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Effect of Low Frequency (50Hz) Electromagnetic Field on Germination Process of Wheat (*Triticum aestivum* L.)

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Abstract **Original Research Article**

Electromagnetic field (EMF) and geomagnetic field (GMF) are inescapable environmental factors experienced by all living organisms including plants. Despite the progresses made in the field of plant-senses, the impact of Electromagnetic field (EMF) and geomagnetic field (GMF) on plant growth and development is not fully explored. In this context influence of exposure intensity and duration of exposure to low frequency (50 Hertz) electromagnetic field produced by a locally designed monoaxial Helmholtz Coil on germination parameters of *Triticum aestivum* L. (Wheat) has been studied in the present work. Wheat seeds were exposed to magnetic field strengths of 30, 60 and 75 Gauss for different periods of time (10, 20 and 30 minutes) inside the Helmholtz Coil. Control plants were grown under the local geomagnetic field (50 micro Tesla). Germination parameters like mean germination time, germination percentage, co-efficient of germination etc. were calculated for each test condition. Results indicated that among all the test conditions the alternating magnetic field with exposure rate of 60 Gauss/30 minutes found most conducive to Wheat germination which is reported here for the first time. These results are encouraging to work further on the effect of magnetic field on biochemical and molecular changes that may be resulting these observed phenomena.

Keywords: Electromagnetic field (EMF), Geo-Magnetic field (GMF), *Triticum aestivum* L., Helmholtz coil, Germination parameters.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the second most-produced cereal after maize (United Nations, FAOSTAT, 2014). It is an important source of carbohydrate, multiple nutrients and dietary fibre (Shewry PR and Hey SJ, 2015). Demands of global strategies for food and nutrition security with minimum environmental toxicity are encouraging study and adaptation of green technologies worldwide. Among these, magnetic field (Rath D and Padhi S, 2022) and sound waves (Padhi *et al.*, 2022) are gaining special attention for their bio stimulation effects. Dearth of magnetic field exposure devices with large uniform surface area restricts optimization of field exposure. The main objective of this work is to design a Helmholtz coil with large uniform surface area that is capable of producing different intensities of magnetic field and to quantify the effects of different magnetic exposure on germination characters of Wheat.

MATERIAL AND METHODS

Plant Material

Wheat seeds (*Triticum aestivum* L.) variety-HD 3117, India) were collected from Division of Seed Science and Technology, ICAR-IARI, New Delhi.

Soil Sample

Soil sample was collected from regions adjoining Noamundi Iron Mines, Noamundi, Jharkhand. The soil samples were analysed (Table 1) and used in the experiment after sieving.

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Table 1: Analysis result for soil sample collected from region adjoining Noamundi Iron Mines, Noamundi, Jharkhand

Location	Soil type	Taxonomic group	pH[a]	Organic Carbon[b] (%)	Total Nitrogen[c] ($mg.kg^{-1}$)	Phosphorus[d] ($mg.Kg^{-1}$)	Potassium[e] ($mg.Kg^{-1}$)	Iron[f] ($mg.Kg^{1}$)
Noamundi Iron Mines, Noamundi, Jharkhand	Laterite	Haplustulf	6.35	0.49	345	< 3	77	< 5

[a] Measured by taking 1:1.25 soil water ratio by Elico-digital pH meter
[b] Determined by Walkey-Black method
[c] Estimated by Kjeldal method
[d] Estimated by Bray's No.1 method
[e] Estimated by Platenic-chloride gravimetric method
[f] Estimated by Citrate dithionite-bicarbonate method

Design of Helmholtz Coil

Helmholtz coil (single axis) was designed (Gyawali S. R., 2008) and constructed in the present work. The structure was assembled using plywood that was cut into circular shape. As any type of metal and nails could not be used, glue was used to fix the structure with the desired specification (Table 2). 10 Standard wire Gauge (SWG) copper wire were used for winding. Each coil has 100 turns of copper wire and both the coils were connected in series. A base was constructed using plywood and fixed at the centre of the structure for exposure with uniform magnetic field (figure 1). The magnetic field produced inside the Helmholtz coil was calculated and measured using a gauss meter for accuracy of the obtained magnitude and uniformity data.

Table 2: Helmholtz coil specification

Type	Monoaxial
Coil radius	47.5 Cm
Number of turns per coil	100
Coil height	130 Cm
Copper wire (Gauge)	10 SWG Copper
Material	Plywood

Figure 1: Helmholtz coil with base for magnetic field exposure

Design of Autotransformer

Autotransformer was designed to get multiple output voltage from a single AC input of 220 Volt, 50 Hertz so that multiple output current was allowed to pass through the Helmholtz coil. As a result, different flux densities of magnetic field (30 G, 60 G and 75G) could be generated. The autotransformer was also capable of producing DC current. Generation of DC output was done using a bridge rectifier consisting of four diodes in series installed in the autotransformer.

Experimental Methods

Three different alternating magnetic flux densities (30 G, 60 G and 75G) produced by AC output were used for three different time intervals per day (10, 20 and 30 minutes) in the present experiment for fourteen days. All the test conditions were carried out and compared with control (Plants grown under Earth's magnetic field (GMF)). The experiments were carried out in triplicate in a controlled environment with constant temperature, humidity and Light.

The germination parameters were calculated as follows:

Germination percentage (GP)

Germination percentage is an estimate of the survivability of the population of seeds. The equation to calculate germination percentage is (Ranal MA, Santana DG, 2006):

$$\mathrm{GP} = \frac{\sum_{i=1}^{k} n_i}{N} \times 100$$

n_i= number of seeds germinated in the *i*th time

N= Total number of seeds used

Relativized percentage (RV)

The germination percentage can be relativized by the following equation (Labouriau, L. G., 1978):

$$\mathrm{R}\,(\%) = \frac{\mathrm{AP}}{\mathrm{HP}}\mathrm{X}100$$

AP= actual percentage

HP= highest percentage amongst the group of data

This standardization allows comparisons among treatments equivalent when the amount of dormancy broken varied.

Mean germination time (MGT)

Mean germination time is a measure of the average time for the seed to emerge or germinate. The following formula was used to calculate the mean germination time (Crosier *et al*., 1970):

$$\bar{t} = \frac{\left(\sum_{i=1}^{k} n_i t_i\right)}{\left(\sum_{i=1}^{k} n_i\right)}$$

$n_i t_i$= The product of seeds germinated at interval *i*th with the corresponding time interval

n_i= number of seeds germinated in the *i*th time

Mean germination rate (MGR)

The mean germination rate is the reciprocal of the mean germination time as shown below (Chougule Sb, 2018).

$$\bar{v} = \frac{1}{\bar{t}}$$

$\bar{t}$ = Mean germination time

Uncertainty of germination process (U)

The uncertainty of the germination process indicates the degree of uncertainty associated with the distribution of relative frequency of germination. Uncertainty is calculated using the following equation (Jones, K. W., and Sanders, D. C. 1987):

$$\mathrm{U} = \sum_{i=1}^{k} f_i \log_2 f_i$$

$$f_i = \frac{n_i}{\sum_{i=1}^{k} n_i}$$

fi= Relative frequency of germination

Low values of uncertainty indicate frequencies with few peaks (i.e., germination more concentrated in time). A low value (towards zero) indicates more synchronized germination.

Synchrony of Germination Process (Z)

Evaluate the degree of overlapping certain demographic. The synchronization index produces a number if and only if there are two seeds finishing the germination process at the same time. It is calculated using the following formula (Jones, K. W., and Sanders, D. C. 1987):

$$Z = \frac{\sum_{i=1}^{k} C_{n_i,2}}{C_{\Sigma_{n_i},2}}$$

$C_{n_i,2} = n_i(n_i - 1)/2$

$C_{n_i,2}$= combinations of seeds germinated in the *i*th time, two by two.

n_i= number of seeds germinated in the *i*th time

The number Z equals one when all seeds sprout at the same time. And when Z is zero, at least two seeds may sprout simultaneously, and so on.

Coefficient of variation of germination time (CVt)

The coefficient of variation of the germination time is calculated by the following formula (Chougule Sb, 2018).

$$\mathrm{CVt} = \frac{S_t}{\bar{t}} \times 100$$

S_t= standard deviation of germination time and calculated as

$$S_t = \sqrt{\frac{\sum_{i=1}^{k} n_i (t_i - \bar{t})^2}{\sum_{i=1}^{k} (n_i - 1)}}$$

$\bar{t}$ = mean germination time

Samples with only one seed germinated do not have the value of this measurement because the divisor of variance of germination time is zero.

Germination index (GI)

The germination index is an estimate of the time (in days) it takes a certain germination percentage to reach its goal. It is expressed as (Coolbear *et al*., 1984)

$$\mathrm{GI} = \frac{\sum_{i=1}^{k} n_i}{t_i}$$

n_i= number of seeds germinated in the ith time

t_i= time taken for seeds to germinate at the ith count

Coefficient of the velocity of germination (CVG)

The coefficient of the velocity of germination was calculated using the following formula (Adams, J. C., and Farrish, K. W., 1992).

$$\mathrm{CVG} = \frac{\sum_{i=1}^{k} n_i t_i}{\sum_{i=1}^{k} n_i} \times 100$$

Time to 50% germination (T_{50})

Time to 50% germination (T_{50}) indicates the time taken for half of the seeds to germinate. T_{50} can is calculated using the following formula (Czabator, F. J., 1962):

$$T_{50} = \frac{t_i + \left(\frac{\sum_{i=1}^{k} n_i}{2} - n_i\right)(t_j - t_i)}{n_j - n_i}$$

In the above equation to find out the value of $n_i n_i$ and $n_j n_j$ there is a need to look at the cumulative number of seeds germinated for which the condition is given below.

$$n_i < \left(\frac{\sum_{i=1}^{k} n_i}{2}\right) < n_j$$

n_i= nearest cumulative number of seeds germinated $C_{n_i} < \left(\frac{\sum_{i=1}^{k} n_i}{2}\right)$

n_j = nearest cumulative number of seeds germinated $C_{n_j} > \left(\frac{\sum_{i=1} n_i}{2}\right)$

Other time-related germination parameters like T_{10}, T_{25}, $T_{75,}$ and T_{90} were calculated using the same above formula by replacing $\frac{\sum_{i=1}^{k} n_i}{2}$ with $\frac{\sum_{i=1}^{n} n_i}{10}$, $\frac{\sum_{=1}^{k} n_i}{4}$, $3\frac{\sum_{=1}^{k} n_i}{4}$ and $9\frac{\sum_{=1}^{k} n_i}{10}$ respectively.

Mean Daily Germination Percent (MDG)

It represents the mean number of seeds germinated per day. This can also be defined as the number of seeds germinating daily relative to the maximum number of germinated seeds. It is calculated using the following expression (Vicient CM., 2017)

$$\mathrm{MDG} = \frac{\mathrm{GP}}{T_n}$$

GP= final cumulative germination percentage

T_n= total number of intervals required for final germination

Peak value (PV)

It is the accumulated number of seeds germinated at the point on the germination curve at which the rate of germination starts to decrease. It is computed as the maximum quotient obtained by dividing successive cumulative germination values by the relevant incubation time (Vicient CM., 2017).

Germination value (GV)

Germination value is calculated by combining both speed and completeness of germination into a composite score as described by Czabator, F. J. (1962).

GV=MDG×PV

MDG= mean daily germination

PV= peak value or largest quotient obtained when all the cumulative germination percentages were divided by the respective time interval.

Statistical Tools

A minimum of three replicates were conducted for each experimental group, and the results were calculated as mean ± standard deviation. Different letters (a–f) among the group denote the significant differences between the mean at $p < 0.05$ using one-way analysis of variance (ANOVA), Tukey's HSD test, and Scheffe's test using R-statistical tool version 4.2.2 and R-Studio version 2023.03.0-386.

RESULTS AND DISCUSSION

The maximum Relativized percentage found in seeds treated with 60 Gauss for 30 minutes (Table 3) followed by seeds treated with 60 Gauss for 20 minutes as compared with the control showing its minimum value. Relativized percentage establishes a linear relationship between treatments (30 Gauss and 60 Gauss) and thus it establishes that the seeds broke dormancy and germinated better under these treatment conditions (Fig 2). Relativized percentage decreased with increasing exposure duration past 60 Gauss (75 Gauss for 10, 20 and 30 minutes). The mean germination time (MGT) was reduced to 2.35 days for plants treated with 60 Gauss for 30 minutes followed by 3.06 days for plants treated with 60 Gauss for 20 minutes. Mean germination time 5.08 days recorded for control plants (Fig 3). The highest value of mean germination rate (MGR) was 0.43 days for plants treated with 60 Gauss for 30 minutes followed by 0.33 days for plants treated with 60 Gauss for 20 minutes as compared with control 0.20 days (Fig 4). The coefficient of variation of germination time (CVt) between control and seeds exposed to electromagnetic fields differs significantly (Table 3).

Table 3: Estimation of "Relativized Percentage of Germination (RPG), Mean Germination Time (MGT), Mean Germination Rate (MGR) and Coefficient of Variation of Germination Time (CVt)" of different treatments. Different letters (a–f) among the group denote the significant differences between the mean at $p < 0.05$ using one-way analysis of variance (ANOVA), Tukey's HSD comparison test, and Scheffe's test

Applied EMF (Gauss)	Treatment Period (Minutes)	Revitalization Percentage (RP) [%]	Mean Germination Time (MGT) [day]	Mean Germination Rate (MGR) [day^{-1}]	Coefficient of Variation of germination time (CVt) [%]
0	0	78.91±0.10^{f}	5.08±0.02^{a}	0.20±0.001^{e}	17.24±0.251^{c}
30	10	84.35±0.10^{de}	4.32±0.04^{ab}	0.23±0.002^{de}	22.15±0.065^{bc}
30	20	86.73±0.08^{d}	3.77±0.01^{bc}	0.27±0.001^{cd}	24.80±0.277^{c}
30	30	89.12±0.05^{cd}	3.62±0.00^{bc}	0.28±0.000^{bcd}	25.45±0.487^{c}
60	10	92.52±0.13^{bc}	3.28±0.01^{c}	0.31±0.001^{bc}	25.39±0.140^{c}
60	20	94.90±0.22^{ab}	3.06±0.00^{cd}	0.33±0.001^{b}	31.09±0.811^{c}
60	30	99.32±0.10^{a}	2.35±0.00^{d}	0.43±0.001^{a}	48.91±0.160^{bc}
75	10	94.56±0.10^{ab}	3.17±0.02^{c}	0.32±0.002^{bc}	46.08±0.115^{ab}
75	20	87.76±0.08^{cd}	3.53±0.01^{bc}	0.28±0.001^{bcd}	44.92±0.141^{ab}
75	30	80.27±0.10^{ef}	3.71±0.03^{bc}	0.27±0.002^{cd}	32.78±0.296^{a}

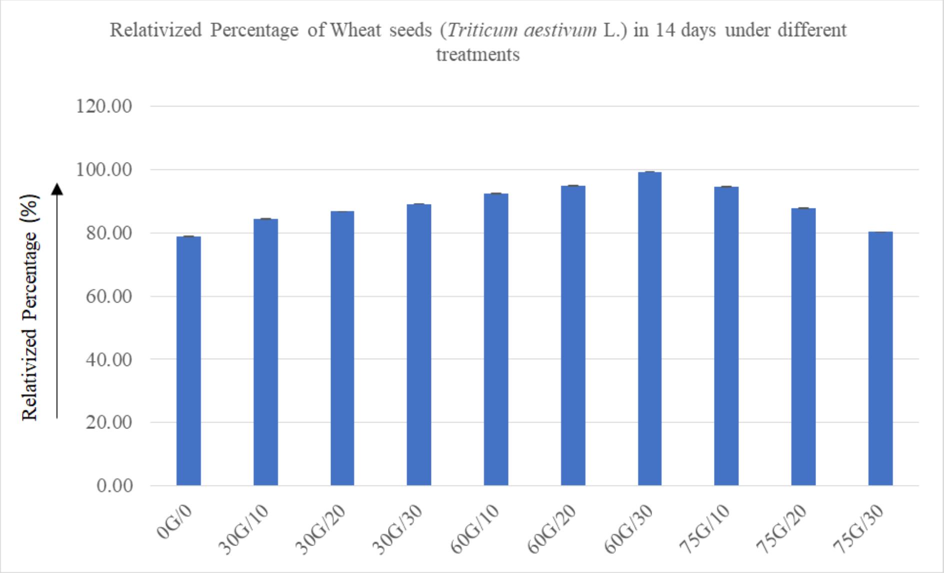

Fig. 2: Relativized percentage of Wheat seeds

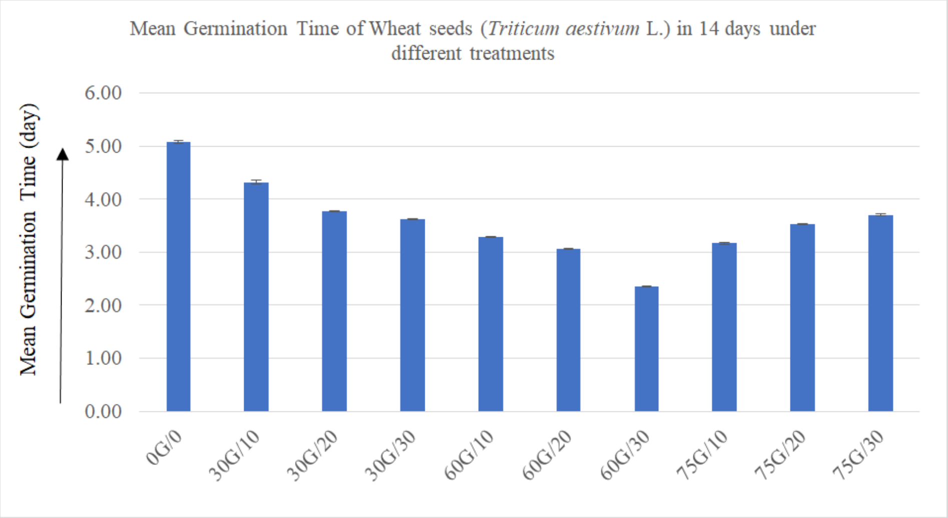

Fig. 3: Mean germination time of Wheat seeds

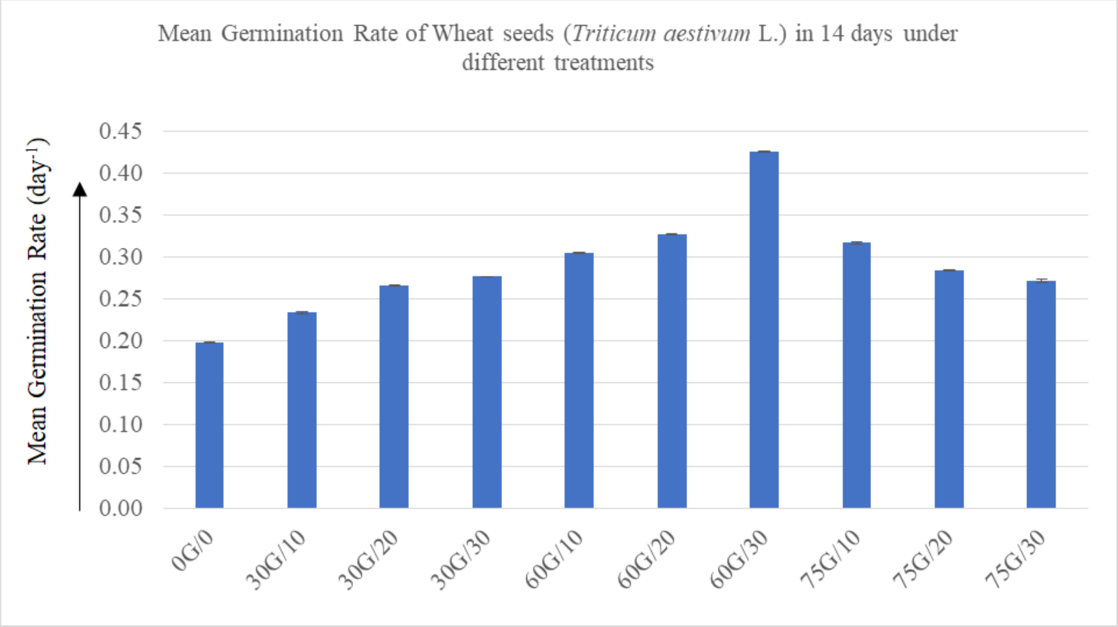

Fig. 4: Mean germination rate of Wheat seeds

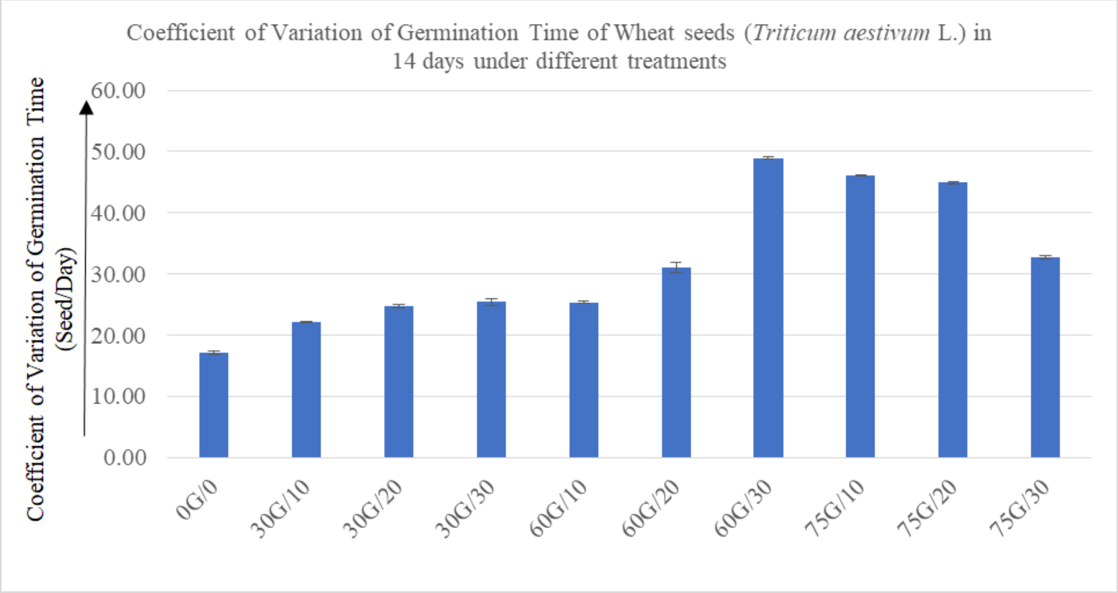

Fig. 5: Coefficient of Variation of Germination Time of Wheat seeds

Similarly in Table 4, the Germination percentage (GP) was highest at 97.33 % for seeds treated with 60 Gauss for 30 minutes followed by 93.00% for seeds treated with 60 Gauss for 20 minutes. The control had the least GP at 77.33% (Fig 6). Linearity was observed in values of the Uncertainty process (U) (Fig 7). It has been suggested that low values of Uncertainty of the germination process (*U*) have focused germination over the period. Hence all the treatment values close to zero indicate coordinated germination. The low values Synchronization Index (*Z*) across the experimental setup plants suggest that more than two seeds in the three groups finished the germination process at the same time. This verifies the experimental setup (Fig 8) (Oguis *et al*., 2022). Mean Daily Germination (MDG) was highest for seeds treated with 60 Gauss for 30 minutes than for seeds treated with 60 Gauss for 20 minutes and least for control (Fig 9).

Table 4: Estimation of "Germination percentage (G%), Uncertainty of Germination Process (U), and Synchronization Index (Z) and Mean Daily Germination Percent (MDG)" of different treatments. Different letters (a–c) among the group denote the significant differences between the mean at $p < 0.05$ using one-way analysis of variance (ANOVA), Tukey's HSD comparison test, and Scheffe's test

Applied EMF (Gauss)	Treatment Period (Minutes)	Germination percentage (G%) [%]	Uncertainty of germination process(U) [bit]	Synchronization Index (Z)	Mean Daily Germination (MDG) [%]
0	0	77.33±0.09^{f}	1.23±0.018^{bcd}	0.49±0.007^{abc}	5.52±0.007f
30	10	82.67±0.09^{de}	1.41±0.018^{bcd}	0.46±0.004^{abcd}	5.90±0.007de
30	20	85.00±0.08^{d}	1.55±0.004^{d}	0.39±0.001^{ab}	6.07±0.006d
30	30	87.33±0.05^{cd}	1.62±0.006^{d}	0.37±0.005^{a}	6.24±0.003cd
60	10	90.67±0.12^{bc}	1.63±0.014^{d}	0.35±0.000^{a}	6.48±0.009bc
60	20	93.00±0.22^{ab}	1.70±0.015^{cd}	0.33±0.004^{abc}	6.64±0.015ab
60	30	97.33±0.09^{a}	1.89±0.029^{cd}	0.32±0.006^{abc}	6.95±0.007a
75	10	92.67±0.09^{ab}	2.26±0.004^{abc}	0.21±0.001^{bcd}	6.62±0.007ab
75	20	86.00±0.08^{cd}	2.39±0.003^{ab}	0.20±0.000^{cd}	6.14±0.006cd
75	30	78.67±0.09^{ef}	2.67±0.013^{a}	0.15±0.002^{d}	5.62±0.007ef

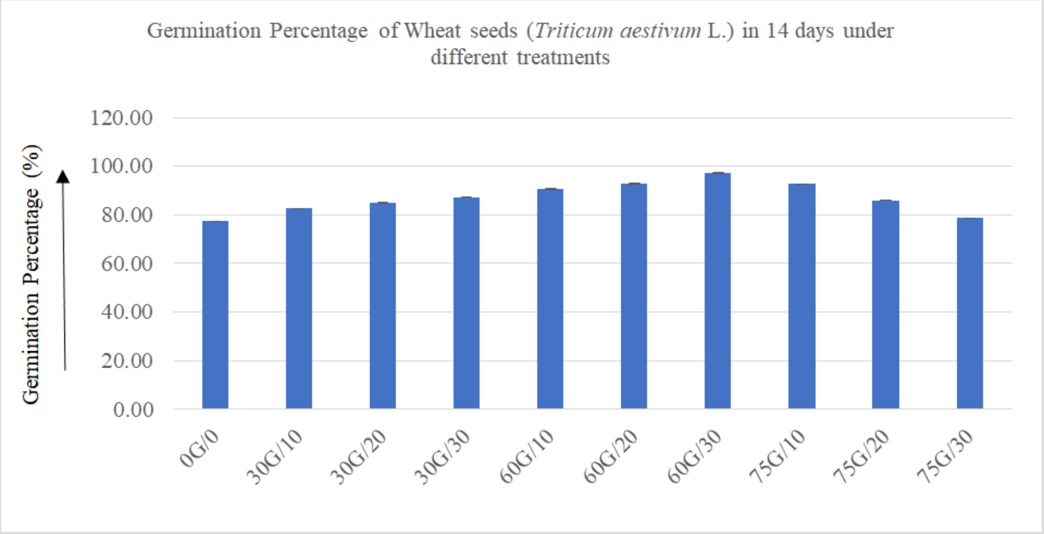

Fig. 6: Germination Percentage of Wheat seeds

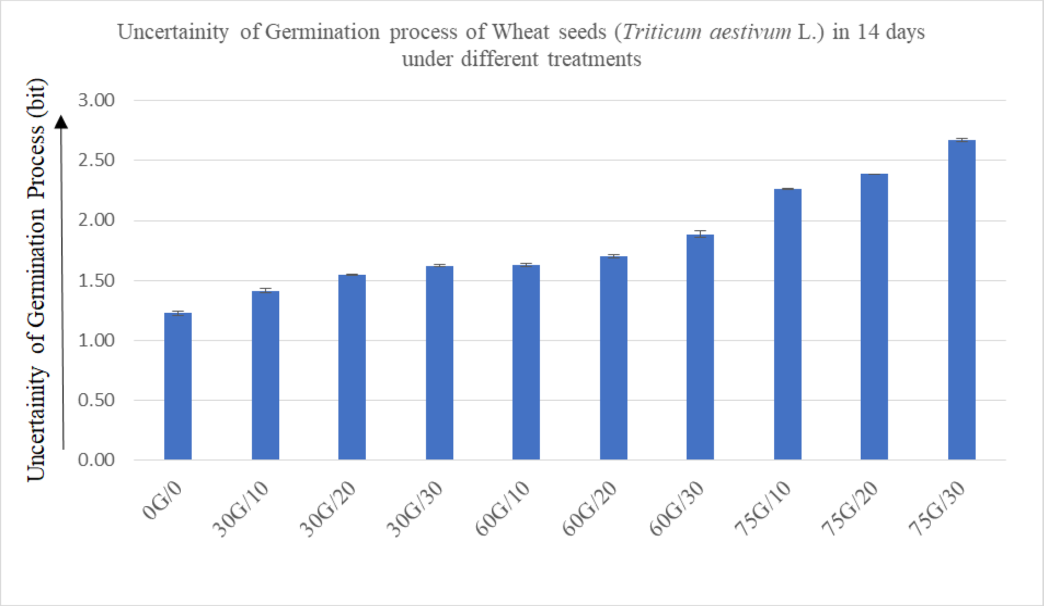

Fig. 7: Uncertainity of Germination process of Wheat seeds

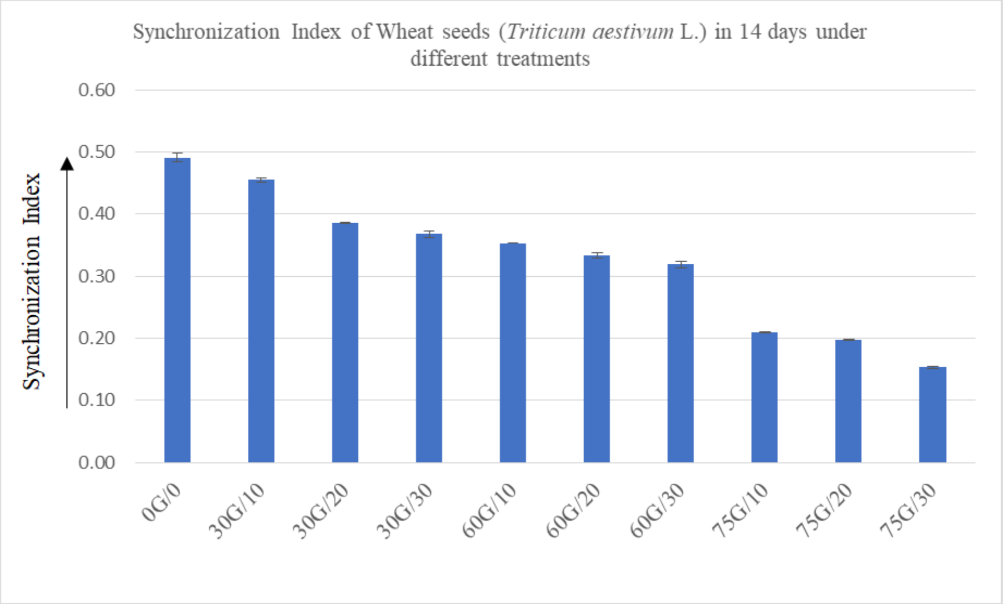

Fig. 8: Synchronization Index of Wheat seeds

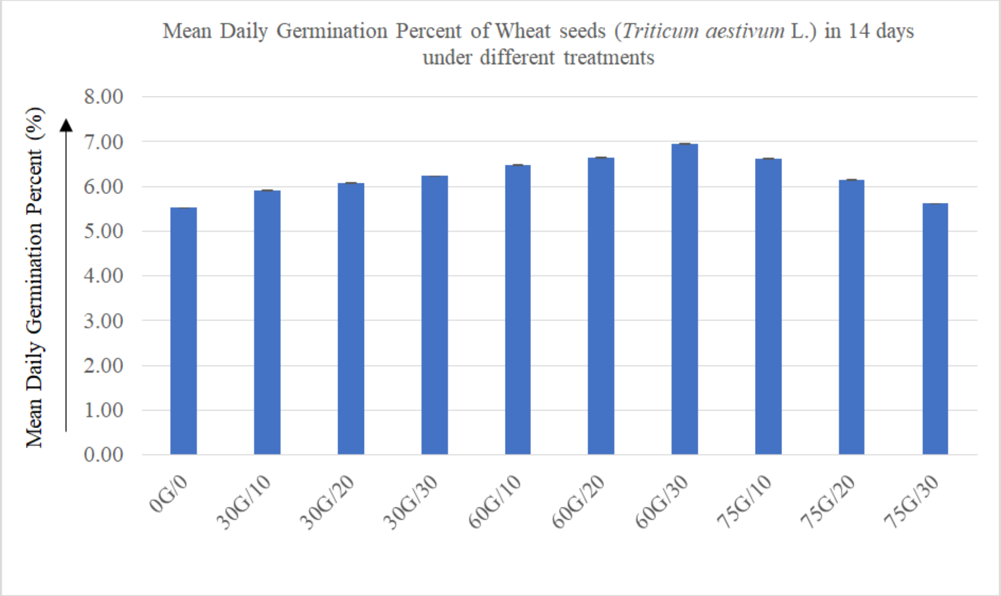

Fig. 9: Mean Daily Germination Percent of Wheat seeds

Germination Index (GI) is a measure of percentage of germination occurring at a particular period and speed of the germination process. A significant difference between experimental and control sets can be observed for GI (Fig 10). Seeds treated with 60 Gauss for 30 minutes had the highest value of 47.28 days followed by seeds treated with 60 Gauss for 20 minutes with 32.48 days. The GI value for control recorded 16.06 days. A higher GI value indicates a higher percentage and a higher rate of germination (East, C. 2020). The peak value of germination describes the germination rate of a seed lot to describe its quality (East, C. 2020). The treatment of electromagnetic field on seeds increased their quality to germinate. The order of peak value ranged from 11.58 for control seeds to 30.67 for seeds treated with 60 Gauss for 30 minutes (Fig 11). Further, Germination Value (GV) can be determined from Peak Value as it is defined as an index of combining speed and completeness of seed germination (Khalid F.,2023) Thus, higher the germination value greater is the germination process. Seeds treated with 60 Gauss for 30 minutes had the maximum value (213.1) among all the experimental conditions followed by seeds treated with 60 Gauss for 20 minutes (154.2) as compared to control (63.9) where lowest value recorded (Fig 13). The *coefficient of velocity of germination* (CVG) (Jones and Sanders, 1987) gives an indication of the rapidity of germination. Its value increases when the number of germinated seeds increases and the time required for germination decreases (Talská *et al*., 2020). Seeds treated with 60 Gauss for 30 minutes have a larger value (42.52) followed by seeds treated with 60 Gauss for 20 minutes (32.69). Control seeds (19.72) had the least value (Fig 13). This suggests that seeds treated with 60 Gauss for 30 minutes are faster in germinating.

Table 5: Estimation of "Germination Index (GI), Peak Value of Germination (PV), Germination Value (GV), and Coefficient of Velocity of Germination (CVG)" of different treatments. Different letters (a–c) among the group denote the significant differences between the mean at $p < 0.05$ using one-way analysis of variance (ANOVA), Tukey's HSD comparison test, and Scheffe's test

Applied EMF (Gauss)	Treatment Period (Minutes)	Germination Index (GI) [day]	Peak Value of germination (PV) [day^{-1}]	Germination Value (GV)	Coefficient of Velocity of germination time (CVG) [%]
0	0	16.06±0.03^{f}	11.58±0.07^{e}	63.9±0.3^{f}	19.72±0.08^{e}
30	10	20.83±0.20^{ef}	15.50±0.18^{cde}	91.6±1.1^{def}	23.30±0.19^{de}
30	20	23.53±0.05^{e}	18.42±0.03^{bcd}	111.8±0.2^{cde}	26.55±0.06^{cd}
30	30	24.79±0.03^{de}	20.75±0.09^{bc}	129.5±0.7^{bc}	27.61±0.03^{bcd}
60	10	29.25±0.12^{cd}	23.61±0.20^{b}	153.1±1.5^{b}	30.51±0.08^{bc}
60	20	32.48±0.06^{c}	23.19±0.07^{b}	154.2±0.8^{b}	32.69±0.05^{b}
60	30	47.28±0.15^{a}	30.67±0.10^{a}	213.1±0.5^{a}	42.52±0.07^{a}
75	10	39.96±0.12^{b}	19.43±0.11^{bc}	128.6±0.7^{bcd}	31.64±0.18^{bc}
75	20	34.19±0.10^{c}	16.64±0.11^{cde}	102.2±0.6^{cde}	28.33±0.06^{bcd}
75	30	30.67±0.15^{c}	13.33±0.12^{de}	74.9±0.7^{ef}	27.12±0.20^{cd}

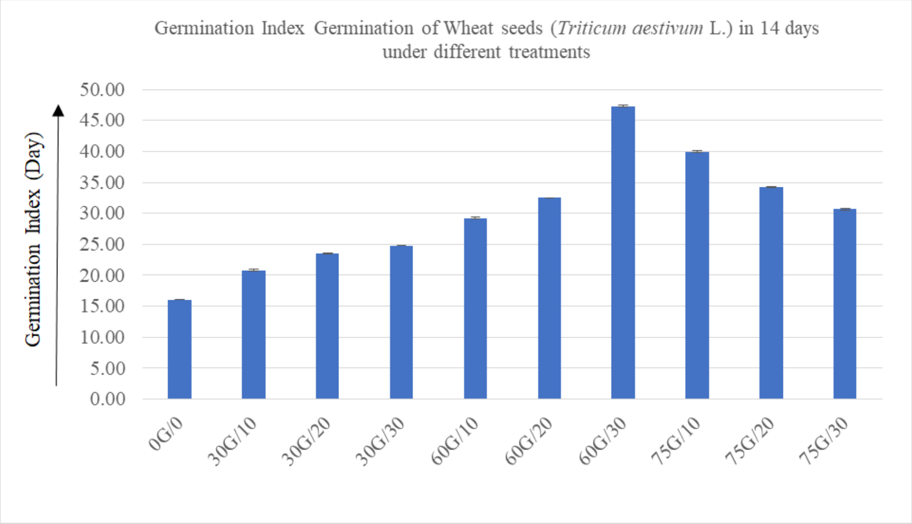

Fig. 10: Germination Index Germination of Wheat seeds

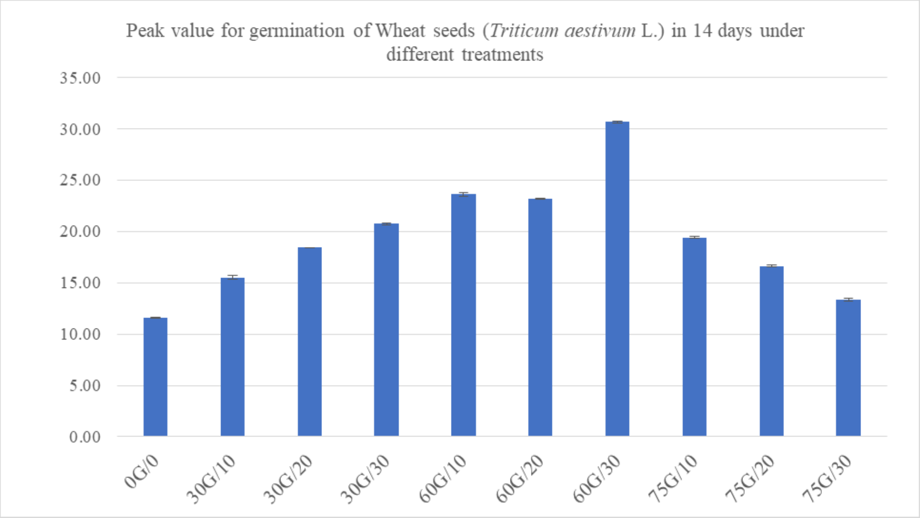

Fig. 11: Peak value for germination of Wheat seeds

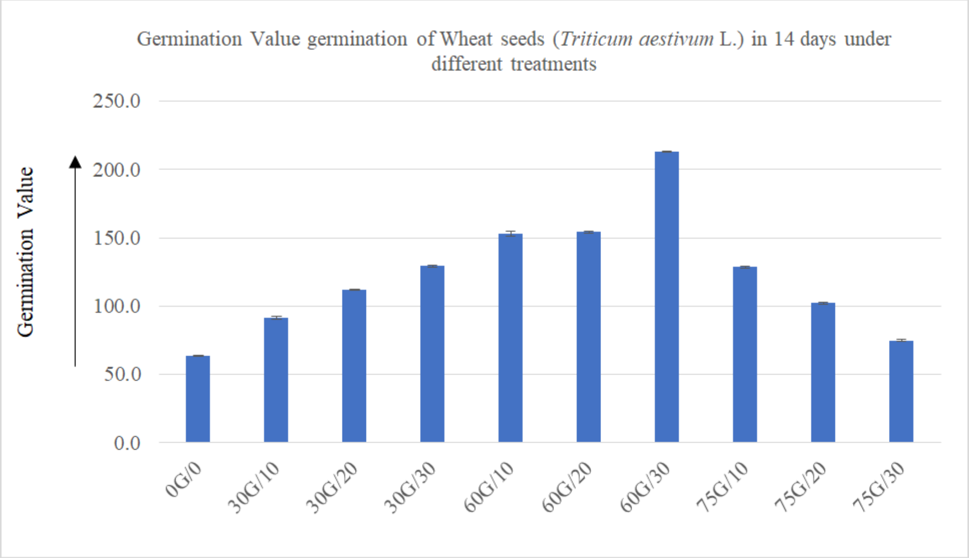

Fig. 12: Germination Value germination of Wheat seeds

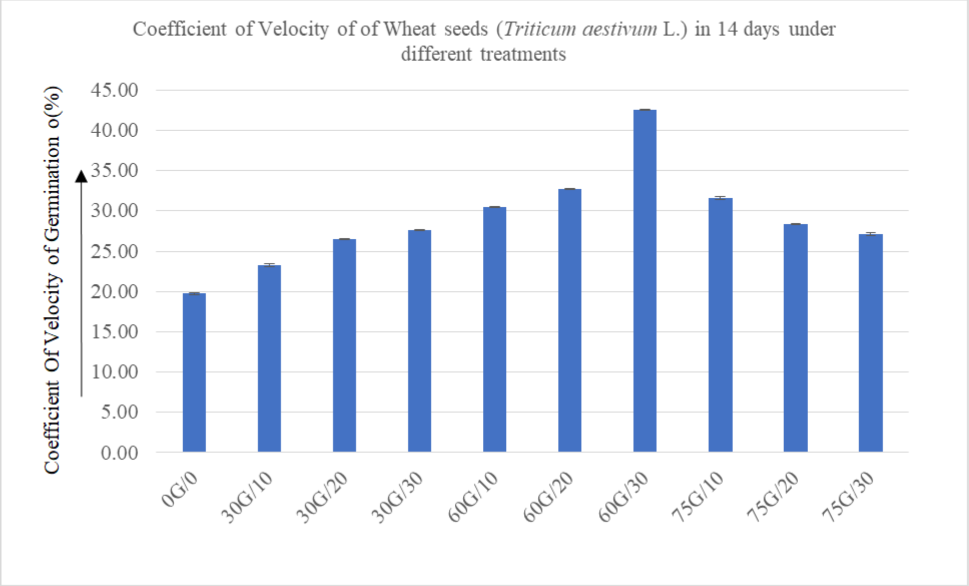

Fig. 13: Coefficient of Velocity of Wheat seeds

Table 6 showed that seeds treated with electromagnetic field germinate much faster than the control Seeds treated with 60 Gauss for 30 minutes observed to be most suited for germination process followed by Seeds treated with 60 Gauss for 20 minutes (Fig 6).

Table 6: Description of “Time to T_{10}, T_{25}, T_{75}, and T_{90} germination” of *Triticum aestivum* L. seeds

Applied EMF (Gauss)	Treatment Period (Minutes)	T_{10}(Day)	T_{25}(Day)	T_{50}(Day)	T_{75}(Day)	T_{90}(Day)
0	0	3.23±0.002	3.57±0.007	4.18±0.017	5.63±0.063	6.61±0.018
30	10	2.37±0.013	2.87±0.029	3.61±0.052	4.30±0.046	5.47±0.094
30	20	2.24±0.003	2.60±0.009	3.15±0.014	3.72±0.007	4.45±0.013
30	30	2.23±0.001	2.57±0.004	3.11±0.006	3.61±0.004	3.91±0.006
60	10	1.97±0.010	2.24±0.006	2.63±0.009	3.06±0.020	4.00±0.047
60	20	1.42±0.003	2.01±0.005	2.53±0.005	3.08±0.010	3.70±0.004
60	30	1.05±0.000	1.27±0.004	1.81±0.006	2.44±0.011	2.90±0.007
75	10	0.00	1.38±0.008	2.76±0.041	4.00±0.021	4.60±0.008
75	20	0.00	2.13±0.006	3.05±0.019	4.25±0.003	5.21±0.005
75	30	0.00	1.83±0.015	3.10±0.012	4.65±0.048	5.78±0.060

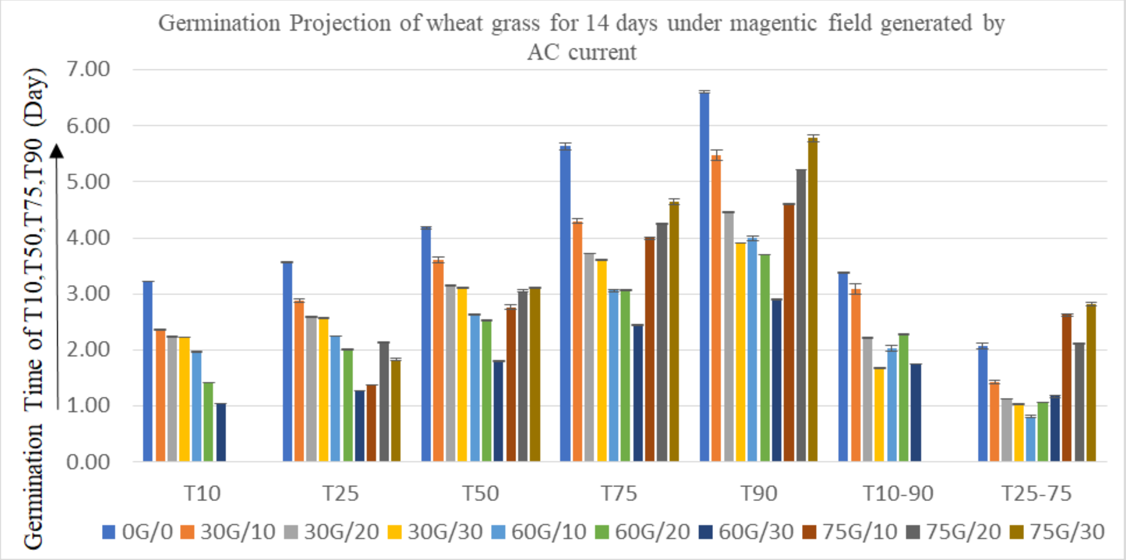

Fig. 14: Germination Projection of wheat grass

CONCLUSION

Among the experimental lot, seeds treated with 60 Gauss for 30 minutes are most suited for the germination process followed by seeds treated with 60 Gauss for 20 minutes when compared with control. These results demand further investigation at morphological, biochemical, and molecular level with the goal to search for novel biomolecules and to study the mechanism of magneto reception. The global growing population presents sustainable agriculture as the foremost challenge to combat issues like world hunger and malnutrition demanding a more environment friendly and economic measure over the current agricultural practice. Standardisation of magnetic field application protocol can further boost production and improve nutritional value of the highly valued crops to face the upcoming global challenges of hunger and malnutrition.

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