Phyton-
International Journal of
Experimental Botany

Tech Science Press

http://doi.org/10.32604/phyton.2026.084457

ARTICLE

Electromagnetic-Field Priming of Broccoli Seeds under Water and Salt Stress: Effects of Exposure Regime and Duration

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Received: 23 April 2026; Accepted: 14 July 2026; Published: 28 August 2026

ABSTRACT: Water deficit and soil salinity can severely impair seed germination and crop establishment, creating a need for sustainable, non-chemical approaches to improve seed performance under stress. Electromagnetic-field (EMF) priming is a promising physical treatment; however, its effectiveness may depend strongly on the exposure regime and duration. This study evaluated the effect of electromagnetic-field (EMF) priming on broccoli (*Brassica oleracea*) seed germination under water and salt stress. Seeds were exposed to one or two EMF exposure regimes for different durations and were assessed through germination tests, digital image analysis, photoacoustic spectroscopy and FTIR spectroscopy. EMF priming produced treatment-dependent responses. A short exposure of 7.5 min under one exposure regime improved germination under water stress by 15% at 36 h compared with untreated control. Longer exposures increased the photoacoustic signal associated with carotenoid-related absorption bands, particularly after 120 min, whereas FTIR analysis showed reduced absorbance in O-H bands at 1600 and 3400 cm^{-1}, indicating changes in seed moisture-related properties. However, not all EMF treatments improved germination, and longer or repeated exposures sometimes produced neutral or negative effects. These results suggest that EMF priming may enhance broccoli seed performance under stress only within specific exposure windows, highlighting the need to optimize treatment duration and exposure regime before practical application.

KEYWORDS: Electromagnetic-field priming; broccoli seeds; water stress; salt stress; germination; FTIR spectroscopy; photoacoustic spectroscopy

1 Introduction

Abiotic stresses such as drought and soil salinity are among the main factors limiting crop establishment and productivity [1]. Salinity stress can impair seed germination and early seedling growth by altering water status, pigment content and oxidative balance [1]. These stresses negatively affect seed germination and early plant development, reducing agricultural yields worldwide. For this reason, the search for sustainable strategies to improve crop performance under adverse environmental conditions has become increasingly important.

Seed conditioning, also known as seed priming, has emerged as an effective alternative to enhance germination and seedling vigor. Different priming methods have been developed, including hydropriming, magnetopriming, osmopriming, nutripriming and biopriming [2,3]. These techniques have demonstrated beneficial effects on seed physiological quality, improving attributes such as germination rate, vigor and water uptake. However, the response to priming depends on the treatment type, concentration and plant species [4].

Conventional agrochemicals can adversely affect environmental quality and human health [5]. Consequently, biological and biophysical approaches have gained attention as potentially more environmentally friendly alternatives [6].

Among the physical methods, magnetic and electromagnetic field treatments have shown promising results as a non-chemical tool for seed stimulation. Several authors have reported that magnetic field exposure can improve germination and seedling development, particularly under water and salt stress conditions. For example, Putri et al. [7] observed enhanced germination in rice seeds treated with a magnetic field under drought stress. Similarly, recent evidence in *Rubus fruticosus* seeds showed that static magnetic field exposure may contribute to germination-associated metabolic activation through changes related to starch mobilization and amylase activity [8].

In addition to physiological evaluations, spectroscopic techniques have been widely used for seed characterization. Photoacoustic spectroscopy (PAS) is a non-invasive and non-destructive method that allows the analysis of optical and biochemical properties of biological materials [9]. Fourier transform infrared spectroscopy (FTIR), although destructive, provides valuable information about molecular functional groups and structural changes occurring in seeds [10]. FTIR-based analyses have been used to monitor seed deterioration and germination-related changes, while seed moisture content is an important determinant of germination performance [11,12].

Electromagnetic-field treatments may influence seed germination through several interconnected physiological and biochemical processes. During early imbibition, changes in membrane permeability and ion transport can affect water uptake, solute balance and metabolic reactivation. EMF exposure may also modulate enzyme activity, respiratory metabolism and reactive oxygen species signaling, which are involved in reserve mobilization and stress responses during germination. However, these effects are expected to depend strongly on exposure intensity, duration and regime, since excessive exposure may disturb hydration balance or induce stress-like responses. Therefore, identifying an optimal exposure window is essential for interpreting the biological effect of EMF priming.

In this context, the present study aimed to evaluate whether EMF priming improves broccoli (*Brassica oleracea*) seed germination under water and salt stress and to identify the exposure regimes associated with beneficial, neutral or inhibitory responses. We hypothesized that short EMF exposures would enhance early germination under stress by promoting hydration-related and metabolic processes, whereas longer or repeated exposures could reduce seed performance by altering moisture-related properties. Germination performance was assessed together with physical, spectroscopic and imaging analyses to characterize seed responses to EMF priming.

2 Materials and Methods

2.1 Biological Material

Broccoli (*Brassica oleracea*) sprouting seeds of commercial brand from Barcelona, Spain were used in this research. The data sheet stipulates that they were packaged in May 2023, expiry date December 2028, lot number 86492CIN0DS. With certification ES-ECO-019-CT EU Agriculture. The roundness index was calculated by dividing the polar diameter by the equatorial diameter, following a general morphometric approach used for seed physical characterization [13].

Seed Humidity

The humidity percentage of the seed was obtained using an OHAUS model MB23 thermobalance. 1.5 g of seeds were placed for 10 min at a temperature of 100°C. 3 replicates of each treatment were carried out.

2.2 Electromagnetic Priming

The electromagnetic exposure device consisted of a 255-turn coil made of 15-gauge copper wire (1.4 mm in diameter), with a length of 44.5 cm and a diameter of 20 cm. The coil was energized through a transformer connected to a 120 V, 50 Hz AC power supply. The experimental setup also included a heat sink for thermal dissipation. During EMF treatment, a current of 5 A was supplied to the coil.

The magnetic field strength (β) of the coil was measured using a Lakeshore 410 gaussmeter to obtain the radiation pattern. Fig. 1 shows the instrumentation of the coil device (a) and its radiation pattern (b) for the electromagnetic priming (EMF priming) of seeds, the highest electromagnetic field strength is in the center of the coil and decreases when approaching the edges of the coil.

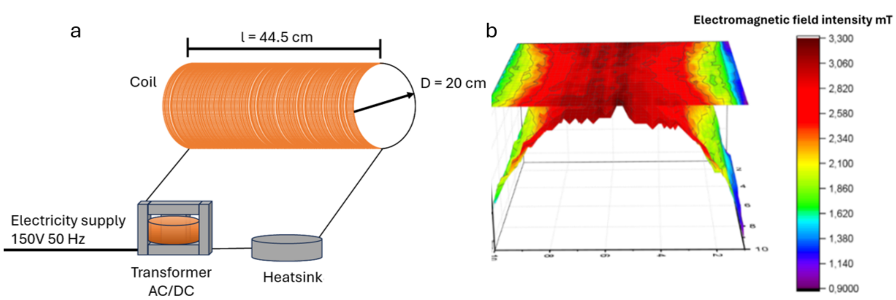

Figure 1: (**a**) Instrumentation of the coil for the treatment of seeds with electromagnetic field according to Domínguez and Hernández [14]; (**b**) radiation pattern.

2.3 Experimental Design and Treatment Structure

The seeds were hydrated for two hours in distilled water before priming by electromagnetic field. The seeds were placed in the center of the coil $\beta = 3.18 \pm 0.12$ mT generated by a current I = 5 A.

Table 1 summarizes the treatment structure, including the experimental phase, EMF exposure regime, exposure duration, water stress (WS) and saline stress condition (SS). The experimental design was organized into four phases. (1) EMF time of 7.5 and 15 min with 1 radiation regime (1R); (2) EMF time of 60 and 120 min with 1 radiation regime (1R); (3) EMF time of 0, 7. 5 and 15 min (2R); (4) EMF time of 60 and 120 min (2R).

After EMF priming, seeds were subjected to three germination conditions: irrigated with distilled water (5 mL), water stress solution and salt stress solution. The water stress solution was made with water and 143 g/l PEG 6000 (Polyethylenglycol, P.M. 6000) to generate an osmotic potential of −0.33 MPa according to Michel and Kaufman [15] (Eq. (1)).

$$\psi s = -(1.8 \times 10^{-2})C - (1.8 \times 10^{-4})C2 + (2.67 \times 10^{-4})CT + (8.39 \times 10^{-7})C2\ T \quad (1)$$

where:

ψs = Osmotic potential

C = concentration de PEG 6000 g/l H_2O

T = temperature°C

Salt stress was induced using a 1.5 g L^{-1} NaCl solution, corresponding to approximately 25.7 mM (0.0257 M) NaCl. This concentration was selected to impose a tolerable level of salt stress on the seeds [16].

Table 1: Electromagnetic pre-conditioning applied to broccoli seeds. WS: water stress, SS: salt stress, control indicates non-exposed seeds; EMF1 and EMF2 indicate the first and second exposure durations within each phase.

Treatment	Condition	Treatment	Condition
Phase 1		**Phase 2**	
T0	Control	T0	Control
T1	WS	T1	WS
T2	SS	T2	SS
T3	EMF1	T3	EMF1
T4	EMF1 + WS	T4	EMF1 + WS
T5	EMF1 + SS	T5	EMF1 + SS
T6	EMF2	T6	EMF2
T7	EMF2 + WS	T7	EMF2 + WS
T8	EMF2 + SS	T8	EMF2 + WS
Phase 3		**Phase 4**	
T0	Control	T0	Control
T1	WS	T1	WS
T2	SS	T2	SS
T3	EMF1	T3	EMF1
T4	EMF1 + WS	T4	EMF1 + WS
T5	EMF + SS	T5	EMF + SS
T6	EMF2	T6	EMF2
T7	EMF2 + WS	T7	EMF2 + WS
T8	EMF2 + SS	T8	EMF2 + SS

The temperature evolution of the seeds before and after being subjected to EMF priming was also determined. A FLIR infrared camera (Systems Wilsonville, OR, USA) model i5; 6.8 mm lens; accuracy ±2%; thermal sensitivity <0.1 at 25°C; thermal image resolution 140 × 140 and temperature range −20 to 250°C was used.

2.4 Germination Test

The germination test was performed according to the recommendations of the International Seed Testing Association [17]. For this test, filter paper was placed in the Petri dishes, and 25 seeds were sown in each one of them. Four replicates per treatment were carried out following a randomized complete block experimental design. The seeds were placed in darkness at 25°C and 73% humidity level. Counts of germinated seeds were carried out every 12 h until the fifth day, when germination counts reached a stable level.

2.5 Seed Characterization Using Digital Imaging

Digital seed image characterization was carried out with a high-resolution scanner (4800 dpi) HP Scanjet G4050 model. A total of 120 images (RGB) of seeds corresponding to 10 replicates of each EMF priming were obtained. A Gaussian filtering was applied to the images to homogenize on the grey scale. The MATLAB R2024b software was used to obtain data matrix of the grayscale images. The MATLAB data were exported to OriginPro 2017 software where an arithmetic XY transformation was performed to obtain the Z-axis values only for the region of interest (seed). The data were sorted considering the Z-axis as the signal intensity. This was smoothed with the Fast Fourier Transform (FFT) to make a representation of the data. Finally, the average of the signals of the images of each treatment was obtained and the data was plotted. Fig. 2 shows the instrumentation used to obtain the digital images of the seeds.

Digital image analysis was used as a complementary method to detect optical and surface-related changes in the seed coat after EMF priming. The image-derived signal should be interpreted as an indirect indicator of changes in seed reflectance, texture or moisture distribution, rather than as a direct physiological measurement of germination potential.

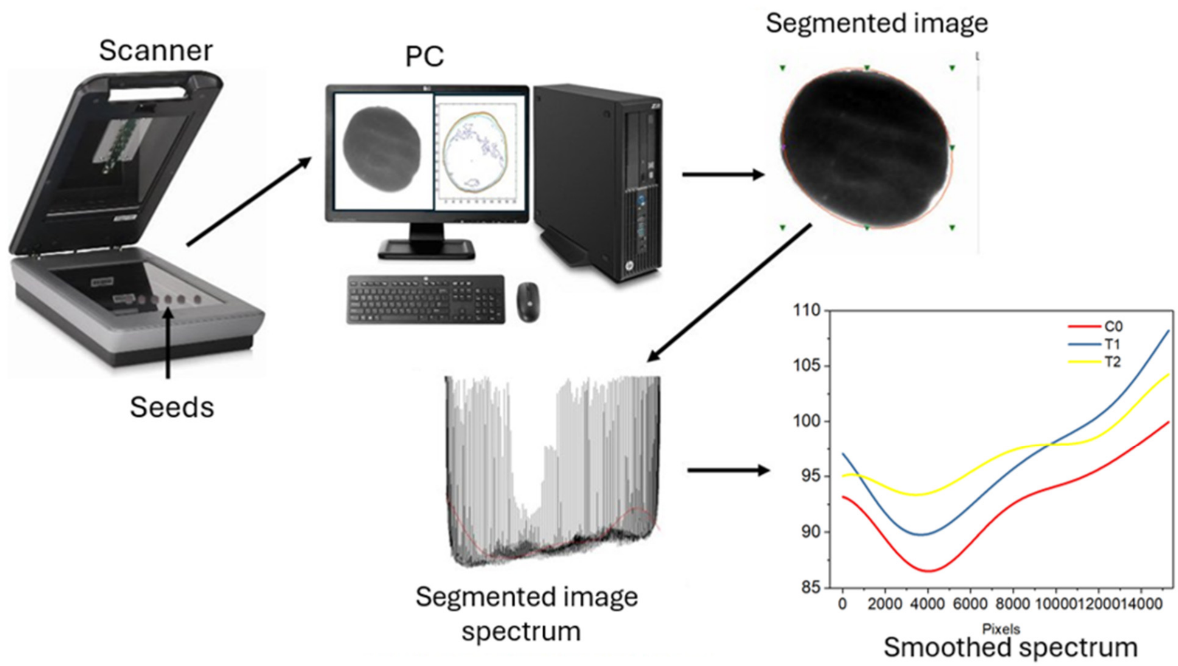

Figure 2: Process for obtaining digital images and analyzing the obtained signal.

2.6 Photoacoustic Spectroscopy

Photoacoustic (PA) spectra were obtained using the photoacoustic spectroscopy (PAS) technique at the physics laboratory of CINVESTAV Mexico City. The seeds were placed inside the PA cell hermetically sealed with silicone grease. The PA spectra was obtained in the range of 250 to 750 nm generated by a 700 W Xenon lamp. The light passed through a monochromator to split the wavelengths and modulated with a chopper at 17 Hz. The signal obtained was amplified by means of a lock-in amplifier and the data were stored on the computer [18].

2.7 Infrared Spectroscopy (FTIR)

Seeds were pulverised into flour to obtain a more homogeneous sample. This was mixed with KBr to make tablets for analysis.

The samples were analyzed using a NICOLET FTIR spectrometer, model 6700 (resolution: 2 cm^{-1}, 200 scans). The FTIR spectrum was measured in transmission mode using KBr disks at room temperature.

2.8 Statistical Analysis

Data was analyzed using analysis of variance (ANOVA). For germination percentage, treatment, germination time and their interaction were considered within each experimental phase. For seed temperature, moisture content, digital image-derived signal intensity and photoacoustic signal intensity, treatment was considered as the main factor. When significant differences were detected, means were compared using Tukey's test at $p \leq 0.05$. Data are presented as mean ± standard error. Statistical analyses were performed using SAS, MATLAB R2024b and OriginPro 2017.

Because the experiment was conducted in four independent phases, each phase was analyzed separately using its corresponding control. Direct statistical comparisons between short-exposure phases and long-exposure phases were not performed. Therefore, comparisons between short and long exposure durations are discussed only as descriptive trends.

To improve clarity, the experimental workflow was organized as follows:

Seed hydration in distilled water for 2 h → EMF priming at $\beta = 3.18 \pm 0.12$ mT → exposure regime selection: one exposure regime (1R) or two exposure regimes (2R) → exposure duration: 0, 7.5, 15, 60 or 120 min depending on the experimental phase → stress condition: no stress, water stress induced by PEG 6000, or salt stress induced by NaCl → seed evaluation through germination test, temperature and moisture determination, digital image analysis, photoacoustic spectroscopy and FTIR spectroscopy.

3 Results

Table 2 shows the physical characteristics: polar diameter, equatorial diameter, roundness index, thickness and weight of 100 seeds.

Table 2: Physical characteristics of the seed.

Polar Diameter (mm)	Equatorial Diameter (mm)	Roundness Index (mm)	Thickness (mm)	Weight of 100 Seeds (g)
0.43	1.95	1.78	1.09	0.33

3.1 Germination Test

Fig. 3 shows the germination response of broccoli seeds exposed to short EMF priming times under one and two exposure regimes. In phase 1 (7.5 and 15 min; 1R), significant differences among treatments were observed at 36, 48, 60 and 72 h. Short EMF exposure produced the most favorable response under specific conditions (Fig. 3a). At 36 h, T6 increased germination by 15% compared with T0, whereas T8 showed lower germination under salt stress. At 48 h, T4 showed the highest positive response, increasing germination by 15% compared with the control. These results suggest that short EMF exposure, particularly 7.5 min under water stress, may improve early germination.

In phase 3 (7.5 and 15 min; 2R), significant differences were observed at 36, 48, 60, 72 and 84 h. Two exposure regimes accelerated early germination at 36 h, especially in T3 and T6 (Fig. 3b). However, this initial advantage was not maintained consistently at later counting times. At 48 h, T3 increased germination, whereas T6 showed a decrease compared with T0. At 60 h, positive responses were observed in T6 and T4, but under salt stress T8 showed lower germination at 72 and 84 h. Overall, phases 1 and 3 indicate that short EMF exposures may promote early germination, but repeated exposure was not consistently beneficial, particularly under salt stress.

Overall, EMF priming produced treatment-dependent effects on broccoli seed germination. Short exposure times improved early germination under specific stress conditions, whereas longer exposures, particularly under two exposure regimes, produced neutral or negative effects. The clearest positive response was observed after 7.5 min under one exposure regime in seeds subjected to water stress.

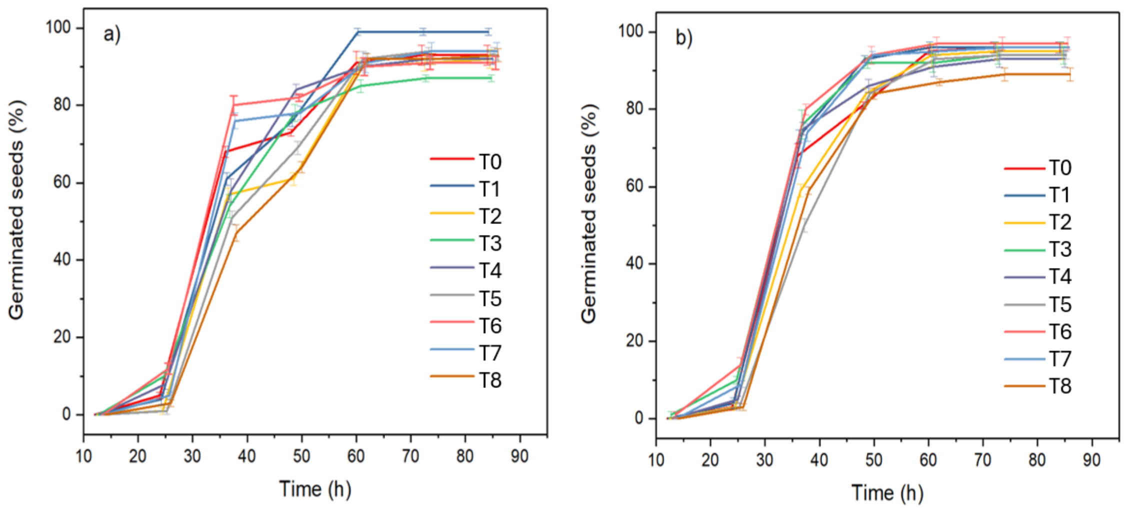

Figure 3: Germination percentage of broccoli seeds subjected to EMF priming under phases 1 and 3. Values represent mean ± SE: (**a**) Phase 1 (7.5 and 15 min; 1R) and (**b**) Phase 3 (7.5 and 15 min; 2R).

Fig. 4 shows the germination response of broccoli seeds exposed to long EMF priming times under one and two exposure regimes. Fig. 4a shows in phase 2 (60 and 120 min; 1R), significant differences among treatments at 12, 36, 48 and 60 h. The clearest negative responses were associated with salt stress, particularly in T5 and T8, which showed lower germination than the non-stressed control during early germination. At 36 h, the salt-stressed control also showed reduced germination compared with T0, indicating that salinity strongly affected early seed performance.

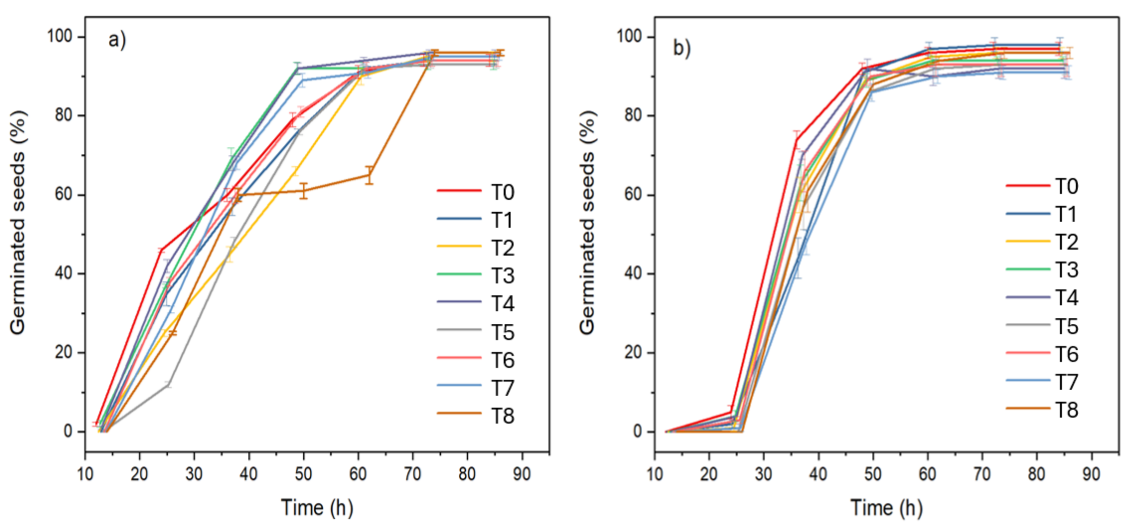

Figure 4: Germination percentage of broccoli seeds subjected to EMF priming under phases 2 and 4. Values represent mean ± SE: (**a**) Phase 2 (60 and 120 min; 1R) and (**b**) Phase 4 (60 and 120 min; 2R).

In phase 4 (60 and 120 min; 2R), EMF priming produced variable effects depending on exposure duration and stress condition (Fig. 4b). At 48 h, T3 increased germination by 16.45% compared with T0, whereas T8 decreased germination by 22.78%. At later counting times, T8 continued to show lower germination than the control, suggesting that long exposure under two regimes was not beneficial under salt stress. Water-stressed treatments also showed reduced germination at early counting times, particularly T7 and T1. Overall, phases 2 and 4 indicate that long EMF exposure did not consistently improve germination and that repeated exposure may reduce seed performance under stress conditions.

Final germination responses also differed among phases and treatments. In phases 2 and 4, the highest final germination values were not always associated with EMF priming, confirming that long exposures did not produce a consistent beneficial effect. In phases 1 and 3, short exposure treatments produced higher early germination in some cases, but the final response depended on the stress condition and exposure regime. Therefore, EMF priming mainly affected the timing and early dynamics of germination rather than producing a uniform increase in final germination across all treatments.

3.2 Seed Characterization by Digital Imaging

Digital optical images and software-processed images of broccoli seeds were obtained for each EMF-priming treatment in all experimental phases. The images were processed to generate smoothed signal-intensity profiles. Fig. 5 shows the smoothed signal intensity obtained using Fast Fourier Transform (FFT)-based processing as a function of pixel position for each EMF-priming treatment.

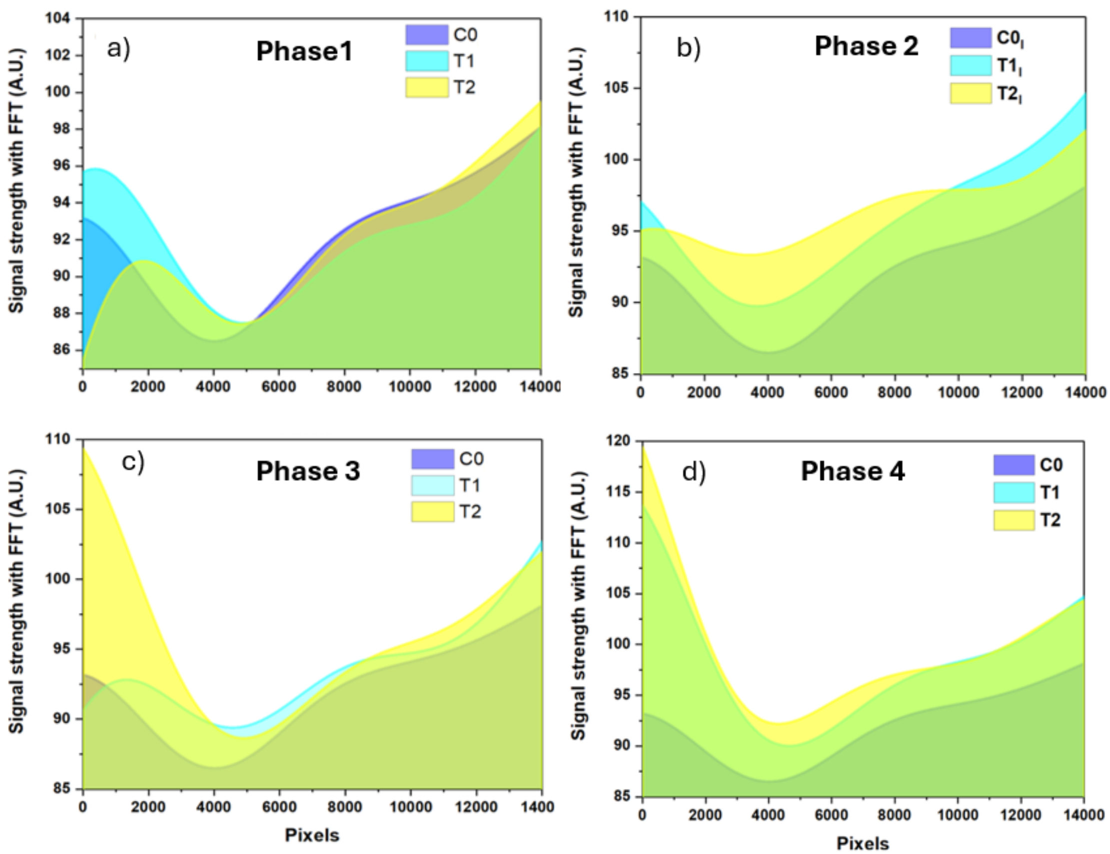

Figure 5: Characterization of broccoli seed using digital images phase 1 (**a**), 2 (**b**), 3 (**c**) and 4 (**d**).

In phases 1 and 3, there was no significant difference at shorter radiation times, although there was a tendency for the signal intensity to increase or decrease. In phase 1 the 2000-pixel range T1 and T2 preconditioning increased to 4.1% (93.15 ± 0.06) and 1.5% (90.81 ± 0.07) in 4000 1.8% (88.14 ± 0.08) and 1.6% (87.95 ± 0.07). In the 8000 range, it decreased (by 1.3% (91.33 ± 0.07) and by 0.3% (92.27 ± 0.08)). In T1 at 12,000, there was a decrease of 1.4% (94.29 ± 0.07), while in T2, there was an increase of 0.5% (96.21 ± 0.09) compared to control (see Fig. 5a). Finally, in the phase 3 T1 and T2 within the same pixel ranges (2000, 4000, 8000, and 12,000), there were increases of 3.3% (92.38 ± 0.06), 9.7% (98.12 ± 0.08), 3.5% (89.60 ± 0.04), 3.4% (89.51 ± 0.11), 1.2% (93.73 ± 0.06), and 0.8% (93.36 ± 0.09) and 1.2% (96.85 ± 0.12) and 2.3% (97.86 ± 0.10), respectively, compared to control (Fig. 3b).

Fig. 5b shows phase 2 treatments T1 (60 min-1R) and T2 (120 min-1R) which had an increase in signal intensity in 2000-pixeles rangeT1 2.57% (91.72 ± 0.06) and 5.21% (94.09 ± 0.06) in 4000 3.8% (89.83 ± 0.03) and 8% (93.48 ± 0.02) with respect to control. An increase in signal intensity of 3.4% (95.71 ± 0.08) and 5.2% (97.38 ± 0.03) was also observed for T1 and T2 in the 8000 range. At 12,000 pixels T1 and T2 increased by 5% (100.50 ± 0.08) and 3.1% (98.68 ± 0.06) with respect to control.

Similarly, on Fig. 5d there was an increase in signal intensity in 2000, 4000, 8000 and 12,000 pixels. The phase 4 T1 (60 min-2R) and T2 (120 min-2R) increased in 2000 11.6% (99.88 ± 0.07) and (100.97 ± 0.07), in 4000 by 4.6% (90.56 ± 0.10) and 6.7% (92.31 ± 0.04), in 8000 by 3.7% (96.01 ± 0.10) and 4.8% (97.02 ± 0.04) and 12,000 by 5% (100.45 ± 0.09) and 5.2% (100.66 ± 0.10) when compared to control.

3.3 Seed Temperature and Humidity

Fig. 6 shows the evolution of temperature and the change in humidity content of broccoli seeds when treated with electromagnetic field at different radiation times. The initial seed temperature was 20 ± 0.14°C at 0 min, and an increase in temperature was observed at all EMF priming times. A statistically significant increase ($p \leq 0.05$) was obtained at the end of the exposure times of 7.5, 15, 60 and 120 min, with increases of 12.5% (22.5 ± 0.2°C), 15% (23 ± 0°C), 25.5% (25.1 ± 0.3°C) and 30.05% (26.07 ± 0.1°C) compared to the initial temperature at 0 min. These temperature changes occurred during EMF exposure and should not be interpreted as differences in germination temperature, since all germination tests were conducted under controlled conditions at 25°C.

In relation to the percentage of humidity of the seeds, a value of 98.66% (0 min) was observed at the beginning. When the seeds were treated with the electromagnetic field, the humidity decreased significantly ($p \leq 0.05$), with a decrease of 0.65% for one regime (1R) (7.5, 15, 60 and 120 min). For 2 regimes it decreased 1.33% (7.5 and 15 min) and 2% (60 and 120 min) when compared to the initial humidity.

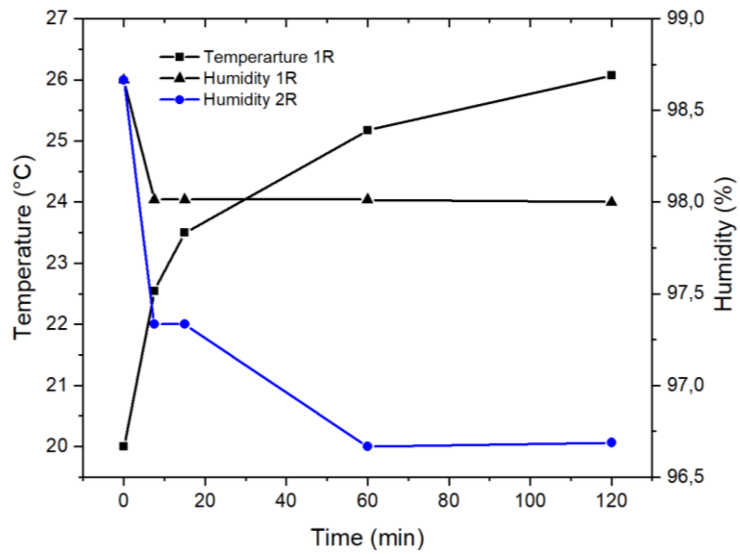

Figure 6: Evolution of temperature and decrease in seed humidity EMF priming.

3.4 Photoacoustic Spectroscopy of Seed

Fig. 7 shows the photoacoustic spectra in the 250–750 nm range of broccoli seeds conditioned by electromagnetic field (60 and 120 min) in regime 1 (1R) and regime 2 (2R). These show a similar pattern of behavior with an absorbing band of 330–560 nm. Fig. 7a, b shows the range of the photoacoustic signal intensity which oscillates between 0.052–0.146 mV and 0.0785–0.1862 mV corresponding to regime 1 and regime 2, respectively. The highest PA signal intensity levels were 0.146 and 0.1862 mV in T2 at phase 2 and 4 (120 min) preconditioning increasing 19.93 and 8.95% when compared to control. On the other hand, the lowest PA signal intensities were 0.0522 and 0.0785 mV in the T1 and T1 (60 min) EMF priming decreasing by 4.92 and 5.08% when compared to control.

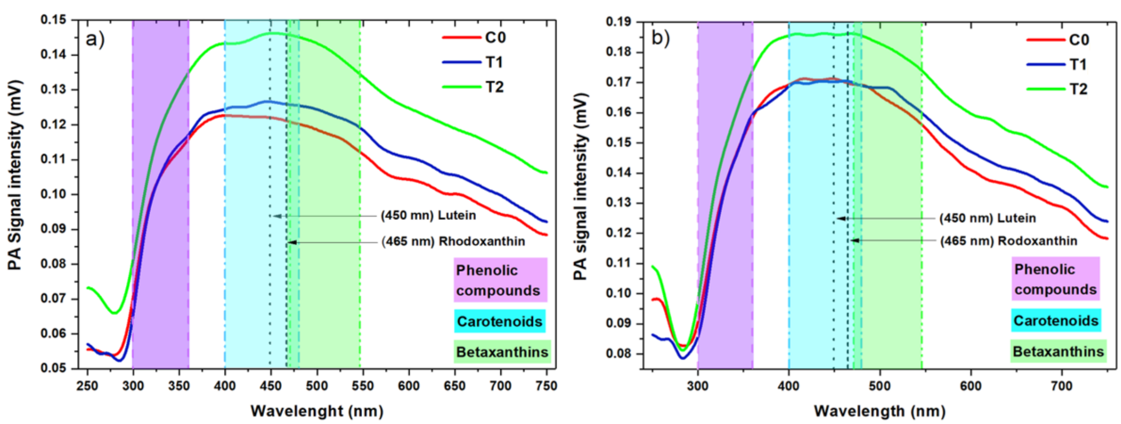

Figure 7: Photoacoustic spectrum of EMF priming broccoli seeds. (**a**) 1 radiation regime, (**b**) 2 radiation regimes.

Fig. 8 shows the first derivative of the photoacoustic signal spectra where it is possible to observe the maxima (crossing of the derivative with 0) and minima (peaks of the derivative). In phase 2 (Fig. 8a) the maximum was at wavelengths 409 (T2), 418 (T1), 435–440 and 648 nm (control) and a minimum at 310 nm. For phase 4 (Fig. 8b), maxima were found at 418 (T1), 423 (T2), 432 (control), 449 (T1), 455 (T2) and 498 nm

(T1II) and the minimum the same as in phase 1 (310 nm). This confirms the findings of the photoacoustic absorption spectrum and the absorbing peaks of the spectrum.

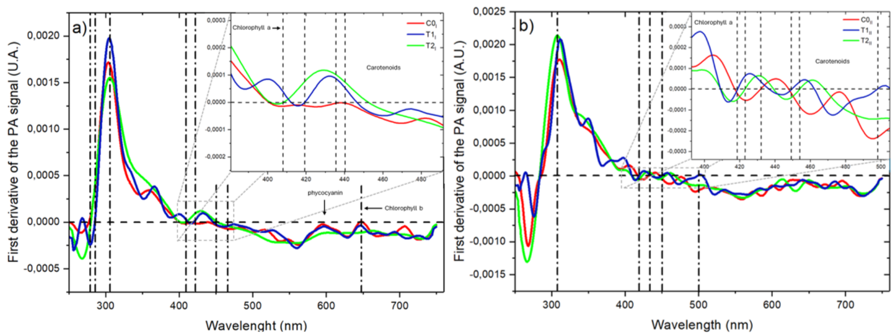

Figure 8: First derivative of the PA signal (**a**) phase 2 spectrum, (**b**) phase 4 spectrum.

3.5 Infrared Spectroscopy

Fig. 9 shows the FTIR absorbance and transmittance spectra of untreated and EMF-primed broccoli seeds over the wavenumber range of 500–4000 cm^{-1}. Panels 9a and 9b show the treatments conducted under one exposure regime (phases 1 and 2), whereas panels 9c and 9d show the treatments conducted under two exposure regimes (phases 3 and 4). In phase 2, the EMF-only treatments applied for 60 min (T3) and 120 min (T6) showed lower absorbance than the untreated control in the ranges of 420–1753 and 2855–3624 cm^{-1}. The untreated control showed the highest absorbance at approximately 1600 and 3400 cm^{-1}. At 1600 cm^{-1}, absorbance decreased by 50.19% and 29.95% in the 60- and 120-min treatments, respectively. At 3400 cm^{-1}, a decrease of 42.34% was observed for both treatments relative to the control. Consistently, the transmittance of the EMF-primed seeds was higher than that of the untreated control in these wavenumber regions.

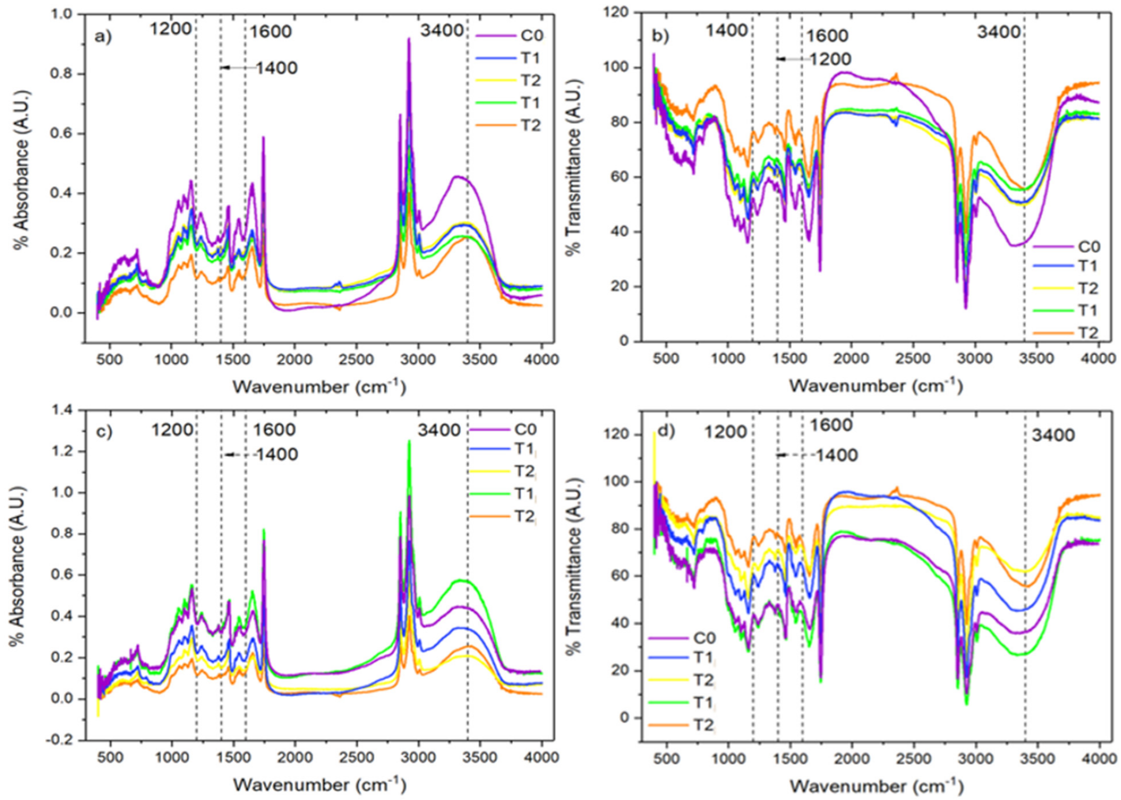

Figure 9: FTIR spectra of untreated and EMF-primed broccoli seeds. (**a**) Absorbance spectra for phases 1 and 2 under one exposure regime; (**b**) transmittance spectra for phases 1 and 2 under one exposure regime; (**c**) absorbance spectra for phases 3 and 4 under two exposure regimes; and (**d**) transmittance spectra for phases 3 and 4 under two exposure regimes.

The absorbance spectra of phases 1 and 3 had a higher absorbance percentage trend than the treatments in phase 3 T1, T2 and T2 for phase 4.

3.6 Relationship between Seed Moisture and Germination

Table 3 and Fig. 10 show the relationship between germination percentage and seed moisture content after EMF priming. Seed moisture decreased with increasing EMF exposure, particularly under two exposure regimes. This decrease was associated with lower germination percentages after longer exposure times, suggesting that excessive EMF exposure may negatively affect seed hydration status. In contrast, moderate exposure under one regime maintained higher moisture levels and was associated with better germination performance. These results support the idea that the effect of EMF priming depends on the balance between exposure duration, moisture retention and seed germination response.

Table 3: Germination percentage and humidity content of the electromagnetically pre-conditioned seeds.

Radiation	Time (min)	Germination (%)	Humidity (%)
1	0	19.75	98.66667
	7.5	19.5	98.01325
	15	20.5	98.01325
	60	23	98.01325
	120	20.25	98
2	0	19.75	98.66667
	7.5	19	97.33333
	15	20	97.33333
	60	15.75	96.66667
	120	16.50	96.68874

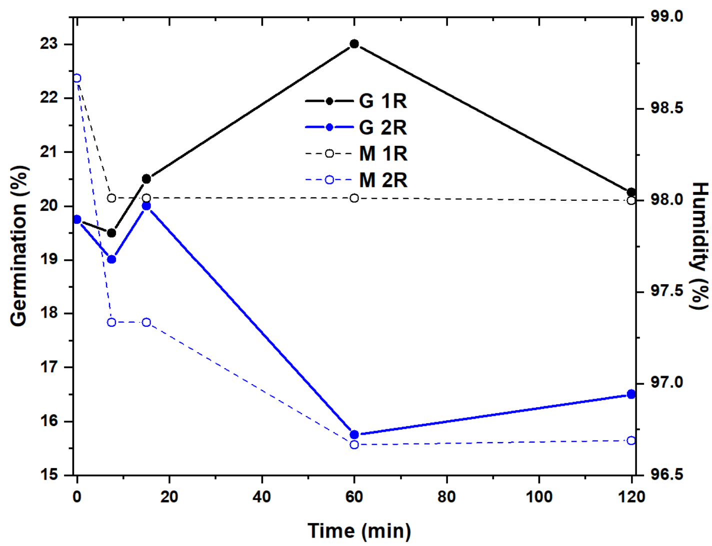

Figure 10: Germination percentage and Humidity content of seeds treated with electromagnetic field.

4 Discussion

The present study showed that EMF priming affected broccoli seed germination in a treatment-dependent manner. The response was not uniformly positive across all exposure regimes, exposure durations and stress conditions. Short exposure times were more favorable in specific cases, particularly under water stress, whereas longer or repeated exposures produced neutral or negative responses. This pattern suggests the existence of an exposure window in which EMF priming may stimulate germination-related processes, while excessive exposure may impose additional stress on the seeds.

The beneficial effects observed under specific EMF treatments may be associated with physiological changes occurring during early imbibition. Seed germination involves rapid water uptake, membrane reorganization, ion homeostasis, and metabolic reactivation. Positive effects of magnetic-field treatments on germination and early seedling development have been reported in several vegetable crops [19] and in cucumber [20]. In maize, EMF treatment improved germination kinetics and seedling growth [21], while magnetopriming mitigated the adverse effects of salinity on germination, growth, and photosynthetic performance [22]. Magnetopriming also modified water absorption in triticale seeds exposed to PEG- and NaCl-induced stress [23]. At the mechanistic level, magnetoprimed soybean seeds under salinity showed increased nitric oxide synthesis, changes in germination-related phytohormones, and a reduced Na^+/K^+ ratio, which were associated with improved germination and salt tolerance [24]. However, these physiological and biochemical mechanisms were not directly measured in the present study.

However, the results also showed that EMF priming did not always improve germination. In some treatments, particularly under longer exposure times and two exposure regimes, germination was reduced or showed no clear advantage compared with the control. This variability may be explained by an excessive EMF dose. Prolonged exposure increased seed temperature and reduced seed moisture content, especially under two exposure regimes. Although the temperature reached during EMF treatment remained moderate and all germination tests were conducted under controlled conditions at 25°C, the associated moisture loss may have modified the hydration status of the seed and reduced the benefits of priming. Therefore, the inconsistent responses observed in this study may reflect the balance between stimulatory effects at short exposure times and stress-like effects at longer or repeated exposures.

The FTIR results support the interpretation that EMF exposure modified moisture-related seed properties. The O-H functional group is commonly associated with absorption bands located near 1600 and 3400 cm^{-1} [25]. Higher water content is generally related to greater absorbance in these regions [26,27]. In the present study, absorbance decreased at these wavenumbers, particularly in seeds exposed to two regimes and longer treatment times. This agrees with the measured decrease in seed moisture content and suggests that prolonged EMF exposure affected the water status of the seeds. Similar changes in moisture-related absorption bands have been reported in biological materials during dehydration or storage processes [25]. Since water availability and hydration dynamics are critical during imbibition, these FTIR changes may help explain why longer or repeated EMF exposures did not consistently improve germination.

Changes in the O-H stretching region may also be related to modifications in hydrogen bonding. Previous studies have suggested that electromagnetic fields can influence the organization of water molecules and hydrogen bonds [27]. Because water is a dipolar molecule, it can be partially aligned under an electric field [28]. In addition, the O-H stretching band around 3400 cm^{-1} is associated with hydrogen-bonded water molecules, and changes in this region can indicate modifications in the interaction among water molecules [29]. Therefore, the FTIR spectra observed in this study may reflect changes in seed moisture distribution and hydrogen-bond organization after EMF exposure.

Digital image analysis provided complementary evidence of physical changes induced by EMF priming. The higher pixel intensity values observed after longer exposures may reflect changes in seed surface reflectance, texture or moisture distribution. These changes do not necessarily indicate improved physiological performance, but they support the idea that EMF exposure can modify seed coat-related properties without causing the type of surface degradation associated with more aggressive physical treatments such as UV-C radiation [30]. Therefore, digital image analysis should be interpreted as an indirect indicator of physical seed changes rather than as a direct measure of germination potential.

Photoacoustic spectroscopy also revealed treatment-dependent changes in the optical absorption profile of the seeds. The increased PA signal observed after longer exposures may be associated with changes in compounds absorbing in the carotenoid-related spectral region. Previous studies have reported increases in photosynthetic pigments, including chlorophylls and carotenoids, after magnetic-field treatment in lettuce seeds [31]. However, because PAS provides indirect spectral information, the present results should be interpreted as evidence of biochemical or optical changes rather than as direct quantification of carotenoid concentration. Future studies should combine PAS with biochemical assays to confirm the compounds responsible for these spectral differences. Recent studies further show that seed priming can improve early plant performance under abiotic stress, although the response depends on the crop species and treatment conditions. KNO_3 priming enhanced salt-stress tolerance in maize, whereas chemical priming improved chilling tolerance in rice seedlings [32,33].

Taken together, the germination, FTIR, moisture, imaging and PAS results indicate that EMF priming can modify both physiological and physical seed properties. Nevertheless, the direction of the response depends on the exposure conditions. Short EMF exposure may favor early germination by improving hydration-related and metabolic processes, whereas longer or repeated exposures may reduce seed moisture and produce stress-like effects. This interpretation is consistent with the concept that magnetic and electromagnetic treatments are not universally beneficial, but depend on the intensity, duration, exposure regime, crop species and stress condition.

This study has some limitations. First, the physiological mechanisms underlying the observed responses were inferred from germination, moisture, imaging and spectroscopic measurements, but direct biochemical markers such as antioxidant enzyme activity, reactive oxygen species accumulation, membrane integrity or ion content were not quantified. Second, the beneficial effect of EMF priming was restricted to specific exposure conditions, indicating that the response cannot be generalized across all regimes or stress types. Third, the study was conducted under controlled laboratory conditions; therefore, further experiments under nursery or field conditions are needed to confirm the agronomic relevance of the optimized treatments. Future research should also include direct measurements of enzyme activity, oxidative status, ion balance and seedling growth to better explain the mechanisms involved in EMF-induced seed responses. Another limitation is that the short- and long-exposure treatments were evaluated in independent experimental phases, each with its own control. Therefore, direct statistical comparisons between short and long exposure times were not performed, and cross-phase comparisons should be interpreted descriptively.

5 Conclusions

This study shows that electromagnetic preconditioning can positively influence germination of broccoli seeds under stress conditions only in specific treatments and exposure times. The effect was not consistent across all radiation regimes, and in some cases no improvement or even slight reductions in germination were observed. Therefore, the beneficial impact of electromagnetic priming cannot be generalized and depends on the particular treatment conditions. It is very important to know the radiation window that generates benefits in the seeds because if they are exposed for a longer time the electromagnetic field stresses the seeds inhibiting their development.

The electromagnetic treatment caused a moderate increase in seed temperature, which was correlated with a decrease in seed moisture content. This temperature rise represents a physical effect of the treatment and was not considered a direct factor affecting germination, as all germination tests were performed at a constant temperature of 25°C. This is evidenced by the decrease in absorbance in the wavenumber range 1600 and 3400 cm^{-1} where the O-H functional group is located.

Digital image analysis revealed that seeds exposed to longer electromagnetic radiation exhibited higher pixel intensity values, which suggests modifications in the optical and structural properties of the seed coat. These changes may be related to alterations in surface texture or moisture distribution induced by the electromagnetic treatment. Therefore, image processing can be considered a useful complementary tool for detecting physical changes in seeds subjected to electromagnetic conditioning.

Acknowledgement: The authors acknowledge support of the National Polytechnic Institute through the projects SIP (Research Secretary), EDI (Researcher Performance Incentives), COFAA (Commission for the Operation and Promotion of Academic Activities). We would like to thank the support of the project entitled UV-C light and hydro

and nutrient priming systems in sprout production as a strategy to add foods with low and minimal postprandial response: educational intervention.

Funding Statement: This research was funded by Secretariat of Research and Graduate Studies (SIP-IPN) by Project SIP 20254142 and 2026344.

Author Contributions: The authors confirm contribution to the paper as follows: Conceptualization, Claudia Hernandez-Aguilar; methodology, all authors; investigation, Julio Antonio Gutierrez-Dominguez and Raúl Romero-Galindo; writing—original draft preparation, Julio Antonio Gutierrez-Dominguez, Raúl Romero-Galindo and Claudia Hernandez-Aguilar; writing—review and editing, José Álvarez, María Victoria Carbonell, Arturo Domínguez-Pacheco and Alfredo Cruz-Orea; visualization, Julio Antonio Gutierrez-Dominguez and Claudia Hernandez-Aguilar; supervision, Claudia Hernandez-Aguilar, Arturo Domínguez-Pacheco and José Álvarez. All authors reviewed and approved the final version of the manuscript.

Availability of Data and Materials: The data that support the findings of this study are available from the corresponding author, José Álvarez, upon reasonable request.

Ethics Approval: Not applicable.

Conflicts of Interest: The authors declare no conflicts of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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