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Enhancing germination and vigor of sugar beet seeds through priming treatments under ambient storage

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Abstract

Objective: Sugar beet is primarily cultivated from seeds, and one of its main challenges is the low germination rate caused by seed dormancy due to seed coat coverage. Seed priming is an effective method to overcome dormancy and enhance both germination percentage and speed. However, storing primed seeds reduces their longevity and accelerates deterioration. Therefore, determining the duration of priming effects during ambient storage is important. This study aimed to improve germination and seed vigor of sugar beet seeds and evaluate the storability of primed seeds.

Methods: The experiment was conducted in 2025 at the Seed Science and Technology Laboratory of Hamyaran Keshavarz Company in Zarand, Kerman, Iran. The experimental design was a completely randomized design with seven priming treatments (0.2% nano-urea for 1 hour, 0.3% seaweed extract for 1 hour, 20 ppm gibberellic acid for 24 hours, hydroelectropriming with a current of 15 V for 2 minutes, hydropriming for 12 hours, electropriming with a current of 20 V for 2 minutes, and 20 ppm gibberellic acid for 1 hour), along with the control treatment, each replicated four times with 100 seeds per replication.

Results: Nano-urea (0.2%), hydroelectropriming (15 V for 2 minutes), and seaweed extract (0.3%) had the largest positive effect on germination and seed vigor. The beneficial effects of priming significantly declined during storage, affecting germination, seed vigor, and the activities of antioxidant enzymes, such as catalase, superoxide dismutase, and peroxidase. Hydroelectropriming and nano-urea demonstrated the best seed viability retention over three months of storage.

Conclusion: Hydroelectropriming (15 V, 2 min) and nano-urea priming (0.2%, 1 h), as innovative and environmentally friendly methods, are recommended to maximize germination and vigor while preserving seed storability for three months, supporting sustainable sugar beet production.

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Introduction

Sugar beet (*Beta vulgaris*) is a biennial plant from the Chenopodiaceae (or Amaranthaceae) family and is the most significant crop for global sugar production. It is cultivated as an annual crop for sugar yield and exhibits relative tolerance to cold, heat, drought, and soil salinity. However, seed germination in sugar beet faces several major challenges that can compromise plant establishment. One key issue is the strong adhesion of the seed cap to the seed coat, which prevents seedling emergence despite the initiation of the physiological germination process, often leading to seedling mortality (TeKrony 1969). The seed coat itself can act as a physical barrier, inducing seed dormancy. Specifically, the hard and impermeable seed coat restricts the uptake of water and oxygen, thereby delaying or inhibiting germination. This type of dormancy, resulting from seed coat characteristics, is referred to as physical dormancy (Chu *et al.* 2022).

Seed priming is a proven method for enhancing both the percentage and speed of seed germination, as well as seed vigor, particularly under stressful environmental conditions such as salinity. This technique involves treating seeds with various substances or solutions to initiate water uptake without completing germination, after which the seeds are dried and stored to improve their performance at planting. Priming is a pre-sowing treatment that prepares seeds under controlled conditions, enabling faster, more uniform, and stronger germination after sowing. By facilitating better and quicker water absorption, priming helps seeds overcome physiological or chemical dormancy. Consequently, under adverse conditions like saline soils, the proportion of germinated seeds increases significantly (Shokouhian and Omidi 2021).

Priming activates key enzymes and metabolic pathways within the seed, which enhances the seedling's ability to cope with environmental stresses. This includes improved regulation of ionic balance, increased antioxidant activity, and preservation of cellular structure. Due to the internal preparation initiated during priming, seeds germinate more rapidly than untreated seeds, resulting in uniform emergence and potentially higher yields in the field. These physiological and biochemical changes contribute to more robust plant establishment and improved crop performance, especially in challenging environments (Karimi *et al.* 2025).

This study focused on enhancing seed germination in sugar beet using electro-priming and hydro-electro-priming methods. Research has demonstrated that primed seeds cannot be stored for extended periods due to lipid peroxidation and increased metabolic activity, which accelerate the formation of reactive oxygen species (ROS) and elevate lipid peroxidation levels during storage, leading to rapid deterioration of primed seeds (Maskri *et al*. 2003). However, the impact of electro-priming and hydro-electro-priming methods on post-priming storage longevity has not been fully assessed. This research is particularly significant because many farmers lack adequate storage facilities, resulting in seeds being kept at room temperature.

Materials and Methods

In this study, sugar beet seeds of the Arya variety, supplied by Pakan Bazr Company, were used. This research was conducted at the Seed Science and Technology Laboratory of Hamyaran Keshavarz Company in Zarand, Kerman, Iran. The experimental design was implemented as a completely randomized design with seven different priming treatments alongside a control (Table 1). Each treatment was replicated four times, with 100 seeds per replication.

Seed priming methods

The initial seed moisture content was 10%. For chemical priming, sugar beet seeds were soaked for 1 and 24 hours in gibberellic acid (GA3) solution at concentrations of 10, 15, and 20 ppm, nano-urea solution at 0.2%, 0.3%, and 0.4%, and seaweed extract at 0.2%, 0.3%, and 0.4% for 1 hour. Hydropriming was conducted by soaking seeds in double-distilled water for 12 hours at 25 °C. Electro- priming was performed using a horizontal gel electrophoresis apparatus with a 0.5% NaCl solution. Electric currents at voltages of 10, 15, and 20 V were applied for 1 and 2 minutes. For hydroelectropriming, seeds were first soaked in double-distilled water for 12 hours at 25 °C, as in the hydro-priming method, followed by electrical treatments identical to those used in electropriming (10, 15, and 20 V for 1 and 2 minutes).

After each priming method, seeds were dried to restore their initial moisture content of 10%. Seed germination tests were conducted in a germination apparatus at 20 °C, following ISTA protocols for sugar beet seeds. According to ISTA standards, germination tests for sugar beet are typically performed in darkness on moistened-pleated germination paper (grade 3014 or similar), with seeds evenly spaced and maintained at a constant temperature for 14 days. The number of germinated seeds was recorded and analyzed daily over 14 consecutive days, ensuring uniformity and reproducibility as required by international seed testing rules.

Post-priming storage

Seven treatments that demonstrated considerable improvement in seed germination and seedling growth characteristics were selected for the storage stage. These treatments included: 0.2% nanourea for 1 hour, 0.3% seaweed extract for 1 hour, 20 ppm GA3 for 24 hours, hydroelectropriming with a current of 15 V for 2 minutes, hydropriming for 12 hours, electropriming with a current of 20 V for 2 minutes, and 20 ppm GA3 for 1 hour, along with the control treatment (Table 1). Primed seeds were stored for four months in closed containers at a constant temperature of 25 °C.

During storage, germination and seed vigor characteristics were periodically evaluated to assess the effects of priming on maintaining seed quality over time. This approach helped to identify the most effective priming treatments for extending seed shelf life and preserving germination capacity, thereby optimizing seed storage processes.

Biochemical analysis

To assess enzymatic activity, primed and control seeds were soaked in distilled water for 24 hours at 25 °C. Then, 1 g of soaked seeds was ground under cold conditions with 5 ml of phosphate buffer (pH 7.5), containing 1 mmol polyethylene glycol, 1 mmol phenylmethylsulfonyl fluoride, 8% (w/v) polyvinylpyrrolidone, and 0.01% (v/v) Triton X-100. The resulting homogenate was centrifuged at 12,000 rpm for 20 minutes at 4 °C, followed by a second centrifugation at 14,000 rpm for 10 minutes at 4 °C. The final supernatant was used to measure the activities of catalase (CAT), peroxidase (POD), and superoxide dismutase (SOD).

Table 1. Details of treatments selected for post-priming storage of sugar beet seeds.

Treatment	Treatment detail	Application rate	Treatment duration
T1	Nano-urea	0.2%	1 h
T2	Sea weed extract	0.3%	1 h
T3	GA3 priming	20 ppm	24 h
T4	Hydroelectropriming (HEP)	Soaking in double-distilled water for 12 h + 15 V for 2 m	12 h + 2 min
T5	Hydropriming (H)	Soaking in double-distilled water	12 h
T6	Electropriming (EP)	20 V	2 min
T7	GA3 priming	20 ppm dH_2O	1 h
C	Control	-	-

***Catalase activity*:** CAT activity was measured using the method described by Aebi (1973). In this assay, enzyme activity was determined by monitoring the rate of hydrogen peroxide (H_2O_2)

decomposition at a wavelength of 240 nm. The activity was expressed as the amount of micromoles of H_2O_2 decomposed per milliliter of solution per minute per milligram of protein ($\mu mol \cdot mL^{-1} \cdot min^{-1} \cdot mg^{-1}$).

***Peroxidase activity*:** POD activity was measured using a spectrophotometer following the method described by Chance and Maehly (1955). In this assay, the rate of tetraguaiacol oxidation was monitored at a wavelength of 470 nm, and the reaction rate was calculated using an extinction coefficient of 25.5 $\mu mol.cm^{-1}$.

***Superoxide dismutase activity*:** SOD activity was determined according to the method described by Dhindsa *et al.* (1981). In this assay, the absorbance of samples was measured at a wavelength of 560 nm using a spectrophotometer. One unit of enzyme activity was defined as the amount of enzyme required to reduce the absorbance by 50% compared to control tubes without enzyme. Enzyme activity was expressed as units per minute per milligram of protein.

Traits

Data on post-priming traits, including initial count of germination, germination percentage, germination rate, seedling length, seedling fresh and dry weight, EC (electrical conductivity), and pH were recorded. The EC of seeds was measured following the protocol of Milosevic *et al.* (2010). The pH of seeds was measured following the method of Santos *et al.* (2019). The formulas related to germination are as follows:

- Germination at initial count = Number of germinated seeds on the 2nd day
- Germination percentage = (Germinated seeds/Total seeds) × 100.
- Germination rate = n1/d1 + n2/d2 + n3/d3 + …

Where, n = Number of germinated seeds on given days and d = Number of days

- Vigor Index 1 = Seedling length × Germination percentage
- Vigor Index 2 = Seedling dry weight × Germination percentage

Statistical analysis

All recorded traits were subjected to one-way analysis of variance (ANOVA) based on a completely randomized design. When the ANOVA indicated significant treatment effects, Fisher's least significant difference (LSD) test at the 5% probability level was used to compare means. Pearson correlation coefficients among traits were calculated using WASP software (version 0.01).

Results and Discussion

Effect of seed priming on seed germination and seedling growth characteristics

The results indicated that nano-urea at a concentration of 0.2% and 15 V for 2 minutes demonstrated the most effective performance in enhancing germination percentage and improving seedling growth characteristics. The detailed effects of the seven selected treatments are provided in Table 2. In the initial germination count, hydroelectropriming at 15 V for 2 minutes achieved the highest germination percentage (28%), followed by treatments of 0.2% nano-urea and 0.3% seaweed extract, both with 27% germination. However, these differences were not significant. In contrast, the control treatment and electropriming at 20 V for 2 minurtes exhibited the lowest initial germination percentage (10%). These results underscore the importance of selecting appropriate priming treatments to enhance seed germination and initial growth. The highest germination percentage was observed in the treatment of 0.2% nan-ourea with 91%, followed by hydroelectropriming at 15 V for 2 minutes with 88%, 0.3% seaweed extract with 84%, electropriming with 75%, and GA3 (24 h) with 74%. The germination rate followed a pattern similar to the germination percentage, with the highest value recorded in the treatment of 0.2% nano-urea (12.26), followed by hydroelectropriming at 15 V for 2 minutes (11.10) and 0.3% seaweed extract (10.42).

The highest seedling vigor index was observed in the treatments of hydroelectropriming at 15 V for 2 minutes and 0.2% nano-urea. These results indicate a positive effect of these treatments on both germination and early seedling growth. These treatments also showed comparable performance across seedling length, seedling fresh weight, and seedling dry weight.

After priming, a reduction in EC was observed, although the differences in EC among treatments were not statistically significant. These results indicate that all priming treatments reduced seed EC compared to the control, suggesting lower membrane damage and better seed quality.

Nano-urea enhances seed nutrient uptake efficiency, promoting rapid germination and seedling vigor in various crops (Nile *et al.* 2022b). According to Seleiman *et al.* (2021), its nanoparticle size enables direct seed penetration, reducing time-to-50% germination by 20-30% and increasing vigor indices by 25-40% compared to hydropriming or controls. Maize seeds primed with nano-urea germinated two days earlier with 35% higher vigor (Nile *et al.* 2022b). In a study in wheat, nano-urea achieved 90% germination rate versus 72% in the untreated control (Ahmed *et al*. 2023). Nano-urea priming upregulates stress-tolerance genes and reduces conventional fertilizer needs, minimizing environmental impacts (Rossi *et al*. 2022; Qureshi *et al*. 2023).

Effect of seed priming on germination characteristics during storage

After one month of storage, a considerable decrease in germination percentage was observed across all treatments, although the germination percentage and rate in the priming treatments remained higher than those of the control. The highest germination percentage after storage was recorded in the hydroelectropriming treatment (80.25%), followed by 0.2% nano-urea (73.25%). These findings demonstrate the positive effect of the applied treatments on enhancing seed stability and maintaining germination capacity during storage. Treatment of 0.3% seaweed extract resulted in a germination percentage of 60%. Seedling growth traits in primed seeds also declined after the storage period, with detailed results presented in Table 3. Additionally, EC in the priming treatments significantly decreased compared to the control, especially in the hydroelectropriming and 0.2% nano-urea treatments. pH values showed a slight increase or decrease in some priming treatments after one month of storage (Table 3).

After two months of storage, a sharp decline in germination percentage was observed in GA3 (1 h) and electropriming (20 V for 2 minutes) treatments as compared to one month of storage. Germination percentages in other treatments also decreased, although hydroelectropriming treatment maintained the highest value (79.25%). Increases in EC were noted in all primed treatments during two months of storage compared to one month of storage (Table 4). Priming benefits declined significantly during three months of ambient storage, reducing germination percentage, vigor indices, and antioxidant enzyme activities (CAT, SOD, and POD) across all treatments.

After three months of storage, the lowest germination percentage was observed in the GA3 (1 h) (13%) treatment, which was significantly lower than that of the control (28%). The highest germination percentage was recorded in 0.2% nano-urea (65%), followed by hydroelectropriming (63%). The maximum germination rate was observed in the hydroelectropriming treatment (5.83 seedlings per day). Priming benefits declined gradually across all treatments during three months of ambient storage, as evidenced by reduced germination percentages (91% to 55% in controls), vigor indices, and antioxidant enzyme activities (CAT, SOD, and POD). EC values increased in all priming treatments compared to two months of storage but they were significantly lower than those of the control. The lowest EC was observed in the hydroelectropriming treatment (Table 5).

After four months of storage, the hydroelectropriming treatment showed a first-count germination of 8% and a final germination percentage of 53.5%. Hydroelectropriming at 15 V for 2 minutes demonstrated the highest germination rate after four months of storage period, reaching 4.01 seedlings per day at four months, followed by the 0.2% nanourea treatment (2.34 seedlings per day). Subsequently, the 0.2% nano-urea and 20 ppm GA3 treatments exhibited EC values of 0.2 dS/m and

0.21 dS/m, respectively. These results suggest reduced cell damage and improved seed quality in these treatments during storage. In contrast, the control treatment recorded the highest EC value in this storage period (Table 6). Hydroelectropriming and nano-urea showed significantly higher seedling vigor indices compared to the control (Vigor index 1: 1268.22 and 1280, and 135, respectively; Vigor index 2: 1.87, 1.75, and 0.27, respectively).

Table 2. Effect of priming treatments on sugar beet seed germination and seedling growth characteristics.

Treatment	Germination at first count (%)	Germination percentage (%)	Germination rate	Seedling length (cm)	Seedling fresh weight (g)	Seedling dry weight (g)	Vigor index 1	Vigor index 2	EC (dsm^{-1})	pH
T1	27	91	13.26	12.8	0.34	0.018	1280.00	1.75	0.123	6.05
T2	27	84	10.42	12.10	0.27	0.017	937.37	1.42	0.176	6.11
T3	15	74	7.36	9.85	0.18	0.018	684.68	1.37	0.154	6.03
T4	28	88	11.10	13.38	0.39	0.020	1268.22	1.87	0.243	6.52
T5	18	59	7.78	12.26	0.22	0.015	820.36	0.91	0.142	6.76
T6	10	75	5.62	11.14	0.27	0.015	666.34	0.79	0.196	6.61
T7	14	62	9.88	9.27	0.23	0.015	612.39	0.86	0.108	6.68
C	0	55	2.84	4.85	0.18	0.009	135.00	0.27	0.603	6.65
LSD (5%)	11.47	26.55	5.634	2.844	0.055	0.0038	352.411	1.087	0.158	0.085
CV	45.24	24.75	45.33	18.24	14.53	16.53	30.22	64.58	49.83	0.91

T1: Nano urea, T2: Sea weed extract, T3: GA3 (24 h), T4: Hydro-electro-priming (15 V for 2 min), T5: Hydro-priming, T6: Electro-priming (20 V for 2 min), T7: GA3 (1 h), C: Control.

Table 3. Effect of different priming treatments on germination characteristics of sugar beet after one month of post-priming storage.

Treatment	pH	EC (dSm^{-1})	Germination rate	Germination (%)	Germination at first count (%)
T1	6.17	0.15	6.26	73.75	13
T2	6.19	0.188	5.16	60.00	10
T3	6.09	0.163	6.36	57.50	13
T4	6.60	0.120	11.50	80.25	29
T5	7.07	0.163	5.37	64.00	9
T6	6.76	0.215	3.15	55.50	4
T7	6.79	0.190	3.02	44.00	6
C	6.66	0.600	1.50	30.50	0
LSD (5%)	0.131	0.034	2.382	20.232	7.7
CV (%)	1.37	10.48	30.91	23.87	49.28

T1: Nano urea, T2: Sea weed extract, T3: GA3 (24 h), T4: Hydro-electro-priming (15 V for 2 min), T5: Hydro-priming, T6: Electro-priming (20 V for 2 min), T7: GA3 (1 h), C: Control.

Table 4. Effect of different priming treatments on germination characteristics of sugar beet after two months of post-priming storage.

Treatment	pH	EC (dSm^{-1})	Germination rate	Germination (%)	Germination at first count (%)
T1	6.58	0.185	5.22	66.25	8
T2	6.56	0.215	2.25	35.75	3
T3	6.61	0.185	4.09	53.25	8[b]
T4	6.40	0.130	6.09	79.25	12
T5	6.46	0.220	2.64	45.25	3
T6	6.58	0.373	1.58	28.50	2
T7	6.64	0.218	1.94	34.50	2
C	6.73	0.695	1.10	28.00	0
LSD (5%)	0.055	0.046	0.954	12.948	2.2
CV (%)	0.57	11.43	21.04	19.18	40.31

T1: Nano urea, T2: Sea weed extract, T3: GA3 (24 h), T4: Hydro-electro-priming (15 V for 2 min), T5: Hydro-priming, T6: Electro-priming (20 V for 2 min), T7: GA3 (1 h), C: Control.

Table 5. Effect of different priming treatments on germination characteristics of sugar beet after three months of post-priming storage.

Treatment	pH	EC (dSm^{-1})	Germination rate	Germination (%)	Germination at first count (%)
T1	6.51	0.197	4.49	65.50	6
T2	6.51	0.215	2.09	34.50	3
T3	6.56	0.197	3.24	49.00	5
T4	6.30	0.14	5.83	63.00	12
T5	6.39	0.225	1.40	28.00	0
T6	6.57	0.390	0.49	13.00	0
T7	6.56	0.233	1.98	24.00	4
C	6.58	0.655	1.30	27.50	0
LSD (5%)	0.097	0.088	1.201	13.778	3.5
CV (%)	1.02	12.50	31.68	24.85	63.48

T1: Nano urea, T2: Sea weed extract, T3: GA3 (24 h), T4: Hydro-electro-priming (15 V for 2 min), T5: Hydro-priming, T6: Electro-priming (20 V for 2 min), T7: GA3 (1 h), C: Control.

After five months of storage, the seed germination rate and seedling vigor indices decreased in all treatments, with values falling below those of the control (data not provided). This decline highlights the impact of prolonged storage on seed quality, even in the presence of priming treatments.

Seed priming significantly enhances germination and seedling vigor during storage, and the method and intensity of priming play a key role in maintaining seed viability. Among all tested treatments, hydroelectropriming at 15 V for 2 minutes provided the most stable and sustained

improvement, maintaining the highest germination percentages and lowest electrical conductivity over four months of storage. These findings demonstrate that optimized priming conditions can effectively delay seed deterioration and extend storage life. Future studies should explore the physiological and biochemical mechanisms underlying this increased stability and evaluate the long-term storage potential of electroprimed seeds under different environmental conditions. These results highlight the positive effect of priming treatments in maintaining and improving seed germination rates during storage, similar to the results of Zhao *et al.* (2021).

Table 6. Effect of different priming treatments on germination characteristics of sugar beet after four months of post-priming storage.

Treatment	pH	EC (dSm^{-1})	Germination rate	Germination (%)	Germination at first count (%)
T1	6.45	0.20	2.34	44.00	2
T2	6.54	0.24	1.36	31.50	0
T3	6.72	0.21	1.16	20.50	0
T4	6.60	0.15	4.01	53.50	8
T5	6.75	0.25	1.21	28.00	0
T6	6.71	0.32	0.35	9.00	0
T7	6.70	0.26	0.91	18.50	0
C	6.67	0.63	1.02	28.20	0
LSD (5%)	0.0.76	0.043	0.584	7.576	2.7
CV (%)	0.79	10.52	25.95	17.84	146.06

T1: Nano urea, T2: Sea weed extract, T3: GA3 (24 h), T4: Hydro-electro-priming (15 V for 2 min), T5: Hydro-priming, T6: Electro-priming (20 V for 2 min), T7: GA3 (1 h), C: Control.

Effect of seed priming on enzyme activity

Following priming, CAT activity in the control group was lower than that in the primed treatments on day 0. Over the subsequent three months, CAT, SOD, and POD activities declined continuously across all treatments (Table 7). However, after three months of storage, all treatments, except GA_3 (1 h) (0.03 CAT/mg protein/min) and electropriming (20 V, 2 min) (0.050 CAT/mg protein/min) maintained higher CAT activity than the control (0.09 CAT/mg protein/min) (Table 7).

SOD activity increased after priming compared to the control. Nano-urea (0.2%) and hydroelectropriming showed the highest initial SOD activity (Table 7). After three months of storage, hydroelectropriming (15 V, 2 min) maintained the highest SOD activity, followed by nano-urea (0.2%), with the control showing the lowest value at 0.023 units/mg protein/min (Table 7).

At the start of storage, the hydroprimed seeds showed the lowest POD activity (4.5 units/mg protein/min), followed by hydroelectropriming (7.8 units); both were lower than the control (10.2

units) (Table 7). Despite the low POD activity, these treatments achieved higher germination (91% and 88% for hydropriming and hydroelectropriming, respectively) and Vigor Index 1 (1280 and 1268.22 for hydropriming and hydroelectropriming, respectively) than the control (55% germination and 135 Vigor Index 1) (Table 2). The highest POD activity occurred in electropriming (16 units) and GA_3 (24h) (15.9 units), followed by seaweed extract (14.3 units) and nano-urea (14.1 units). All priming treatments showed a POD decline after three months of storage (Table 7).

Priming enhanced sugar beet germination and vigor, but primed seeds showed greater declines in germination and enzyme activity during storage compared to the control. Nano-urea (0.2%, 1h) was the most effective initially, increasing germination to 91% and vigor index to 1280 (Table 2), likely due to nitrogen-enhanced metabolism. Nano-priming creates cell wall nanopores via controlled ROS signaling that loosens tissues and accelerates germination (Nile *et al.* 2022). Aquaporins facilitate beneficial H_2O_2 signaling for water uptake and starch hydrolysis (Charkhandaz and Sorkheh 2022).

Table 7. Catalase (CAT), peroxidase (POD), and superoxide dismutase (SOD) activity in sugar beet seeds after priming and after two and four months of post-priming storage.

Treatments	CAT (mg/protein/min)			SOD (mg/protein/min)			POD (mg/protein/min)		
	Zero day	Two month	Three month	Zero day	Two month	Three month	Zero day	Two month	Three month
T1	0.49	0.35	0.30	0.089	0.076	0.040	14.1	12.20	11.8
T2	0.30	0.28	0.19	0.080	0.033	0.019	14.3	9.98	8.4
T3	0.30	0.29	0.28	0.078	0.051	0.039	15.9	14.70	13.8
T4	0.44	0.37	0.30	0.089	0.076	0.062	7.8	6.00	5.8
T5	0.17	0.21	0.12	0.060	0.038	0.038	4.5	4.00	2.0
T6	0.12	0.10	0.05	0.040	0.022	0.012	16.0	11.90	10.0
T7	0.16	0.10	0.03	0.042	0.034	0.014	13.7	10.30	10.8
C	0.10	0.10	0.09	0.027	0.033	0.023	10.2	10.00	10.5

T1: Nano urea, T2: Sea weed extract, T3: GA3 (24 h), T4: Hydro-electro-priming (15 V for 2 min), T5: Hydro-priming, T6: Electro-priming (20 V for 2 min), T7: GA3 (1 h), C: Control.

Hydroelectropriming (15 V for 2 minutes) was another treatment that produced the highest improvements in seed germination rate, seedling growth, and SOD and CAT activity. As reported by Das and Prabha (2024), hydroelectropriming represents an innovative approach to enhancing the vigor and viability of stored onion seeds. Electrostatic field exposure may alter enzyme structure, and such structural modifications could contribute to the observed increase in SOD activity. This

enhanced enzymatic activity leads to more efficient scavenging of free radicals, thereby reducing the accumulation of hydrogen peroxide and superoxide radicals. Structural modifications in enzymes are generally driven by changes in charge distribution and transition states, which ultimately lower the activation energy required for catalysis (Abdoli 2020). Similarly, Garcia *et al.* (2021) observed that hydroelectrohybrid priming decreased emergence time and significantly improved the germination rate and vigor index of tomato seeds compared with the control, electric field, and hydropriming treatments. Shi *et al.* (2014) reported that a 12-hour soaking treatment followed by electropriming significantly increased germination percentage, germination rate, and seedling vigor index, demonstrating that the combination of pre-soaking and electrical priming can enhance seed germination and early seedling growth. The hybrid hydroelectropriming further improves the mobilization and efficient use of seed storage reserves, which strengthens overall seed performance, germination capacity, and early seedling establishment. Zhao *et al.* (2021) showed that hydroelectropriming provides additional benefits over hydropriming alone, including greater recovery of seed vigor, accelerated germination, and longer-lasting priming effects. This hybrid approach also enhances the utilization of stored reserves, leading to improved seedling growth, higher yield potential, and greater stability during the early stages of development (Zhao *et al.* 2021). Combining individual priming methods provides superior performance in maintaining seed health and vigor by reducing the drawbacks of individual approaches and boosting antioxidant activities (Jatana *et al.* 2024).

Seed priming with the plant hormone GA3 at a concentration of 20 ppm and a soaking duration of 24 hours was shown to positively influence seed germination characteristics and the activity of antioxidant enzymes CAT, SOD, and POD. GA3 treatment improves germination percentage and speed, boosts seed vigor, and enhances the activity of defense enzymes that protect against biotic and environmental stresses (Kibiniza *et al.* 2011). Interestingly, our experiment revealed that POD activity was lower in seeds treated with hydropriming and hydroelectropriming compared to the control and other treatments. This finding contradicts the common expectation that priming increases POD activity, which is typically associated with plant stress responses. In contrast, electropriming elevated POD activity after priming, suggesting that the type of priming method determines the enzyme's response. Higher POD activity is generally observed under stress conditions, indicating that hydropriming may result in minimal stress for seeds, while electropriming induces a more pronounced defense response. While some studies suggest that hydropriming does not significantly affect POD activity under stress (Zhang *et al.* 2023), the reasons for the observed decrease in POD activity, particularly with hydropriming, remain unclear and warrant further investigation.

In the present study, the germination percentage for the nano-urea and hydroelectropriming (15 V for 2 min) treatments gradually decreased over three months of storage, yet both maintained a germination rate above 60% (the minimum threshold required for sugar beet seed certification and acceptable field performance. However, after four months of storage, the hydroelectropriming treatment dropped to 50.53% germination, falling below the standard. This indicates that while priming provides stable effects on germination and viability in the short term, its benefits may diminish over extended storage periods, particularly under suboptimal conditions.

Seeds treated with GA3 and electropriming experienced a rapid decline in germination capacity after just one month of storage. This aligns with previous research showing that long-term storage negatively impacts seed germination, largely due to the reduction of starch and increased hydrolysis of soluble sugars, which significantly lowers germination rates in rice seeds (Hussain *et al.* 2015). Yan (2017) also reported decreased activities of antioxidant enzymes (SOD, POD, and CAT) in stored Chinese cabbage seeds, highlighting the metabolic changes that occur during storage. Increased metabolic activity in seeds, which raises molecular mobility and accelerates specific degradation reactions during storage. While hydroelectroprimed seeds maintain their germination percentage for up to three months, hydroprimed and electroprimed seeds begin to lose viability after one month. Zhao *et al.* (2021) found that hydroelectropriming significantly improved germination rate, vigor index, and metabolic efficiency in carrot seeds, reducing limitations of individual hydropriming or electropriming methods. Overall, the benefits of priming are most evident in the short term, but the longevity of primed seeds is often reduced during extended storage, especially under suboptimal conditions.

Our study demonstrates that various sugar beet seed priming treatments improve germination characteristics, such as early germination, germination capacity, and seed vigor. These treatments activate physiological and biochemical processes in seeds, promoting better root and stem development and increased seedling growth. The duration of beneficial effects under standard storage conditions was also reported, with some methods offering longer and more stable maintenance of seed quality and germination performance. In summary, seed priming is recognized as an effective method for enhancing germination and increasing seed resistance to environmental stresses and storage, a conclusion supported by numerous studies.

Conclusion

The study demonstrates that seed priming with nano-urea (0.2%) is one of the most effective methods for enhancing germination characteristics and seed vigor in sugar beet, followed by hybrid

hydroelectropriming (15 V for 2 minutes), and seaweed extract treatment (0.3%). These approaches regulate phytohormones and boost metabolic activity, leading to improved germination rates and seed resilience. Regarding post-priming storage, the 0.2% nano-urea and hydroelectropriming (15 V for 2 minutes) treatments showed favorable results in maintaining germination percentage and seed vigor, highlighting their practical value for farmers aiming to improve yield and storage stability. Further research is needed to clarify the molecular mechanisms behind the reduction in POD activity observed in hydroprimed sugar beet seeds. Understanding these mechanisms could refine priming techniques and enhance their efficiency, given POD's crucial role in stress response and oxidative balance in seeds. These findings underscore seed priming as a reliable method for improving germination and seed resistance, with broad implications for sustainable agriculture.

Data availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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