopsis thaliana L. and Lactuca sativa L. seeds prevented the reduction in germination, caused by seed incubation at high relative humidity and temperature. The objective of this study was to stimulate the germination of zinnia seed using magnetic field to overcome the germination difficulties concerned to mentioned climatic conditions.

MATERIALS AND METHODS

The Zinnia seeds were purchased from Pakan Seed Company (Pakan & Co.) and were cleaned from trash, leaves and other external materials. The review of available literature revealed that the magnetic field generators are developed for a limited field strength in which the field provided low or high exposure range. Since the present study is a part of an expand project of herb pre-sowing treatment in which an extremely wide range of magnetic field was required, it was decided to design and fabricate a custom built set up in which corresponding magnetic field range would be selected. The experimental set up was developed in Department of Agricultural Engineering, University of Jiroft, Jiroft, Iran. The set up was consisted of a stand, two magnetic spool coils, two aluminum sliding cores and a power supply (Fig. 1). The power supply was able to produce two separate lines of 220/24 Volt DC and 500 W (Watt) active power. The spool coils were cylindrical in shape with 160 mm in length, 8.6 mm and 8.8 mm inside and outside diameter, respectively. The number of turns per coil was 2250 and a resistance of each coil was 31 Ohm. The experimental set up was adjusted by sliding aluminum cores. The more sliding core inside of the spools, the lower field could be obtained. The calibration process was carried out using a 3-axial digital Tesla meter model LMF-828 (Lutron Company, Taiwan). Since the magnetic field strength of 15, 100, 400 and 800 µT (micro Tesla) were required, the positions of the sliding cores for these values were determined and marked. Many used high intensity magnetic fields to initiate plant seeds. However, it is not recommended because in some cases high intensity magnetic fields showed inhibitory effects on seed germination. The use of high intensity magnetic fields are also cost consuming in commercial range (Krizaj and Valencic, 1989). Therefore, authors decided to use lower magnetic intensity in range of 15 to 800 µT. A variation of 0.5 µT in horizontal direction and 1.1 µT in vertical direction were measured and considered to be neglected.

Seeds seemingly healthy without visible defects, insect damage and malformation were randomly tested in order to ensure high viability. Petri dishes of 80 mm in diameter and 10 mm depth with filter paper were used as germinators. The seeds and all experimental materials were sterilized with Sodium hypochlorite 5%, washed three times with distilled water and dried at room temperature. Seeds were placed on filter paper soaked with 3 mm of distilled water in Petri dishes. Three replicates with 50 seeds in each Petri dish were used in experimental design. The study was performed under laboratory conditions with natural light, average room temperature of 20°C and relative humidity of 35% (Dole and Wilkins, 1999).

The zinnia seeds were exposed to various magnetic strengths of 15, 100, 400 and 800 μ T in duration of 30, 60, 120 and 240 min. At the same time, the control, during each exposure time, was kept in the same condition but without being in the magnetic field. Seed samples were placed in germination room at 22-24°C daily night temperature, relative humidity of 50% and with natural light cycle 14-h light/10-h darkness. These conditions were suggested by Dole and Wilkins (1999) for zinnia seed germination. Number of germinated seeds was scored 8 times per day for the time necessary to achieve the final number of germinated seeds. Seeds were considered as germinated when their radicle showed at least 2 mm length. For evaluation of seed germination some seed parameters had to be measured. They were root length, shoot length, fresh and dry root weight, fresh and dry shoot weight, seedling length and weight, germination percentage, speed of germination, vigor index I and II. The vigor indices calculated using the following equations (Abdul-Baki and Anderson, 1973):

$$VI = (GP) \times (SdL)$$
 (1)

$$VII = (GP) \times (SdW)$$
 (2)

Where, GP is germinated percentage, SdL and SdW are seedling length and weight, respectively. To calculate the speed of germination, GS, the germinated seeds were recorded until no

more seeds germinated and calculated as (Vashisth & Nagarajan, 2008):

$$GS = (N1/T) + ... + (Nn/Tn)$$
(3)

Where, GS is germinated speed, N1 and Nn are the number of germinated seeds in the first and last count, respectively, T1 and Tn are the time of first and last count, respectively.

The fresh and dry root weight, fresh and dry shoot weight and seedling weight were measured by a digital scale (SCALTEC INSTRUMENT, SBA 32, 120g, Heiliy Enstadt, Germany) with 0.0001g precision.

The exposure strengths of 15, 100, 400 and 800 μ T in durations of 30, 60, 120 and 240 min were further examined for their effect on early growth and emergence in soil. Seeds were sowed in homogenous garden soil in 120 pots. In each pot, four treated seeds were sowed and kept under the conditions of 22-24°C daily night temperature, relative humidity of 50% and natural light cycle 14-h light/10-h darkness. Irrigation was provided as and when required. The seeds were considered emerged when shoot length were at least 2 mm (Martinez *et al.*, 2009).

A number of emerged seeds were recorded four times a day. After being reached to the maximum emergence in each pot, the strongest seedling was kept and the rest were removed. Three replicates were taken into account for each level of exposure and duration. The same procedure was carried out for the control but without being exposed to electromagnetic fields. When maximum emergence percentage was achieved, seedlings were gently removed, cleaned and washed from soil. Having measured, root, shoot and seedling length, seedling shoot was separate from root in order to weigh fresh root and shoot. They were dried at room temperature for three days and measured dry root and shoot weight (Vashisth and Nagarajan, 2008; Nimmi and Madhu, 2009). The soil emergence factors were root length, shoot length, fresh and dry root weight, fresh and dry shoot weight, seedling length, emergence percentage, emergence speed and the ratio of dry root weight to dry shoot weight. The filed emergence speed was calculated using the following equation (Zamiran, 2011):

$$ES = \sum \frac{NES}{ND}$$
 (4)

Where, ES, is emergence speed, NEs, number of emerged seeds per day, ND, days after planting.

The data was analyzed using SPSS (ver. 17) for both experiments one-factor analysis of variance was performed on a complete randomized design.

RESULTS

Laboratory results

The germination parameters determined for each treatment, expressed as mean of the three replicates are provided in Table 1. To analyze the effect of different magnetic fields on seed parameters, average values of different durations of each magnetic field were compared. The values of germination parameters were obtained for treated seeds and compared with corresponding control. Fig. 2 shows the germinated seeds under laboratory conditions. Exposure of zinnia seeds to different magnetic field intensities significantly increased germination-related characteristics. Among the various magnetic strengths, $400~\mu T$ was the most effective one (p \leq 0.05) in enhancement of all germination parameters. It follows by $100~\mu T$ and $800~\mu T$ which significantly affected seed germination parameters except for shoot length.

To analyze the effect of exposure durations on seed germination parameters, the average values of different magnetic fields of each exposure time were compared. Among the four duration exposure times, 240 min was the most effective one ($p \le 0.05$) in enhancement of all germination

parameters. It follows by 120 min which was significantly affected seed germination parameters except for shoot length.

It can be concluded that the most effective field intensity and duration exposure time were $400 \mu T$ and $240 \min$, respectively for zinnia seed germination enhancement.

Greenhouse results

The emergence parameters determined for each treatment, expressed as mean of the three replicates are provided in Table 2. To analyze the effect of different magnetic fields on seed emergence parameters, average values of different durations of each magnetic field were compared. The values of emergence parameters were obtained for treated seeds and compared with untreated seeds. Fig. 3 shows the emerged seeds under greenhouse conditions. Exposure of zinnia seeds to different magnetic field intensities significantly increased emergence-related characteristics while compared with control. Among the various magnetic strengths, 400 μ T was the most effective one (p≤0.05) in enhancement of all emergence parameters. This exposure field intensity was not affected significantly FSW (fresh shoot weight) and DRW/DSW (the ratio of dry root weight to dry shoot weight). Another effective magnetic field intensity was 100 μ T which significantly affected seed emergence parameters except for root length and emergence speed. Results obtained in emergence test allow us to conclude that magnetic treatment of 400 μ T improved zinnia emergence parameters better than other field intensities.

To analyze the effect of exposure durations on seed emergence parameters, the average values of different magnetic fields of each exposure time were compared. Among the four duration exposure times, 30 min was the most effective one ($p \le 0.05$) in enhancement of emergence parameters except for FSW (fresh shoot weight) and DRW/DSW (the ratio of dry root weight to dry shoot weight).

DISCUSSION

Magnetic field intensity

This study showed that exposure of seeds in both experiments of laboratory and greenhouse generally affected the most of germination and emergence parameters of zinnia seeds. The most effective magnetic field intensity was 400 µT which accelerate germination and emergence parameters of zinnia seeds while compared with untreated seeds. In accordance to the present study, Podlesny et al. (2004 & 2005) confirmed the positive effect of magnetic field of 30 and 85 mT on the emergence of broad bean. Celestino et al. (2000) reported enhanced germination and growth of Quercus suber seedling when exposed to electromagnetic filed. Results obtained from the present study indicated that the magnetic field clearly affect root length, fresh root weight, seedling length, germination percentage, germination speed and vigor index I. This finding is supported by Vashisth and Nagarajan (2008) who reported an increase of germination percentage, speed of germination, seedling length, seedling weight and vigor index I caused by magnetic exposure for chickpea. The main phenomenon investigated by different authors, found to be responsible for enhanced seed germination capacity and acceleration of morphogenetic processes, is increased water absorption by seeds (Reina et al., 2001, Burtebayeva et al., 2003, Fischer et al., 2004). Water exposed to the action of a magnetic field indicate an altered, usually reduced, surface tension, viscosity (Pang and Deng, 2008) as well as heat of evaporation, manifested in a faster vaporization of such water (Galland and Pazur, 2005; Nakagawa et al., 1999). These three phenomena are interrelated by the power of actions defined as hydrogen bonds. Changes in these parameters result in a faster penetration of water inside seeds inducing higher rate of enzymatic reactions consequently resulted in faster and more effective germination (Burtebayeva et al., 2003). Such an enhanced enzymatic activity under the influence of a magnetic field is observed in case of catalase (Piacentini et al., 2001), horseradish peroxidase (the participation of H3O+ ion), esterase (Galland and Pazur, 2005), amylase, protease and lipase (Rajendra, 2005). Some of these enzymes participate in seed germination.

Exposure duration

The maximum effect of exposure on germination parameters was obtained on 240 min. This finding is supported by Florez *et al.* (2008) who examined 250 mT in durations of 10 min, 20 min, 1 h, 24 h and chronic exposure on alfalfa seeds and reported that the greatest germination speed differences between treated seeds and control where obtained when seeds where treated for 24 h and chronically exposed. However, the most effective exposure duration on zinnia seed emergence was obtained on 30 min. Nagy *et al.* (2005) reported that longer exposure time cancels the stimulating effects of electromagnetic on wheat and sunflower seeds. Thus, results from greenhouse experiments demonstrated that the longer duration may prevent emergence enhancement. In other words, the exposure to magnetic field for longer than 30 min may have an inhibitor effect on zinnia seed emergence. It can be concluded that, zinnia seeds germination can be accelerated using electromagnetic field to obtain stronger seedlings. The combination of exposure time and field intensity plays the main role in seed enhancement and there is a limitation in use of magnetic exposure intensity and duration which requires further investigations to be proven. At present, we hope to attract the attention of the scientific community to study this interesting phenomenon, so that the results on other ornamental seeds can be investigated in the future.

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Tables

Table 1. Effect of magnetic field and its duration on seed germination parameters of zinnia.

Parameter	RL (mm)	SL (mm)	FRW (mg)	FSW (mg)	DRW (mg)	DSW (mg)	Mean SdL (mm)	SdW (mg)	GP	GS	I	VII
Magnetic field (μT)												
Control	39.42 a	18.67 a	33.74 a	7.68 a	2.20 a	0.40 a	58.08 a	2.61 a	72.00 a	4.76 a	4206 a	188 :
15	66.00 b	22.25 ab	51.56 b	9.06 ab	3.07 ab	0.73 ab	88.25 b	3.81 ab	93.00 Ь	8.73 b	8225 b	354 a
100	61.58 b	23.50 ab	61.22 bc	11.03 bc	5.33 c	0.89 bc	85.08 b	6.22 c	89.67 b	7.91 b	7645 b	558
400	70.42 b	26.83 b	67.48 c	11.42 с	5.21 c	1.24 c	97.25 b	6.45 c	93.01 b	8.67 b	9058 Ь	600
800	64.75 b	22.92 ab	51.93 b	10.14 bc	4.05 bc	0.95 bc	87.67 b	5.00 bc	91.83 b	8.59 b	8063 b	461 b
Duration (min)												
Control	39.42 a	18.67 a	33.74 a	7.68 a	2.20 a	0.40 a	58.08 a	2.61 a	72.00 a	4.76 a	4206 a	188 :
30	65.75 b	22.50 ab	60.15 b	10.06 ab	4.72 b	1.03 b	88.25 b	5.75 b	88.17 b	7.37 b	7794 bc	5051
60	60.75 b	23.00 ab	51.89 b	9.50 ab	4.27 b	0.85 ab	83.75 b	5.13 b	89.17 bc	7.26 b	7495 b	4581
120	68.33 b	22.92 ab	60.28 b	10.75 b	4.36 b	0.96 b	91.25 b	5.33 b	95.00 с	9.35 с	8668 bc	5051
240	67.92 b	27.08 b	59.88 b	11.34 b	4.31 b	0.98 b	95.00 b	5.30 b	95.17 c	9.92 с	9034 с	5051
Control 39.42 a 18.67 a 33.74 a 7.68 a 2.20 a 0.40 a 88.25 b 3.81 ab 15	39.42 a 66.00 b 61.58 b 70.42 b 64.75 b 65.75 b 60.75 b 68.33 b 67.92 b	18.67 a 22.25 ab 23.50 ab 26.83 b 22.92 ab 18.67 a 22.50 ab 22.50 ab 22.50 ab 23.00 ab 27.08 b 27.08 b	33.74 a 51.56 b 61.22 bc 67.48 c 51.93 b 51.89 b 60.15 b 51.89 b 60.28 b 59.88 b me letters are	7.68 a 9.06 ab 11.03 bc 11.42 c 10.14 bc 7.68 a 7.68 a 10.06 ab 9.50 ab 10.75 b 11.34 b	2.20 a 3.07 ab 5.33 c 5.21 c 4.05 bc 2.20 a 4.72 b 4.27 b 4.36 b 4.36 b 4.31 b 2.30 a	0.40 a 0.73 ab 0.89 bc 1.24 c 0.95 bc 0.40 a 1.03 b 0.85 ab 0.96 b 0.98 b 1.098 b 0.98 b	Control 39,42a 18,67a 33,74a 7,68a 2,20a 0,40a 88,08a 2,61a 72,00a 4,76a 4,96a 188 15 66,00b 22,25 ab 51,36b 9,06 ab 3,07 ab 0,73 ab 88,25b 3,81 ab 93,00b 87,3b 822,5b 354 100 61,58b 22,50 ab 61,22 bc 11,03 bc 5,33 c 0,89 bc 85,08 b 6,22 c 89,67 b 79,1b 7645 b 558 400 70,42 b 26,83 b 61,22 bc 11,42 c 5,21 c 1,24 c 97,25 b 6,45 c 93,01 b 867 b 90,85 b 600 b 400 4,75 b 22,92 ab 51,93 b 10,14 bc 4,05 bc 9,95 bc 87,75 b 5,00 bc 91,83 b 86,05 b 90,88 b 600 b 90 4,75 b 22,92 ab 1,04 bc 4,25 b 0,40 a 87,75 b 5,00 bc 91,83 b 86,65 b 461 Control 39,42 a <	2.61 a 3.81 ab 6.22 c 6.45 c 5.00 bc 2.61 a 2.61 a 5.75 b 5.13 b 5.33 b 5.33 b 5.30 b	72.00 a 93.00 b 89.67 b 93.01 b 91.83 b 91.83 b 72.00 a 88.17 b 89.17 bc 95.00 c 95.17 c 95.17 c	4.76 a 8.73 b 7.91 b 8.67 b 8.59 b 4.76 a 4.76 a 7.37 b 7.26 b 9.35 c 9.92 c	72.00 a 4.76 a 4206 a 188 93.00 b 8.73 b 8225 b 354 a 89.67 b 7.91 b 7645 b 558 89.67 b 8.67 b 9058 b 600 91.83 b 8.59 b 8063 b 461 b 72.00 a 4.76 a 4206 a 188 88.17 b 7.37 b 7794 bc 505 88.17 b 7.26 b 7495 b 458 95.00 c 9.35 c 8668 bc 505 95.17 c 9.92 c 9034 c 505 test, RL, root length, SL, shoot lengths, SdW, seedlling weight, GP, germinate	188: 354: 558 600 461: 188: 5051 458: 5051 5051 5051 5051 5051

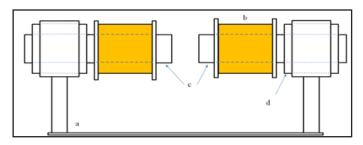
percentage, GS, germination speed, VI, vigor index I, VII, vigor index II.

Table 2. Effect of magnetic field and its duration on seed emergence parameters of zinnia.

Parameter	RL (mm)	RL (mm) SL (mm)	FRW (mg)	FSW (mg)	FRW (mg) FSW (mg) DRW (mg)	DSW (mg)	Mean SdL (mm)	DRW/DSW	ES	ЕP
Magnetic field (μΤ)										
Control	57.00 a	61.58 a	11.16 a	90.93 a	0.35 a	3.83 a	118.58 a	0.087 a	0.96 a	54.17 a
15	76.25 ab	83.08 b	15.51 ab	102.08 ab	0.73 b	6.01 b	159.33 b	0.119 ab	1.33 ab	75.00 ab
100	78.58 ab	98.38 b	17.83 b	137.28 b	0.89 b	6.61 b	176.96 b	0.146 b	1.36 ab	77.08 b
400	85.25 b	84.22 b	18.86 b	119.02 ab	0.95 b	7.11 b	169.47 b	0.140 ab	1.71 b	93.75 b
800	73.58 ab	84.58 b	17.44 ab	111.98 ab	0.83 b	6.05 b	158.17 b	0.141 ab	1.37 ab	77.08 b
Duration (min)										
Control	57.00 a	61.58 a	11.16 a	90.93 a	0.35 a	3.83 a	118.58 a	0.087 a	0.96 a	54.17 a
30	87.83 c	89.64 b	19.95 в	128.67 a	0.91 b	6.88 b	177.47 b	0.140 ab	1.58 b	89.58 b
60	83.92 bc	90.45 b	17.23 ab	120.45 a	0.93 b	6.39 b	174.36 b	0.155 b	1.27 ab	70.83 ab
120	77.42 abc	79.33 ab	16.27 ab	99.17 a	0.79 b	5.78 ab	156.75 b	0.142 ab	1.49 b	83.33 b
240		00011	16.17 ab	122.08 a	0.76 b	6 73 h	155.34 ah	0.108 ah	1 40 1	79.17 h

Means in columns followed by the same letters are not significantly different at α = 0.05 level according to Tukey's test, RL, root length, SL, shoot length, FRW, fresh root weight, FSW, fresh shoot weight, DRW, dry root weight, DSW, dry shoot weight, SdL, seedling length, DRW/DSW, the ratio of DRW to DSW, ES, emergence speed, EP, emergence percentage.

Figures



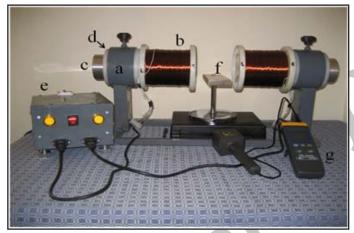


Fig. 1. Up, schematic diagram and down a figure of the electromagnetic field generator , a. stand, b. coil, c. sliding aluminum core, d. insulation ring, e. power supply, f. sample holder, g. Tesla meter



Fig.2. Germination of zinnia seed for various applied magnetic fields and exposure durations.



Fig. 3. Emergence of zinnia seed for various applied magnetic fields and exposure durations.