The Journal of Science and Medicine
Vol 4 No 1 (2023): The Journal of Science and Medicine
Articles

Inductively Coupled Electrical Stimulation - Part 6: Duration of PEMF-induced enhanced seed germination

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The objective of this study was to directly answer the practical question: how long lasting are the observed effects of pulsed electro-magnetic fields (PEMF) on seed germination? Earlier studies have shown that the effect of PEMF treatment on seed germination can be positive, neutral, or negative, exhibiting a tri-phasic inverse hormesis behavior as a function of total PEMF dosage during a single treatment session. This allows an optimal dosage range to be determined for maximum progerminative effect. Dosages in this range were found in earlier studies to increase the germination rate of pepper seeds to ~ 85%, from the baseline germination rate of ~56%, amounting to an increased germination rate of ~150% of baseline.

One major practical consideration of this observation is whether the PEMF treatment is durable enough to allow central processing and long-term storage of the seeds for later use, or whether seeds treated by PEMF need to be planted shortly after treatment due to rapid loss of effect over time. If the latter, what is the allowable time between PEMF treatment and planting? In addition to their practical value, these observations will have considerable theoretical value as well. As the underlying biophysical mechanism of PEMF remains to be elucidated, quantitative assessments of the durability of the effect of PEMF treatment will give important clues as to the potential mechanisms involved; for example, whether they involve the activation of enzyme systems that remain activated or taper off, whether they initiate the release or activation of substances in reserve in the germ that are exhausted upon a single use, whether multiple mechanisms are involved as evidenced by multi-phasic decay over time, or whether the progerminative effect can be repeated more than one time in any given seed.

For this study, the duration of the progerminative effect was measured by applying a previously-determined "optimal" dose of PEMF to pepper seeds, then storing the seeds in their original paper envelopes in a darkened room at 50% RH and room temperature for a period of up to one year post-treatment before planting, and determination of germination rate at two weeks (14 days). Finally, we study whether the lost progerminative effect can be recovered in stored seeds by reapplication of PEMF at the same dose immediately before planting.

Our findings were that (1) the progerminative effect lasted only about 2 days after PEMF exposure, then returned about half way back to baseline and began a slow decline thereafter, but remained at approximately half-way, never returning to baseline even for periods of storage as long as 1 year, (2) a second application of PEMF restored almost all of the progerminative effect even after storage periods of up to one year. Storage periods longer than 1 year were not studied.

These results add to the discussion of the mechanisms of PEMF across the span of complex organisms by suggesting the involvement of multiple mechanisms, some durable while others are semi-permanent.

Introduction

The effects of electromagnetism on plant growth have been the subject of academic study for about a century and a half [1]. For most of this time, the results have been inconsistent, largely due to a lack of standardization of the exact type of electromagnetism applied, which ranged from solid, fixed magnetic fields, to sinusoidal waves, to so-called “square” waves, and many other waveform types.

In most cases, both in animal and plant research with PEMF, the exact nature of the electromagnetic fields and pulses has been poorly characterized by the researchers, generally due to the omission of the key parameters of interest in favor of the less important but widely referenced parameters of “frequency” and “Gauss”. These are unfortunately not the key parameters of interest when studying the biological effects of PEMF.

Therefore, attempts to combine and generate coherent models of the range of electromagnetic effects on plant growth have been presented [2], but are of limited by the paucity of reliable and accurate information about the exact nature of the applied PEMF, which undermines the foundation upon which these models are built. Some broad generalizations do emerge however. It was generally concluded by review of this body of literature that steady magnetic fields were of dubious effect, whereas “alternating” fields tended to result in more reliably positive results. [2]

The key electromagnetic parameters for eliciting reliable biological effects in mammalian cells and tissues have been identified, and effective ranges are beginning to be elucidated [3,4]. The question then became whether or not these same parameters in the same ranges would elicit biological effects in a broader scope of living systems, including plants.

In a recent paper [5], we were able to show clear and repeatable effects of PEMF on the germination rate of various types of seeds, and were then able to show the dose-response curve for this effect. In this research project our aim has been to provide repeatable, simple, and practical information of commercial value to agriculture, both for terrestrial use, as well as for the future needs of human space exploration, where both the efficiency and effectiveness of all life-critical activities is paramount, and this includes the production of food. In our and current research, very small amounts of electrical energy (~2 W-h) were able to process more than 7,000 seeds and were subsequently shown to elicit significant increases in germination rate for some types of seeds.

In this report, we endeavor to directly answer the practical question: how long lasting are the observed effects of pulsed electro-magnetic fields (PEMF) on seed germination? Earlier studies have shown that the effect of PEMF treatment on seed germination can be positive, neutral, or negative, depending upon the dose of PEMF applied, and exhibiting a tri-phasic inverse hormesis behavior as a function of total PEMF dosage during a single treatment session. This allows an optimal dosage range to be determined for maximum progerminative effect.

In addition to the practical commercial agricultural aspects of this research endeavor, the resulting information will provide insight into the span of effects of PEMF across two of the kingdoms of complex living organisms, and to the extent that repeatable effects are observed, they must be explainable by any proposed or putative fundamental mechanisms of action of PEMF on this broad range very of living systems. The use of seeds rather than adult plants or animals affords the use of many thousands of test specimens, facilitating the collection of sufficient data to begin to fill in the detailed dose-response and time course (with terminal data points) effects of PEMF exposure. Briefly, it is a rather simple matter to measure germination rate using many thousands of seeds, each yielding clear and simple data (germination rate and time to germination), whereas any experiment on such a scale using fully-grown plants or animals would be hundreds of times more costly and difficult, with outcomes far more difficult to quantify and analyze. Therefore, from an experimental standpoint, the use of germinating seeds as a model system to probe fundamental biophysical mechanisms is superior even to the use of tiny insects such as drosophila, or flatworms such as planaria.

Having demonstrated the dose-response behavior of PEMF-treated pepper seeds [5], additional questions of practical importance to agriculture follow: “How long does the positive effect last?” and “if the effects decay over time, what is the nature of that decay?”. A long-lived effect would allow seed producers to harvest and process seeds, apply PEMF, and then store them until use months or perhaps years later. On the other hand, an effect that lasts only minutes would render the process virtually impractical for agricultural use. If the progerminative effects have a moderate duration, say for example many hours to days or weeks, then it would be necessary to process seeds shortly before planting. This might require point-of-use processing facilities, or decentralized processing if longer delays are practical. This will all depend on what the data tell us.

It is also possible that initial benefits may lead after prolonged storage to a reduction of germination below the original baseline, in which case the application of PEMF could be considered to be harmful if the seeds were not subsequently planted in the appropriate time window. This would be analogous to allowing seeds to initially sprout under conditions where there is insufficient moisture, warmth, or light to sustain growth. In the case of the effects of PEMF on seeds, this has been entirely unknown.

If the present study has positive results, it will also be of interest to determine the storage durability for other types of seed, and would be essential to know for any species before a large-scale agricultural use of this technology were undertaken. The testing however is simple to perform, and a method is proposed whereby this information can be gathered for any seed stock of interest. This report should therefore be viewed as a proof-of-principle for such specific applications.

In addition to the practical need to know the durability of enhanced progerminative effects for any particular type of seed, it is also of great scientific interest to know: is it the same or very different for different types of seed? Do we see patterns for different phyla or classes of plants? What do these patterns tell us about the fundamental mechanisms involved?

The following very simple experiment is offered as a first step in this direction.

Methods

Selection of Seed Type

This experiment is an extension of one reported earlier, in which the dose-dependent progerminative effects of PEMF were identified in Jupiter bell peppers [5]. Jupiter Bell Pepper Seeds have an intermediate germination rate, and in our experience the germination rate is only about half that typically advertised by the seed provider. Bell peppers are a relatively valuable fruit, and for this reason peppers are often the favorite choice of vegetable in food banks, for example, yielding the highest ‘value per bag’ of vegetable produce generally available. The nutritional value of peppers is also highly regarded. The seeds for peppers can be relatively expensive, so, all things considered, an improvement in the actual germination rate may have commercial importance. In earlier studies we found germination rates typically in the range of 30% to 40%, and more recently 50% to 56%, and therefore it was thought that the effect of PEMF on germination rate could be observed clearly whether the rate was increased or decreased by PEMF exposure, making this model suitable for experiments where PEMF exposure and storage time could result in either beneficial or potentially damaging effects. It is possible, for example, that a PEMF exposure protocol that would result in greater germination rate if the seeds were planted immediately might also cause a reduction in germination rate for those same seeds if they were stored for a longer period before planting. This would be expected to be the case if it turns out that the beneficial effects of PEMF when seeds are planted immediately also set into motion irreversible mechanisms that, once thwarted by failure to plant and germinate, might actually reduce the capacity to sprout at a later time. So the selection of a seed type with a germination rate of about 50% is advantageous, in order to allow any changes, positive or negative, to be evident in the data. So far as we are aware, such information is entirely absent from the scientific literature. For these reasons, Jupiter Bell Pepper seeds were selected for this study.

Experimental Conditions

The source of seeds and the method for the application of PEMF was exactly as described in detail elsewhere [5]. The PEMF dosage applied was 15 pulses per second (pps) for a duration of 75 minutes, using the same PEMF pulse waveform and at the same intensity described earlier [5]. This PEMF dosage was in the optimal range for enhanced germination rate.

Seeds were divided into three categories:

Baseline (Control, no exposure to PEMF)

Storage (Single initial exposure to PEMF with subsequent storage for various time periods)

Recharge (Initial single PEMF exposure, storage, then re-exposure immediately prior to planting)

Baseline (control) seeds were planted immediately without exposure to PEMF, **Storage** seeds were stored prior to planting for the following time periods: 23 minutes (planted “immediately”, i.e. all within 23 minutes, after PEMF exposure, which was as fast as possible under the experimental conditions), 1 hour, 2.4 hours, 6 hours, 12 hours, 1 day, 2 days, 4, 7, 14, 30, 90, 135, 180, or 360 days. **Recharge** seeds were subjected to a second PEMF exposure and then planted immediately at 90, 135, 180, and 360 days of storage after the first PEMF exposure.

Each experimental group time point consisted of 240 seeds, grouped as 8 rows of 6 columns with 5 seeds per germination cell. For statistical purposes, to estimate error, each of 8 rows was taken as a value for number of germinated cells, ranging from 0 to 30. Germination rate in each row was determined as the number of sprouted seeds per row of 30 seeds. For each timepoint, 8 rows of seeds were used to calculate the mean and standard deviation of the germination rate. Raw data will be made available for investigators who wish to analyze the data differently.

Storage conditions: the seeds were exposed to PEMF and then stored in their original paper envelopes in a darkened room at 50% relative humidity and at a temperature of 20 - 21 C for a period up to one year post-treatment prior to planting.

Seed planting, growth conditions and the determination of germination rate (determined 14 days after planting) were exactly as described earlier, using the same materials and procedures as described earlier [5].

Decisions made during the course of the experiment:

The **Recharge** seeds were actually about 1000 extra seeds that were exposed to PEMF and were held in reserve in storage, in parallel with the experiment described above, so that additional “recharge” PEMF exposures could be applied at time points not yet set at the beginning of this experiment in the event that it was determined, based upon ongoing data trends as they emerged, that a reapplication of PEMF might be necessary. As it turns out, it was decided that groups of the extra seeds would be re-exposed to a second dose of PEMF, identical to the first PEMF exposure, after long-duration storage times had shown a stable trend of partial loss of progerminative effects, if any. During the ongoing experiment, the selected time points for PEMF recharge (re-exposure) were 90, 135, 180, and 360 days. At these time points, extra seeds which had been originally exposed to a single PEMF exposure and then held in reserve were subsequently exposed to a second dose of PEMF and then planted immediately as described above. All other experimental conditions being the same, the germination rate was again determined at 2 weeks (14 days) after planting.

Results

Germination rates for **baseline** (no PEMF exposure), **storage** (single PEMF exposure followed by storage before planting), and **recharge** (exposed to an initial PEMF dosage, stored, then exposed to a second PEMF dosage immediately before planting) are shown in Table 1 and Figure 1.

Group	Days	Mean	Std. Dev.
Baseline	-	55.8	3.70
Storage	0.016	86.3	2.70
Storage	0.042	87.1	2.53
Storage	0.100	85.0	2.78
Storage	0.250	86.7	2.14
Storage	0.500	85.8	2.55
Storage	1	84.6	2.97
Storage	2	84.2	3.81
Storage	4	71.3	2.13
Storage	7	70.8	2.82
Storage	14	72.9	3.40
Storage	30	71.3	4.66
Storge	90	72.1	2.62
Storage	135	71.3	2.00
Storage	180	68.3	6.12
Storage	360	67.5	6.32
Recharge	90	84.2	2.82
Recharge	135	85.0	3.12
Recharge	180	84.6	2.56
Recharge	360	81.7	4.07

Table 1. *Mean and standard deviation germination rate (%) for each group.* For each row (timepoint) in the table, n = 8 sample groups of 30 seeds for a total of 240 seeds total per time point.

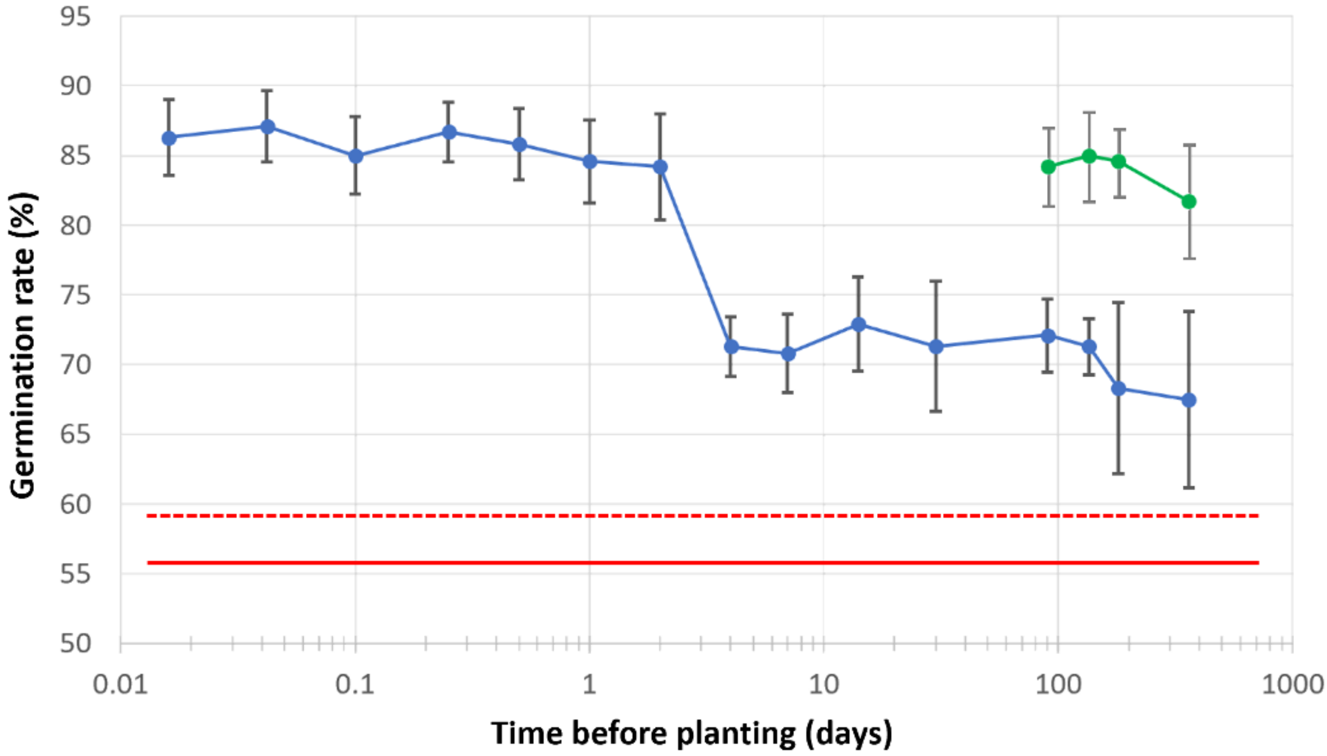

Figure 1. *Germination rates (%) are displayed on a log scale for storage time (days) after exposure to PEMF.* **Red** solid line indicates the **baseline** germination rate (55.8%), red dashed line indicates the baseline germination rate plus one standard deviation (3.70). **Blue** line indicates the germination rate for seeds **stored** for various lengths of time after a single PEMF exposure (± standard deviation). **Green** line shows the germination rate for seeds that were **recharged** with an additional exposure to PEMF immediately before planting, after having an initial PEMF exposure and then being stored for the indicated amount of time.

Discussion

When planted immediately, bell peppers exposed to PEMF as described have a substantial increase in germination rate, from about 56% to about 86%, an increase of more than 50% above baseline. But the effect is not long-lasting, dropping precipitously to from ~86% to ~ 70% somewhere between 2 and 4 days of storage after the application of PEMF. This suggests that the improvement of germination is not durable enough to be applied centrally at the point of seed production, but rather must be applied at the point of use, and certainly within about 2 days of PEMF exposure to achieve the best effect. This is a practical time constraint: the seeds need to be processed with PEMF exposure in bulk at the time of planting, within a day or two. Because the application of PEMF as described herein is simple, does not require a great deal of time, and requires only inexpensive equipment, it may be practical to scale this process for widespread agricultural use even if the PEMF must be applied at the point of use.

To determine the usefulness of this method it will be necessary to determine the durability of benefit for each particular seed type for which it is being considered. Some seeds may have large and durable effects, which would suggest different strategies for processing and storage.

It is however more likely that the benefits of this process would be better utilized when the seeds are of high individual value, when the germination rate is generally intermediate or low, or when large numbers of seeds are not generally available, such as in remote or inaccessible locations, in isolated communities, and so forth. This method might also prove especially useful for seeds after prolonged storage such as in seed banks, where in the event of a global agricultural catastrophe every seed will count.

Practical Recommendations for Implementation at Commercial Agricultural Scale

Recent history clearly indicates that it is always wise to question the claimed practical value of academic research, and to independently verify results before making a large investment. In the case of the results reported herein, these experiments can be readily applied to seeds of any type to verify the general results, to optimize the PEMF parameters to be applied, and then to estimate the potential commercial value of this method for each type of seed. The procedure is not costly in terms of equipment or supplies.

The following procedure is recommended:

1. Determine the approximate dose-response curve for the specific seeds to be planted [5].
2. Identify the optimal dosage range [5], test more dosages if necessary.
3. Arrange a brief version of the experiment described above to identify the time of persistence of the progerminative effect.

This procedure will allow the determination of the maximal beneficial effect to be expected, the PEMF dosage range to be applied to achieve the desired effect, and the time allowed between PEMF exposure and planting. With these numbers, the potential value of the PEMF treatment can be calculated, and the practical limitations appreciated, such as the required PEMF processing and handling time, achievable processing volume/rate, and necessity to provide additional equipment to achieve the desired scale.

Conclusion

When applied as described, PEMF can improve germination rate of bell pepper seeds by approximately 50% greater than baseline. This effect persists for about 2 days after PEMF exposure, but then the benefit drops by about half within 4 days after exposure and persists thereafter at about that level for at least a year. Re-exposure to PEMF can again restore the progerminative effect if the seeds are promptly planted after the second PEMF treatment.

The practical agricultural use of this technique at scale should take into account the commercial value of the seeds in question, the persistence of the beneficial effect of PEMF for each particular seed type, and the ability to process and store seeds at the point of use rather than centrally. It is not known if the results described here can be generalized to seed types other than bell peppers. Therefore, a simple course of experiments is recommend to optimize the beneficial effects of PEMF for improved seed germination rate for each type of seed being considered.

Statement of Potential Conflict of Interest

The author of this report (R.G. Dennis) declares both a scientific and a commercial interest in ICES®-PEMF technology: He is owner of Micro-Pulse LLC (manufacturer of the technology), holds several patents for ICES®-PEMF technology and receives royalty payments from NASA-Johnson Space Center for the commercial licensing of this technology, which he developed in its initial form (TVEMF) as a consultant for NASA in the mid-1990’s.

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