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Enhancing Seedling Growth in *Capparis spinosa* L. Seeds: Effects of Soaking and Cold Stratification

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ABSTRACT

The caper bush (*Capparis spinosa* L.), a hardy Mediterranean plant, exhibits remarkable resilience to arid conditions while offering a wealth of medicinal and industrial applications. However, low and inconsistent seed germination has often hindered large-scale cultivation, primarily due to physical and physiological dormancy. This study investigates the effects of various seed soaking durations and stratification times on different germination substrates, specifically sand and paper. Nine caper ecotypes were selected from diverse provinces of Iran. The study included control seeds and evaluated three soaking periods: soaking seeds in tap water at room temperature for 24 hours, 15 days, and 30 days. Additionally, five stratification periods were assessed at 4 °C for 7, 14, 21, 28, and 35 days. The results indicated that cold stratification applied over various periods in a sand substrate was the most effective method for breaking dormancy. Among the ecotypes studied, Alborz exhibited the highest germination percentage (91%) following a 35-day cold stratification period. The treatment demonstrated a positive impact on both the germination percentage and the mean daily germination rate. suggesting that cold stratification is the most effective approach to overcoming seed dormancy in caper ecotypes. Furthermore, our findings revealed that sand is the superior substrate for enhancing caper seed germination. The results imply that the incidence of fungal infection in sterilized sand is lower than in paper, and moisture levels are more uniformly maintained during germination. It was also noted that caper plants derived from freshly collected seed do not consistently develop fully, with only approximately 10% of fresh seeds germinating. The reactions of six caper ecotypes to cold stratification indicate that physiological dormancy is the primary factor contributing to the low germination percentage and rate. Additionally, both physical factors (such as the hard seed coat) and physiological factors (including the immature embryo) contribute to seed dormancy in caper seeds. The seed coat and the mucilage layer that developed its surface represent the primary barriers to germination. The responses of six caper ecotypes to cold stratification further confirm that physiological dormancy plays a significant role in low germination percentages and rates. Consequently, low winter temperatures achieved through cold stratification may be critical in facilitating the spread of caper ecotypes in Iran.

Keywords: Germination rate, Germination substrate, Physical dormancy, Physiological dormancy, Pre-treatment.

INTRODUCTION

Caper *(Capparis spinosa* L.) is a perennial shrub belonging to the Capparaceae family. It is native to the Mediterranean region and is also found in East Africa, Madagascar, Southwest and Central Asia, the Himalayas, the Pacific Islands, and Australia [1, 2]. The economic significance of caper has led to a notable expansion in its cultivation and production since the late 1980s [3]. This drought-tolerant species thrives in arid and semi-arid climates, blooming during the summer months. In Mediterranean regions, caper exhibits strong resilience to drought [4] and can withstand harsh conditions for example, strong winds and temperatures that can reach above 40 °C during dry summers [5]. For optimal growth, caper requires an annual temperature above 14 °C and annual rainfall exceeding 150 mm. While it can survive winters as a stump, frost during the vegetative phase can be detrimental. The plant typically flourishes at low altitudes, although specimens have been found at elevations over 1000 m above sea level [6, 7]. Caper is also known for its tolerance to calcareous and saline soils and its ability to endure challenging environmental conditions [8]. Traditionally, caper has been used as a food seasoning, with its fruits and buds consumed pickled or salted. Studies have identified alkaloids, lipids, flavonoids, and glucosinolates in capers, contributing to their antioxidant, cancer-preventive, and biopesticidal properties [9, 10].

Seeds can propagate caper plants or stem cuttings can be used; however, both methods present significant challenges for large-scale cultivation. Seed propagation is particularly problematic due to the combined effects of physical (seed coat) and physiological dormancy, which complicate germination [10]. As reported by Nowruzian and Aalami [2], the germination percentage of caper seeds during the initial two to three months following planting was observed to be only 5%. In contrast, Foschi *et al*. [11] found that when seed moisture content is adequate, germination rates can reach as high

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as 90%, with all viable seeds successfully germinating. The results of this study suggest that caper seeds do not exhibit characteristics indicative of physical dormancy, such as a water-impermeable seed coat.

It has been reported that pre-chilling, scarification, and treatments with gibberellic acid (GA_3) or potassium nitrate (KNO_3) are commonly employed methods to enhance the germination of dormant seeds [12, 13]. Olmez *et al*. [14] recommended that caper seeds undergo stratification at 4 °C for 20 to 60 days to address germination challenges. Wang *et al*. [15] demonstrated that soaking caper seeds in tap water for 24 hours effectively reduces hardheadedness, making it the most practical treatment among those evaluated. Additionally, Pascual *et al*. [16] found that warm stratification significantly enhances and accelerates germination in caper seeds, while refrigerated stratification also improves germination, albeit to a lesser extent.

Caper is widely distributed across various regions of Iran [17]. However, research on caper seed germination remains limited globally, and there is currently no documented methodology for enhancing germination among the diverse Iranian ecotypes. This study aimed to evaluate different techniques for breaking the dormancy of caper seeds and analyze their effects on germination rates.

MATERIALS AND METHODS

Plant Material and Experimental Setup

This study aimed to evaluate germination indices and identify the optimal treatments for breaking dormancy in nine caper ecotypes from various regions of Iran. Ripe fruits were harvested in September 2021 from these ecotypes across different Iranian provinces. In 2022, the experiment was implemented at the Seed and Plant Certification and Registration Institute (SPCRI) laboratory, following a factorial design within a completely randomized design (CRD) with four replications. The experiment incorporated three factors: the first factor consisted of nine caper ecotypes, detailed in Table 1. The second factor included nine pre-treatment methods, comprising soaking and stratification techniques. Four soaking durations were assessed: 24 hours (T1), 15 days (T2), and 30 days (T3), during which seeds were immersed in 0.1 L of tap water at room temperature [18]. Additionally, five cold-moist stratification periods were evaluated: 7 (T4), 14 (T5), 21 (T6), 28 (T7), and 35 (T8) days. Seeds were pre-soaked in distilled water for 24 hours at room temperature and then transferred to Petri dishes containing moistened white germination paper. Untreated seeds served as the control group (T9). All seeds were sealed in plastic bags and stored at 4 °C [19]. The third factor examined involved two types of planting substrates: paper and sand, with the sand being sterilized at 120 °C for 12 hours.

Table 1 Geographical distribution of *Capparis spinosa* L. ecotype across selected regions in Iran

Ecotype code	City	Province	Latitude	Longitude	Climate classification
E1	Moghan	Ardabil	39° 20′ 41.88″ N	47° 30′ 23.04″ E	Semi-tropical
E2	Hendijan	Khuzestan	30° 14′ 24″ N	49° 42′ 36″ E	Tropical
E3	Ahvaz	Khuzestan	31° 21′ 0″ N	48° 45′ 0″ E	Tropical
E4	Mehran	Ilam	33° 7′ 19.92″ N	46° 9′ 52.56″ E	Mild summer, cold winters
E5	Mahshahr	Khuzestan	30° 32′ 44.88″ N	49° 10′ 49.08″ E	Tropical
E6	Bojnord	North Khorasan	37° 28′ 12.72″ N	57° 18′ 51.48″ E	Mild summer, cold winters
E7	Ramhormoz	Khuzestan	31° 16′ 41.88″ N	49° 36′ 23.04″ E	Tropical
E8	Dalahoo	Kermanshah	43° 46′ 41.88″ N	64° 35′ 23.04″ E	Mild summer, cold winters
E9	Karaj	Alborz	35° 49′ 12″ N	50° 58′ 12″ E	Mild summer, cold winters

Mature, dark brown caper seeds were harvested from the fruits, rinsed thoroughly with tap water, and air-dried in the shade at room temperature for 48 hours. Subsequently, they were stored in airtight plastic containers at 7 ± 0.5 °C until required for experimentation. Seed viability was determined using the tetrazolium test method described by [16]. A total of 400 control seeds were used, divided equally into four replicates of 100 seeds each. Two treatment types were evaluated to promote seed germination: 1) physical dormancy removal by softening the seed coat and 2) breaking physiological dormancy through pre-chilling. Seeds were surface-sterilized in a 5% sodium hypochlorite solution for 10 minutes to prevent fungal contamination. This was followed by two rinses with sterile tap water and a final rinse with sterile distilled water.

The germination experiments were conducted within confined 9 cm × 6 cm plastic containers using the "between paper" method, which involved placing Whatman No. 1 filter paper (two layers), moistened with distilled water. A sand substrate was also employed. The experimental samples consisted of 100 seeds, randomly selected and distributed into four separate replicates of 25 seeds each. The dishes were subsequently placed in a growth chamber that provided an alternating temperature and light regime: 12 hours at 20 ± 1 °C in darkness, followed by 12 hours at 30 ± 1 °C with a photosynthetic

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photon flux density maintained at 324 μmol m^{-2} s^{-1} for a duration of 30 days. The emergence of the radicle was used as the criterion for germination at least 5 mm from the seed coat.

The parameters used to compare the germination data were as follows [20, 21].

Eq. 1. Germination percentage (GP) = (S/ T) × 100

where S and T represent the number of germinated seeds and the total number of seeds, respectively.

Eq. 2. Mean daily germination (MDG) = FGP / D

where FGP and D represent the final germination percentage (viability) and the duration (in days) to attain maximum germination during the experiment, respectively.

Eq. 3. Meantime germination (MTG) = $\sum$ (nd) / $\sum$n

where *n* denotes the number of seeds germinating on day *d*, with *d* representing the number of days post-germination, and $\sum n$ denotes the cumulative number of germinated seeds.

Eq. 4. Daily germination speed (DGS) =1 / MDG [22]

Statistical analysis

Three-way ANOVA and LSD post-hoc tests were conducted using SAS version 9.1 to analyze treatment effects. A significance level of ≤0.05 was considered statistically significant. Additionally, simple correlations between traits and cluster analysis were performed using Excel 2018 and Minitab version 19.

RESULTS

The results indicated that both the simple and interaction effects of dormancy-breaking treatments (soaking seeds in tap water and cold-moist stratification) and planting substrates significantly influenced germination indices, including germination percentage, mean daily germination, germination rate, mean time to germination, and daily germination speed across different caper ecotypes. According to the mean comparison of the main effects, E4 (Ilam ecotype) and E9 (Karaj ecotype) exhibited the highest germination percentages at 60.56% and 50.2%, respectively. In contrast, E1 (Moghan ecotype) and E5 (Mahshahr ecotype) demonstrated the lowest germination percentages at 19.35% and 25.44%. Among these ecotypes, the highest mean daily germination and germination rates were 0.50 and 0.42 seeds, and 1.82 and 1.79 seeds per day, respectively. Conversely, the lowest values for mean daily germination and germination rates were observed at 0.16 and 0.21 seeds, and 0.51 and 0.62 seeds per day, respectively. Among the pre-treatments tested, T7 (28 days of cold stratification) and T8 (35 days of cold stratification) were found to be the most effective in improving germination percentage, mean daily germination, and germination rate, while untreated seeds exhibited the lowest values for these traits. Furthermore, when comparing planting substrates, sand was more effective than paper in enhancing germination percentage, mean daily germination, and germination rate (Table 2).

Germination percentage

In six ecotypes—Hendijan (E2), Ahvaz (E3), Mehran (E4), Mahshahr (E5), Bojnord (E6), and Karaj (E9)—cold stratification for 7, 21, 28, and 35 days (T4-T8) in a sand substrate effectively overcame caper seed dormancy, resulting in the highest germination percentages. However, for the E9 and E4 ecotypes, soaking seeds in water for 24 hours (T1) followed by 14 days of stratification (T5) in sand was necessary to achieve optimal germination. Overall, the use of a sand substrate significantly outperformed paper in terms of germination percentages across all ecotypes (Fig. 1). The germination percentage across treatments ranged from 4% to 91%, indicating substantial variability. The highest germination percentage (91%) was observed in the E9 ecotype after 35 days of cold stratification (T8) in a sand substrate. In contrast, untreated seeds from all ecotypes exhibited low germination percentages, with the lowest observed in untreated seeds from the Moghan ecotype (E1) in a paper substrate (Table 3).

Mean Daily Germination

Both dormancy-breaking treatments, including cold stratification and water soaking, significantly increased the mean daily germination across all caper ecotypes. The highest mean daily germination of 0.76 was achieved in ecotype E9 after 35 days of cold stratification in a sand substrate. In ecotypes E4 and E2, 21 days of cold stratification (T6) in sand significantly raised the mean daily germination from 0.03 to 0.41 and 0.68, respectively (Fig.1). Cold stratification for various durations significantly enhanced mean daily germination in ecotypes E5, E3, and E6, increasing the values from 0.03, 0.06, and 0.07 to 0.63, 0.35, and 0.67, respectively. Additionally, a soaking duration of 15 days (T2) combined with a sand substrate significantly enhanced the mean daily germination in ecotypes E3 and E7, increasing it to 0.62. In contrast, untreated seeds from ecotypes E7, E5, E1, E8, and E2 in the paper substrate exhibited the lowest mean daily germination (Table 3).

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Table 2 Variations in seed germination indices of *Capparis spinosa* L. influenced by ecotype, different pre-treatments, and planting substrates

	Germination percentage (%)	Mean daily germination	Germination rate (seed per day)	Mean time germination (day)	Daily germination speed
Ecotype					
E1	19.35 ± 8.68	0.16 ± 0.04	0.51 ± 0.13	10.22 ± 3.3	13.94 ± 7.36
E2	35.03 ± 13.28	0.29 ± 0.10	0.93 ± 0.26	10.43 ± 2.43	6.12 ± 2.67
E3	41.69 ± 12.28	0.35 ± 0.10	1.13 ± 0.37	10.74 ± 3	4.74 ± 1.68
E4	60.56 ± 15.4	0.50 ± 0.11	1.82 ± 0.52	9.85 ± 3.04	2.9 ± 0.68
E5	25.44 ± 7.24	0.21 ± 0.09	0.62 ± 0.19	12.77 ± 4.21	6.33 ± 1.54
E6	40.56 ± 12.36	0.34 ± 0.12	1.34 ± 0.56	9.66 ± 3.25	4.32 ± 1.05
E7	40.67 ± 14.1	0.34 ± 0.12	1.07 ± 0.26	11.11 ± 2.35	4.85 ± 1.85
E8	31.36 ± 10.47	0.26 ± 0.10	0.82 ± 0.23	11.29 ± 3.39	5.29 ± 1.38
E9	50.2 ± 15.87	0.42 ± 0.12	1.79 ± 0.71	8.41 ± 3.53	4.41 ± 1.63
Pre-treatment					
T1	42.42 ± 13.79	0.35 ± 0.12	1 ± 0.32	11.84 ± 2.79	6.15 ± 1.41
T2	40.52 ± 14.79	0.34 ± 0.11	1.1 ± 0.44	10.71 ± 2.42	4.5 ± 1.86
T3	31.83 ± 10.72	0.27 ± 0.10	0.77 ± 0.17	10.56 ± 2.81	5.69 ± 1.28
T4	37.89 ± 12.27	0.32 ± 0.11	1.03 ± 0.27	10.16 ± 2.48	5.03 ± 1.98
T5	42.33 ± 13.92	0.35 ± 0.12	1.34 ± 0.43	9.44 ± 3.92	5 ± 5.46
T6	49.5 ± 17.02	0.41 ± 0.13	1.69 ± 0.53	9.41 ± 3.13	4.17 ± 1.12
T7	45.22 ± 12.81	0.38 ± 0.11	1.54 ± 0.52	9.86 ± 3.05	3.7 ± 1.53
T8	42.94 ± 10.93	0.36 ± 0.11	1.25 ± 0.42	10.17 ± 3.81	3.87 ± 1.79
T9	9.63 ± 1.01	0.08 ± 0.01	0.21 ± 0.08	12.8 5± 3.37	15.4 ± 2.92
Substrate planting					
S1	27.45 ± 7.92	0.23 ± 0.05	0.99 ± 0.26	9.14 ± 2.23	7.47 ± 2.09
S2	49.71 ± 14.56	0.41 ± 0.12	1.25 ± 0.39	11.88 ± 2.25	4.06 ± 1.77

The mean (±SD) values were compared using the least significant difference (LSD) test at a 5% probability level. The highest mean values were highlighted in bold, and the lowest mean values were italicized in the text.

E1: Moghan, E2: Hendijan, E3: Ahvaz, E4: Mehran, E5: Mahshahr, E6: Bojnord, E7: Ramhormoz, E8: Dalahoo, E9: Karaj.

T1: Soaking for 24 hours, T2: Soaking for 15 days, T3: Soaking for 30 days, T4: Cold-moist stratification for 7 days, T5: Cold-moist stratification for 14 days, T6: Cold-moist stratification for 21 days, T7: Cold-moist stratification for 28 days, T8: Cold-moist stratification for 35 days, T9: untreated seed.

S1: Paper substrate, S2: sand substrate.

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Germination Rate

The results demonstrated significant variation in seed germination rates across different caper ecotypes, influenced by pre-treatments and planting substrates. The average germination rate ranged from 0.057 to 3.90 seeds per day, indicating considerable differences attributable to the applied treatments. Germination rates were substantially boosted in all ecotypes when subjected to either cold stratification or water soaking. The highest germination rate was recorded for the E9 ecotype after 35 days of cold stratification in a sand substrate, while the lowest germination rate of 0.03 seeds per day was observed in the untreated E1 and E8 ecotypes in the sand substrate (Fig. 1). Additionally, a 21-day cold stratification treatment effectively broke dormancy and increased the germination rate in the E1, E2, E4, E6, E8, and E9 ecotypes. In contrast, a 28-day cold stratification treatment enhanced the germination rate in the E3 and E4 ecotypes (Table 3).

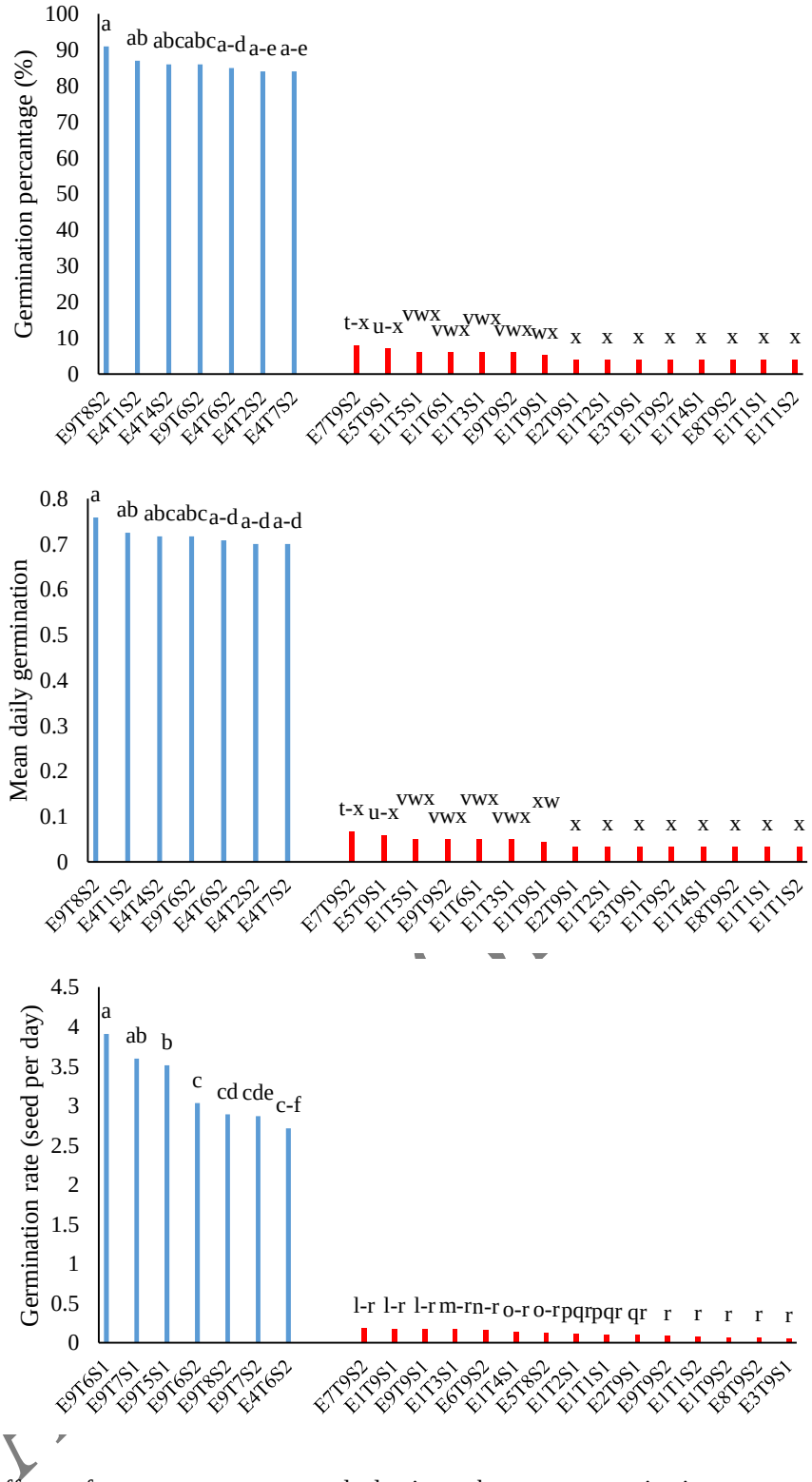

Fig. 1 Interaction effects of ecotype, treatment, and planting substrate on germination percentage (A), mean daily germination (B), and germination rate (C) of *Capparis spinosa* L. ecotypes (Mean values with corresponding upper and lower error bars are shown).

Table 3 Interaction effects of ecotype, pre-treatment, and planting substrates on seed germination indices of *Capparis spinosa* L.

E×T×S	Germination percentage (%)	Mean daily germination	Germination rate (seed per day)	Mean time germination (day)	Daily germination speed
E1T1S1	4 ± 0	0.03 ± 0	0.11 ± 0.02	9.5 ± 1.91	30 ± 0
E1T1S2	4 ± 0	0.03 ± 0	0.08 ± 0.02	17 ± 2.65	30 ± 0

E1T2S1	4 ± 0	0.03 ± 0	0.11 ± 0	9 ± 0	30 ± 0
E1T2S2	22 ± 2.31	0.18 ± 0.02	0.43 ± 0.06	13.03 ± 0.42	5.5 ± 0.58
E1T3S1	6 ± 2.31	0.05 ± 0.02	0.17 ± 0.07	9.5 ± 1.91	22.5 ± 8.66
E1T3S2	21 ± 2	0.18 ± 0.02	0.35 ± 0.03	15.33 ± 0.59	5.75 ± 0.5
E1T4S1	4 ± 0	0.03 ± 0	0.13 ± 0.05	8 ± 2.83	30 ± 0
E1T4S2	13 ± 2	0.11 ± 0.02	0.29 ± 0.06	11.54 ± 0.76	9.38 ± 1.25
E1T5S1	6 ± 2.31	0.05 ± 0.02	0.25 ± 0.09	6.13 ± 0.85	22.5 ± 8.66
E1T5S2	35 ± 3.83	0.29 ± 0.03	0.82 ± 0.13	11.12 ± 1.07	3.46 ± 0.36
E1T6S1	6 ± 2.31	0.05 ± 0.02	0.24 ± 0.07	6.63 ± 1.49	22.5 ± 8.66
E1T6S2	49 ± 6	0.41 ± 0.05	1.31 ± 0.25	9.95 ± 1.12	2.48 ± 0.3
E1T7S1	14 ± 2.31	0.12 ± 0.02	0.43 ± 0.13	9.58 ± 2.75	8.75 ± 1.44
E1T7S2	44 ± 3.27	0.37 ± 0.03	1.13 ± 0.13	10.9 ± 0.7	2.74 ± 0.2
E1T8S1	18 ± 4	0.15 ± 0.03	0.82 ± 0.41	7 ± 2.61	6.88 ± 1.25
E1T8S2	51 ± 3.83	0.43 ± 0.03	1.21 ± 0.25	11.42 ± 1.85	2.36 ± 0.17
E1T9S1	5.33 ± 2.31	0.04 ± 0.02	0.18 ± 0.02	5.67 ± 0.58	25 ± 8.66
E1T9S2	4 ± 0	0.03 ± 0	0.07± 0	14 ± 0	30 ± 0
E2T1S1	23 ± 3.83	0.19 ± 0.03	0.71 ± 0.2	10.42 ± 0.85	5.32 ± 0.84
E2T1S2	50 ± 2.31	0.42 ± 0.02	1.16 ± 0.08	11.09 ± 0.37	2.4 ± 0.11
E2T2S1	19 ± 3.83	0.16 ± 0.03	0. 7± 0.22	8.15 ± 1.06	6.5 ± 1.22
E2T2S2	68 ± 3.27	0.57 ± 0.03	1.69 ± 0.17	11.34 ± 1.35	1.77 ± 0.09
E2T3S1	9 ± 2	0.08 ± 0.02	0.33 ± 0.14	8.63 ± 2.17	13.75 ± 2.5
E2T3S2	42 ± 2.31	0.35 ± 0.02	0.99 ± 0.14	12.38 ± 1.26	2.86 ± 0.16
E2T4S1	17± 2	0.14 ± 0.02	0.6 ± 0.14	8.48 ± 1.62	7.13 ± 0.75
E2T4S2	40 ± 6.53	0.33 ± 0.05	0.8 ± 0.22	13.61 ± 2.71	3.06 ± 0.52
E2T5S1	10 ± 2.31	0.08 ± 0.02	0.29 ± 0.01	8.92 ± 1.66	12.5 ± 2.89
E2T5S2	62 ± 2.31	0.52± 0.02	1.5 ± 0.16	10.96 ± 0.97	1.94 ± 0.07
E2T6S1	18 ± 2.31	0.15 ± 0.02	0.73 ± 0.2	6.9 ± 1.15	6.75 ± 0.87
E2T6S2	81 ± 2	0.68 ± 0.02	1.98 ± 0.17	11.3 ± 0.42	1.48 ± 0.04
E2T7S1	19 ± 2	0.16 ± 0.02	0.65 ± 0.15	9.3 ± 2.96	6.38 ± 0.75
E2T7S2	55 ± 2	0.46 ± 0.02	1.4 ± 0.12	11.01 ± 0.99	2.18 ± 0.08
E2T8S1	22 ± 4	0.18 ± 0.03	0.72 ± 0.23	8.92 ± 3.38	5.63 ± 1.25
E2T8S2	64 ± 3.27	0.53 ± 0.03	1.79 ± 0.22	11.66 ± 1.23	1.88 ± 0.1
E2T9S1	4 ± 0	0.03 ± 0	0.1 ± 0.02	10.5 ± 2.12	30 ± 0
E2T9S2	12 ± 3.27	0.1 ± 0.03	0.22 ± 0.06	14.29 ± 1.11	10.63 ± 3.15
E3T1S1	29 ± 2	0.24 ± 0.02	0.98 ± 0.22	8.86 ± 1	4.15 ± 0.27
E3T1S2	69 ± 3.83	0.58 ± 0.03	1.58 ± 0.16	12.42 ± 0.66	1.74 ± 0.1
E3T2S1	35 ± 3.83	0.29 ± 0.03	1.18 ± 0.15	9.2 ± 0.84	3.46 ± 0.36
E3T2S2	74 ± 5.16	0.62 ± 0.04	2.03 ± 0.16	10.02 ± 0.93	1.63 ± 0.11
E3T3S1	29 ± 2	0.24 ± 0.02	1 ± 0.25	11.31 ± 3.45	4.15 ± 0.27
E3T3S2	43 ± 3.83	0.36 ± 0.03	1.34 ± 0.2	9.22 ± 1.24	2.81 ± 0.24
E3T4S1	26 ± 4	0.22 ± 0.03	0.88 ± 0.07	8.02 ± 1.23	4.69 ± 0.63
E3T4S2	66 ± 4	0.55 ± 0.03	1.45 ± 0.08	13.13 ± 0.45	1.82 ± 0.1
E3T5S1	17 ± 2	0.14 ± 0.02	0.61 ±0.1	7.85 ± 1.68	7.13 ± 0.75
E3T5S2	60 ± 3.27	0.5 ± 0.03	1.29 ± 0.15	12.87 ± 1.48	2 ± 0.11
E3T6S1	21 ± 2	0.18 ± 0.02	0.69 ± 0.05	8.83 ± 2.47	5.75 ± 0.5
E3T6S2	69 ± 2	0.58 ± 0.02	2 ± 0.18	9.78 ± 0.95	1.74 ± 0.05
E3T7S1	28 ± 4.62	0.23 ± 0.04	1.16 ± 0.18	7.66 ± 0.81	4.38 ± 0.72
E3T7S2	75 ± 2	0.63 ± 0.02	2.14 ± 0.2	9.97 ± 1.12	1.6 ± 0.04

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E3T8S1	26 ± 4	0.22 ± 0.03	0.75 ± 0.18	9.98 ± 2.39	4.69 ± 0.63
E3T8S2	54 ± 4	0.45 ± 0.03	0.75 ± 0.07	12.68 ± 1	2.23 ± 0.18
E3T9S1	4 ± 0	0.03 ± 0	0.06 ± 0.01	18 ± 4.36	30 ± 0
E3T9S2	16 ± 3.27	0.13 ± 0.03	0.27 ± 0.05	15.33 ± 1.2	7.75 ± 1.66
E4T1S1	66 ± 4	0.55 ± 0.03	1.57 ± 0.29	12.67 ± 2.1	1.82 ± 0.1
E4T1S2	87 ± 6	0.73 ± 0.05	1.82 ± 0.1	12.66 ± 0.78	1.38 ± 0.09
E4T2S1	80 ± 3.27	0.67 ± 0.03	2.63 ± 0.39	12.54 ± 1.28	1.5 ± 0.06
E4T2S2	84 ± 3.27	0.7 ± 0.03	2.33 ± 0.16	9.8 ± 0.6	1.43 ± 0.06
E4T3S1	36 ± 4.62	0.3 ± 0.04	1.09 ± 0.21	9.99 ± 2.58	3.38 ± 0.43
E4T3S2	82 ± 6.93	0.68 ± 0.06	0.77 ± 0.16	16.62 ± 2	1.47 ± 0.12
E4T4S1	26 ± 2.31	0.22 ± 0.02	0.94 ± 0.05	9.1 ± 1.79	4.64 ± 0.41
E4T4S2	86 ± 7.66	0.72 ± 0.06	2.36 ± 0.34	10.03 ± 1.26	1.4±0.12
E4T5S1	58 ± 5.16	0.48 ± 0.04	2.49 ± 0.39	7.99 ± 0 .8	2.08 ± 0.19
E4T5S2	78 ± 5.16	0.65 ± 0.04	2.45 ± 0.23	8.71 ± 0.51	1.54 ± 0.1
E4T6S1	44 ± 5.66	0.37 ± 0.05	2.26 ± 0.18	6.7 ± 1.76	2.76 ± 0.33
E4T6S2	85 ± 6	0.71 ± 0.05	2.71 ± 0.15	8.31 ± 0.19	1.42 ± 0.1
E4T7S1	55 ± 3.83	0.46 ± 0.03	2.61 ± 0.18	6.16 ± 0.58	2.19 ± 0.15
E4T7S2	84 ± 3.27	0.7 ± 0.03	2.63 ± 0.31	8.71 ± 0.87	1.43 ± 0.06
E4T8S1	46 ± 7.66	0.38 ± 0.06	1.8 ± 0.42	4.43 ± 0.61	2.66 ± 0.42
E4T8S2	67 ± 6	0.56 ± 0.05	1.6 ± 0.13	11.7 2 ± 0.91	1.8 ± 0.16
E4T9S1	13 ±3.83	0.11 ± 0.03	0.38 ± 0.16	10.67 ± 2.97	10 ± 3.54
E4T9S2	13 ± 2	0.11 ± 0.02	0.33 ± 0.05	10.52 ± 0.44	9.38 ± 1.25
E5T1S1	38 ± 5.16	0.32 ± 0.04	1.05 ± 0.14	6.66 ± 1.15	3.2 ± 0.44
E5T1S2	36 ± 3.27	0.3 ± 0.03	1 ± 0.09	10.73 ± 1.46	3.35 ± 0.31
E5T2S1	35 ± 3.83	0.29 ± 0.03	0.92 ± 0.19	10.81 ± 1.1	3.46 ± 0.36
E5T2S2	30 ± 2.31	0.25 ± 0.02	0.68 ± 0.08	12.87 ± 2.48	4.02 ± 0.31
E5T3S1	19 ± 3.83	0.16 ± 0.03	0.62 ± 0.16	10.69 ± 0.38	6.5 ± 1.22
E5T3S2	23 ± 2	0.19 ± 0.02	0.57 ± 0.1	11.24 ± 1.68	5.25 ± 0.5
E5T4S1	18 ± 2.31	0.15 ± 0.02	0.56 ± 0.04	9.71 ± 1.35	6.75 ± 0.87
E5T4S2	42 ± 2.31	0.35 ± 0.02	0.96 ± 0.13	12.11 ± 1.42	2.86 ± 0.16
E5T5S1	18 ± 2.31	0.15 ± 0.02	0.53 ± 0.07	10.26 ± 1.26	6.75 ± 0.87
E5T5S2	40 ± 3.27	0.33 ± 0.03	0.64 ± 0.14	17 ± 2.2	3.02 ± 0.25
E5T6S1	29 ± 2	0.24 ± 0.02	0.91 ± 0.08	12.05 ± 0.6	4.15 ± 0.27
E5T6S2	36 ± 4.62	0.3 ± 0.04	0.8 ± 0.15	18.74 ± 0.19	3.38 ± 0.43
E5T7S1	18 ± 2.31	0.15 ± 0.02	0.36 ± 0.04	15.69 ± 3.85	6.75 ± 0.87
E5T7S2	31 ± 3.83	0.26 ± 0.03	0.46 ± 0.09	18.92 ± 2.07	3.91 ± 0.46
E5T8S1	18 ± 4	0.15 ± 0.03	0.52 ± 0.1	9.5 ± 3.85	6.88 ± 1.25
E5T8S2	10 ± 2.31	0.08 ± 0.02	0.13 ± 0.03	21.13 ± 2.19	12.5 ± 2.89
E5T9S1	7 ± 2	0.06 ± 0.02	0.21 ± 0.1	10.13 ± 3.12	18.75 ± 7.5
E5T9S2	10 ± 2.31	0.08 ± 0.02	0.25 ± 0.08	11.58 ± 1.83	12.5 ± 2.89
E6T1S1	38 ± 5.16	0.32 ± 0.04	0.83 ± 0.13	13.45 ± 0.68	3.2 ± 0.44
E6T1S2	23 ± 3.83	0.19 ± 0.03	0.41 ± 0.12	14.93 ± 2.3	5.32 ± 0.84
E6T2S1	18 ± 4	0.15 ± 0.03	0.74 ± 0.21	6.88 ± 1.25	6.88 ± 1.25
E6T2S2	18 ± 4	0.15 ± 0.03	0.54 ± 0.13	8.67 ±1.03	6.88 ± 1.25
E6T3S1	24 ± 5.66	0.2 ± 0.05	0.78 ± 0.21	8.2 ± 1.07	5.19 ± 1.07
E6T3S2	35 ± 3.83	0.29 ± 0.03	0.9 ± 0.15	10.32 ± 0.78	3.46 ± 0.36
E6T4S1	25 ± 3.83	0.21 ± 0.03	1.1 ± 0.27	6.2 ± 1.02	4.89 ± 0.81
E6T4S2	66 ± 7.66	0.55 ± 0.06	1.66 ± 0.25	10.34 ± 1.25	1.84 ± 0.23

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E6T5S1	40 ± 5.66	0.33 ± 0.05	1.68 ± 0.35	6.88 ± 1.47	3.04 ± 0.39
E6T5S2	75 ± 6.83	0.63 ± 0.06	2.19 ± 0.46	9.63 ± 1.58	1.61 ± 0.14
E6T6S1	51 ± 6.83	0.43 ± 0.06	2.52 ± 0.23	7.53 ± 0.64	2.38 ± 0.31
E6T6S2	80 ± 4.62	0.67 ± 0.04	2.66 ± 0.15	8.22 ± 1.05	1.5 ± 0.09
E6T7S1	47 ± 10.52	0.39 ± 0.09	2.2 ± 0.61	5.71 ± 0.41	2.65 ± 0.61
E6T7S2	76 ± 7.3	0.63 ± 0.06	2.55 ± 0.44	8.3 ± 0.69	1.59 ± 0.15
E6T8S1	48 ± 6.53	0.4 ± 0.05	1.86 ± 0.47	7.76 ± 1.47	2.54 ± 0.35
E6T8S2	43 ± 6	0.36 ± 0.05	1.08 ± 0.17	10.51 ± 1.04	2.83 ± 0.35
E6T9S1	13 ± 2	0.11 ± 0.02	0.27 ± 0.14	14.04 ± 2.73	9.38 ± 1.25
E6T9S2	10 ± 2.31	0.08 ± 0.02	0.16 ± 0.04	16.25 ± 0.87	12.5 ± 2.89
E7T1S1	35 ± 3.83	0.29 ± 0.03	1.01 ± 0.26	10.72 ± 3.89	3.46 ± 0.36
E7T1S2	68 ± 3.27	0.57 ± 0.03	1.6 ± 0.18	11.56 ± 0.52	1.77 ± 0.09
E7T2S1	15 ± 3.83	0.13 ± 0.03	0.53 ± 0.18	8.58 ± 0.72	8.38 ± 1.97
E7T2S2	74 ± 5.16	0.62 ± 0.04	2.02 ± 0.14	10.19 ± 0.08	1.63 ± 0.11
E7T3S1	17 ± 2	0.14 ± 0.02	0.63 ± 0.18	10.8 ± 1.07	7.13 ± 0.75
E7T3S2	51 ± 3.83	0.43 ± 0.03	1.26 ± 0.2	11.41 ± 1.47	2.36 ± 0.17
E7T4S1	14 ± 2.31	0.12 ± 0.02	0.34 ± 0.12	10.58 ± 4.83	8.75 ± 1.44
E7T4S2	68 ± 4.62	0.57 ± 0.04	1.65 ± 0.18	11.05 ± 0.48	1.77 ± 0.12
E7T5S1	24 ± 3.27	0.2 ± 0.03	0.7 ± 0.32	11.72 ± 4.47	5.07 ± 0.7
E7T5S2	71 ± 2	0.59 ± 0.02	1.65 ± 0.19	13.56 ± 0.94	1.69 ± 0.05
E7T6S1	23 ± 3.83	0.19 ± 0.03	0.74 ± 0.23	10.51 ± 3.03	5.32 ± 0.84
E7T6S2	75 ± 3.83	0.63 ± 0.03	1.92 ± 0.32	10.93 ± 1.16	1.6 ± 0.08
E7T7S1	28 ± 3.27	0.23 ± 0.03	0.75 ± 0.15	11.16 ± 2.47	4.33 ± 0.51
E7T7S2	53 ± 2	0.44 ± 0.02	1.34 ± 0.17	11.31 ±1.43	2.27 ± 0.08
E7T8S1	34 ± 4	0.28 ± 0.03	1.14 ± 0.41	9.56 ± 2.74	3.56 ± 0.38
E7T8S2	63 ± 2	0.53 ± 0.02	1.55 ± 0.19	11.04 ± 1.21	1.91 ± 0.06
E7T9S1	11 ± 2	0.09 ± 0.02	0.22 ± 0.08	13.5 ± 2.53	11.25 ± 2.5
E7T9S2	8 ± 0	0.07 ± 0	0.18 ± 0.02	11.88 ± 1.38	15 ± 0
E8T1S1	62 ± 7.66	0.52 ± 0.06	1.45 ± 0.26	12.74 ± 1.2	1.96 ± 0.23
E8T1S2	66 ± 7.66	0.55 ± 0.06	1.16 ± 0.15	15.92 ± 1.78	1.84 ± 0.23
E8T2S1	26 ± 4	0.22 ± 0.03	0.55 ± 0.06	13.17 ± 2.9	4.69 ± 0.63
E8T2S2	26 ± 4	0.22 ± 0.03	0.45 ± 0.05	14.81 ± 1.53	4.69 ± 0.63
E8T3S1	34 ± 4	0.28 ± 0.03	1.03 ± 0.32	8.81 ± 3.08	3.56 ± 0.38
E8T3S2	42 ± 4	0.35 ± 0.03	1.14 ± 0.23	10.08 ± 1.76	2.88 ± 0.25
E8T4S1	22 ± 5.16	0.18 ± 0.04	0.59 ± 0.11	10.61 ± 1.51	5.7 ± 1.39
E8T4S2	40 ± 6.53	0.33 ± 0.05	0.93 ± 0.27	11.69 ± 1.26	3.06 ± 0.52
E8T5S1	23 ± 3.83	0.19 ± 0.03	1.02 ± 0.19	6.51 ± 1.48	5.32 ± 0.84
E8T5S2	19 ± 3.83	0.16 ± 0.03	0.36 ± 0.11	14 ± 1.22	6.5 ± 1.22
E8T6S1	28 ± 7.3	0.23 ± 0.06	1.59 ± 0.33	1.82 ± 0.47	4.52 ± 1.21
E8T6S2	28 ± 8	0.23 ± 0.07	0.42 ± 0.06	17.73 ± 3.38	4.5 ± 1
E8T7S1	18 ± 8.33	0.15 ± 0.07	0.74 ± 0.26	6.4 ± 1.36	8.2 ± 4.72
E8T7S2	32 ± 0	0.27 ± 0	0.71 ± 0.27	14.5 ± 6.01	3.75 ± 0
E8T8S1	32 ± 6.53	0.27 ± 0.05	0.83 ± 0.34	9.72 ± 2.85	3.88 ± 0.83
E8T8S2	34 ± 7.66	0.28 ± 0.06	0.91 ± 0.21	10.32 ± 2.34	3.69 ± 0.94
E8T9S1	8 ± 0	0.07 ± 0	0.18 ± 0.02	11.88 ± 1.38	15 ± 0
E8T9S2	4 ± 0	0.03 ± 0	0.06 ± 0	16 ± 0	30 ± 0
E9T1S1	10 ± 4	0.08 ± 0.03	0.23 ± 0.06	11 ± 2.16	13.13 ± 3.75
E9T1S2	46 ± 7.66	0.38±0.06	0.98 ± 0.19	12.1 ± 1.04	2.66 ± 0.42

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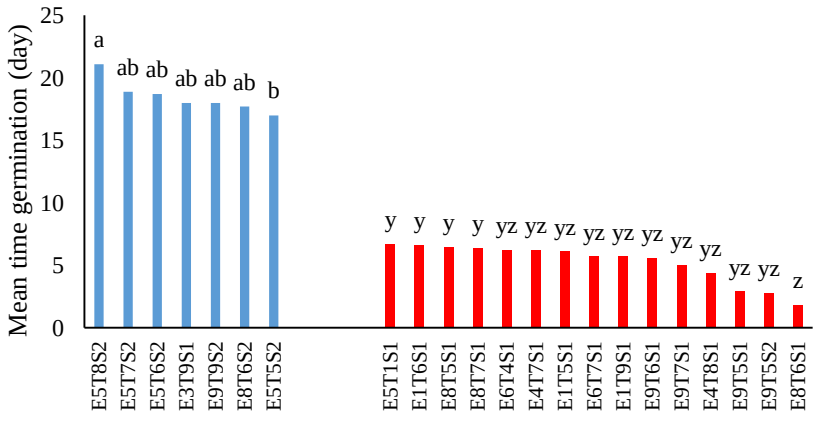
E9T2S1	24 ± 6.53	0.2±0.05	0.58 ± 0.23	9.75 ± 1.29	5.31 ± 1.57
E9T2S2	50 ± 7.66	0.4 2± 0.06	1 ± 0.21	12.76 ± 0.93	2.45 ± 0.41
E9T3S1	22 ± 7.66	0.18 ± 0.06	0.33 ± 0.14	7.54 ± 2.02	5.94 ± 1.88
E9T3S2	38 ± 13.66	0.32 ± 0.11	0.52 ± 0.17	8.11 ± 0.98	3.47 ± 1.21
E9T4S1	42 ± 4	0.35 ± 0.03	1.53 ± 0.28	7.79 ± 1.55	2.88 ± 0.25
E9T4S2	50 ± 4	0.42 ± 0.03	1.36 ± 0.19	9.85 ± 1.6	2.41 ± 0.18
E9T5S1	60 ± 8	0.5 ± 0.07	3.51 ± 0.47	2.91 ± 0.34	2.03 ± 0.31
E9T5S2	66 ± 6.93	0.55 ± 0.06	2.08 ± 0.25	2.84 ± 1.42	1.83 ± 0.18
E9T6S1	82 ± 10.07	0.68 ± 0.08	3.91 ± 0.49	5.64 ± 0.27	1.48 ± 0.17
E9T6S2	86 ±7.66	0.72 ± 0.06	3.03 ± 0.4	7.84 ± 0.26	1.4 ± 0.12
E9T7S1	60 ± 4.62	0.5 ± 0.04	3.59 ± 0.81	4.97 ± 0.71	2.01 ± 0.15
E9T7S2	77 ± 6	0.64 ± 0.05	2.87 ± 0.19	7.31 ± 0.62	1.57 ± 0.12
E9T8S1	52 ± 10.33	0.4 3 ± 0.09	2.24 ± 0.72	7.35 ± 2.09	2.38 ± 0.49
E9T8S2	91 ± 5.03	0.76 ± 0.04	2.89 ± 0.31	8.33 ± 0.48	1.32 ± 0.08
E9T9S1	9.33 ± 4.62	0.08 ± 0.04	0.18 ± 0.09	13.33 ± 0.88	16.67 ± 11.55
E9T9S2	6 ± 2.83	0.05 ± 0.02	0.09 ± 0.05	18 ± 1.41	22.5 ± 10.61
LSD =0.05	7.74	0.10	0.48	1.37	4.65
			Statistical analysis		
E× T × S	**	**	**	**	**
Error	0.068	3.31	0.051	0.0015	22.03
CV (%)	12.16	12.18	20.17	17.32	11.97

The mean (±SD) values were compared using the least significant difference (LSD) test at a 5% probability level.
E1: Moghan, E2: Hendijan, E3: Ahvaz, E4: Mehran, E5: Mahshahr, E6: Bojnord, E7: Ramhormoz, E8: Dalahoo, E9: Karaj.
T1: Soaking for 24 hours, T2: Soaking for 15 days, T3: Soaking for 30 days, T4: Cold-moist stratification for 7 days, T5: Cold-moist stratification for 14 days, T6: Cold-moist stratification for 21 days, T7: Cold-moist stratification for 28 days, T8: Cold-moist stratification for 35 days, T9: untreated seed.
S1: Paper substrate, S2: sand substrate.

Mean Time Germination

Mean germination time was significantly decreased in all ecotypes when subjected to either cold stratification or water soaking treatments. The highest mean time to germination recorded was 21.1 days for the E5 caper ecotype following 35 days of cold stratification in a sand substrate (Fig. 2). In the E2, E3, E6, and E9 ecotypes, maximum mean times to germination of 14.29, 18, 16.25, and 18 days, respectively, were observed under control treatments in the sand substrate. Conversely, the lowest mean time to germination of 1.8 days was recorded for the E8 ecotype after 21 days of cold stratification in paper substrates (Table 3).



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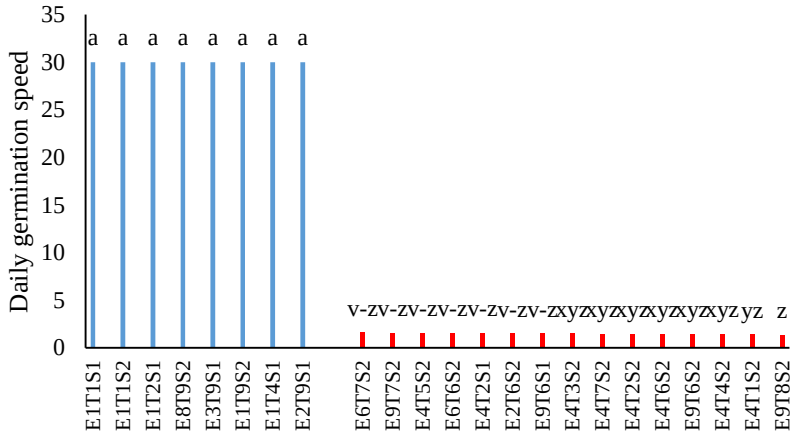

Fig. 2. Interaction Effects of ecotype, treatment, and planting substrate on mean time germination (A) and daily germination speed (B) of *Capparis spinosa* L. ecotypes (Mean values with corresponding upper and lower error bars are shown).

Daily Germination Speed

Based on the mean values, the highest daily germination speed was recorded for the E1 ecotype following a 24-hour water-soaking treatment in both sand and paper substrates. Additionally, untreated seeds of the E8, E3, and E1 ecotypes exhibited the highest daily germination speed averages in sand, paper, and sand substrates, respectively (Fig. 2). Conversely, the lowest daily germination speed was observed in the E9 ecotype, which underwent 35 days of cold stratification in a sand substrate. In the E2, E3, E5, and E8 ecotypes, the highest daily germination speeds were noted in untreated caper seeds using the paper germination method. Similarly, untreated seeds of E6 and E7 in sand substrates also demonstrated elevated daily germination speed values. Notably, the daily germination speed for the E9 ecotype improved after a 24-hour water-soaking treatment using the paper germination method (Table 3).

Coefficient Correlation

A positive and significant correlation was observed between mean daily germination and germination percentage (r = 0.99**). Additionally, significant positive correlations were identified between germination rate and both germination percentage (r = 0.87**) and mean daily germination (r = 0.87**). Conversely, a negative and significant correlation was found between mean time to germination and both germination percentage and mean daily germination (Table 4).

Cluster Analysis

The cluster analysis results and the grouping of the nine ecotypes of caper are presented in Table 5 and Fig. 3. The findings indicate that the nine caper ecotypes were classified into three distinct groups. The first group consisted of two ecotypes, E1 and E5, which represented 22.2% of the total and exhibited superior mean time to germination and daily germination speed. The second group included five ecotypes—E2, E8, E3, E7, and E6—accounting for 55.5% of the total, with no significant differences observed in germination traits among its members. The third group, also comprising 22.2% of the total, was characterized by higher germination percentage, mean daily germination, and germination rate, including the two ecotypes E4 and E9.

Table 4 Pearson correlation coefficients between seed germination indices of *Capparis spinosa* L.

	Germination percentage	Mean daily germination	Germination rate	Daily germination speed	Mean time germination
Germination percentage	1				
Mean daily germination	0.99**	1			
Germination rate	0.87**	0.87**	1		
Daily germination speed	-0.13ns	-0.13ns	-0.44*	1	
Mean time germination	-0.69**	-0.69**	-0.61**	0.15ns	1

ns: non-significant;* and **: significant at α=0.05 and 0.01%.

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Table 5 Cluster analysis and grouping of studied variables in different ecotypes of *Capparis spinosa* L.

Variable	Cluster-1	Cluster-2	Cluster-3	Grand centroid
Germination percentage	22.3996	37.8607	55.3792	38.3179
Mean daily germination	0.1867	0.3155	0.4615	0.3193
Germination rate	0.5645	1.0583	1.8045	1.1144
Mean time germination	11.4925	10.6463	9.1302	10.4974
Daily germination speed	10.1349	5.0634	3.6557	5.8776
Number of ecotypes	2	5	2	-

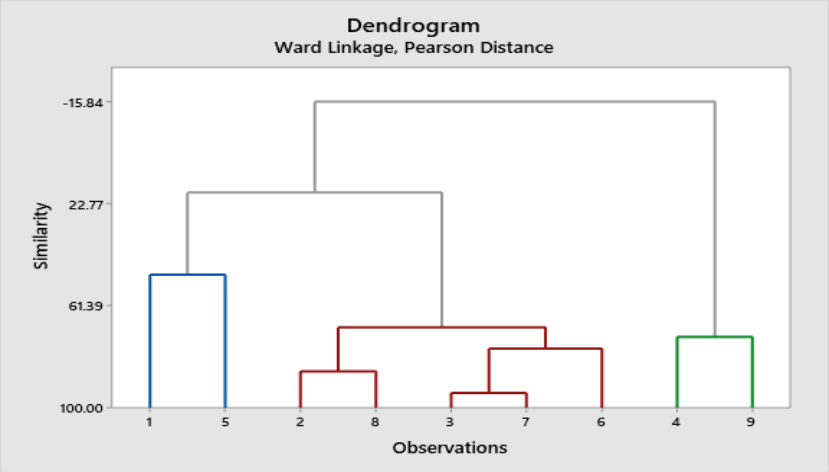

Fig. 3 Cluster analysis dendrogram of different *Capparis spinosa* L. ecotypes
1: Moghan, 2: Hendijan, 3: Ahvaz, 4: Mehran, 5: Mahshahr,E6: Bojnord, 7: Ramhormoz, 8: Dalahoo, 9: Karaj

DISCUSSION

This research aimed to assess germination indices and implement dormancy-breaking treatments across nine ecotypes of caper in Iran, utilizing various pre-treatments and germination substrates. The findings indicate that caper plants grown from fresh seeds do not consistently develop fully, with only approximately 10% of the seeds germinating. As reported by Pascual *et al*. [23], the germination percentage of caper seeds during the initial two to three months following planting was observed to be only 5%. In this study, untreated seeds exhibited a germination percentage of 9.63% and a germination rate of 0.21 seeds per day. Notably, these values increased significantly across all pre-treatment conditions. The most pronounced enhancements were recorded for the 21-day and 28-day scarification treatments (T6 and T7), resulting in germination percentages that were 5.14 and 4.69 times higher, respectively. Furthermore, the germination rates for these treatments increased by factors of 8.04 and 7.33, respectively (Table 2). Several authors have conducted studies to enhance seed germination in caper by addressing potential physical dormancy through various scarification techniques, including mechanical, chemical, thermal, and biological methods [2, 16, 17, 24]. Additionally, physiological dormancy has been effectively mitigated using treatments with GA_3 and potassium nitrate, as reported by Labbafi *et al*. [25], Foschi *et al*. [19], and Nowruzian and Aalami [12].

Kaya *et al*. [26] reported a significant effect of pre-chilling and cold stratification periods on the germination of *C. ovata* seeds. Cold stratification is essential for many species from temperate regions, as it facilitates dormancy breaks during winter, allowing seeds to germinate in spring or early summer, coinciding with optimal conditions for seedling growth [27,28]. In the current study, three caper ecotypes (E4, E7, and E8), seeds with physically dormant, water-impermeable seed coats showed the highest germination percentage after soaking in water for 24 hours and subsequently placing the seeds in a sand substrate for 15 days. In contrast, five other ecotypes (E1, E3, E5, E6, and E9) exhibited varying responses to cold stratification at 4 °C for 21, 28, and 35 days in the sand substrate. Both the specific ecotype and the initial dormancy status of the fresh seeds significantly influenced the outcomes of dormancy break and subsequent germination.

A significant increase in seed germination rates was observed across all seven caper ecotypes following cold stratification at 4°C, with germination occurring in two ecotypes while the seeds were still soaking in water. Early germination during the cold season facilitates the establishment of seedlings at the onset of the growing season. The responses of six caper ecotypes to cold stratification indicate that physiological dormancy is the primary factor contributing to the low germination percentage and rate. Consequently, low winter temperatures achieved through cold stratification likely play a crucial role in promoting the distribution of caper ecotypes in Iran. Additionally, cold and wet storage (stratification) is

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essential for breaking dormancy and enhancing germination in various other plant species, such as *Spartina alterniflora*, which requires a minimum duration of 4–8 weeks at 4 °C [29]. The mechanism of cold stratification encompasses several key physiological processes that aid in seed germination. Primarily, it alleviates physiological dormancy by lowering the levels of inhibitory hormones and enhancing metabolic activity within the seeds [30]. Additionally, cooler temperatures stimulate respiration and the utilization of stored nutrients, supplying the energy required for germination [31].

Additionally, maintaining adequate moisture during this period is crucial to ensure that the seeds remain hydrated and ready for germination [28]. This combination of cold temperatures and moisture creates optimal conditions that significantly enhance the germination potential of seeds, particularly in species like *Capparis*. The significance of cold stratification has been well-documented across various species, including *Primula beesiana* [32], *Aster tripolium*, and *Triglochin maritimum* [33], as well as *Guizotia scabra*, *Parthenium hysterophorus*, and *Verbesina encelioides* [34]. Cold stratification provides several advantages. Firstly, it can enhance superoxide dismutase activity within the seeds, facilitating the removal of accumulated superoxide radicals (O_2^-), a crucial adaptation to the low-temperature conditions experienced during winter. Concurrently, a significant decrease in abscisic acid (ABA) levels, a known germination inhibitor, occurs during cold stratification, aligning with an observed increase in germination capacity [36].

CONCLUSION

In conclusion, this experiment successfully evaluated two straightforward and effective methods for breaking seed dormancy and enhancing the germination of caper seeds: soaking seeds in tap water and cold stratification. The findings indicate that both physical factors, such as the hard seed coat, and physiological factors, including the immature embryo, contribute to seed dormancy in caper seeds. The pre-chilling stratification treatment facilitated embryo maturation and allowed for the accumulation of essential stimulatory substances necessary to initiate germination. The highest germination percentage was recorded for the E9 caper ecotype after 35 days of cold stratification in a sand substrate, resulting in a significant increase in germination rates. Additionally, soaking seeds in water effectively leached out germination inhibitors, thereby promoting metabolic processes that regulate seed germination. The seed coat and the mucilage formed on its surface represent the primary barriers to germination. Soaking seeds in water for varying durations weakened both the seed coat and mucilage, facilitating water entry and initiating the germination process. The dormancy-breaking treatment of 24 hours of soaking in water, combined with a sand substrate, notably improved the germination value in the E4 and E8 caper ecotypes.

The article presents innovative research on enhancing seed germination in *Capparis spinosa* L. (caper) through various pre-treatment methods, specifically focusing on cold stratification and soaking durations. This study addresses the critical issue of low and inconsistent germination rates caused by physical and physiological dormancy, which has impeded the large-scale cultivation of this economically significant plant. By experimenting with different soaking times (24 hours, 15 days, and 30 days) and cold stratification periods (ranging from 7 to 35 days), the authors identified optimal conditions that significantly improved germination percentages and rates across nine ecotypes from Iran. Notably, the findings revealed that cold stratification in a sand substrate was the most effective method, achieving a remarkable 91% germination rate in one ecotype after 35 days. This research not only contributes valuable insights into seed dormancy mechanisms but also offers practical solutions for enhancing the cultivation of capers, thereby supporting agricultural practices in arid regions.

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