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Research Article

The Effects of Mother Tree Size and Flowering Period on Salt Tolerance of *Tamarix Austromongolica* Seeds in the Yellow River Delta

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Abstract

*Tamarix austromongolic*a is a two-season flowering plant with a spring and summer flowering period and both flowering periods produce viable seeds, with flowering extending from mid-May to October and seed maturation from mid-June to October. In order to reveal the germination pattern of *T. austromongolica* seeds in the Yellow River Delta region, this study investigated the salt tolerance of seeds of different age mother trees during the spring and summer flowering periods in the region. Spring and summer flowering seeds of different ground diameter mother trees in the *T. austromongolica* forest in the region were collected and subjected to seed germination experiments under different concentrations of NaCl solution. The results showed that the diameter grade of the mother tree had a significant impact on seed germination, but had no significant effect on early seedling growth (except for the root length of offspring seedlings during the summer flowering period). As the diameter level of the mother tree increases, the germination rate, germination index, and vitality index of seeds during the spring flowering period first increase and then decrease, with the highest values observed at a ground diameter of 10.0-19.9cm. However, the above indicators of summer flowering seeds gradually decreased with the diameter increasing. There were significant differences in salt tolerance of seeds during different flowering periods. The germination rate and germination index of spring flowering seeds significantly decreased between 11‰ and 15‰, but the effect of salt concentration on summer flowering seed germination was not significant. For the early growth of seedlings, as the salt concentration increased, the root length and seedling height of seed offspring seedlings during spring and summer flowering periods decreased after being higher than 5‰. Compared with spring flowering seeds, summer flowering seeds had higher germination ability and seedling weight under salt stress. Therefore, the salt tolerance of the seeds of *T. austromongolica* was determined by the size of the mother tree and the flowering period, which is an important strategy for its adaptation to the Yellow River Delta region.

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Keywords

Yellow River Delta, Mother Tree Size, Salt Stress, Florescence, Seed Germination, Seedling Size

1. Introduction

In the Yellow River Delta region, the water table is shallow and mineralization is high, and groundwater salts are easy to collect on the surface [1]. Soil salinization is a process in which salts from the soil subsoil or groundwater rise to the surface with capillary water, and water evaporation causes salts to accumulate in the surface soil [2]. The formation of soil salinization is influenced by a combination of natural and anthropogenic factors, such as climate, hydrology, geology, over-fertilization and indiscriminate cutting [3]. Excessive salinity in the soil can cause harm to plants, which is mainly divided into osmotic stress and ionic toxicity, the water potential in the soil is usually lower than that of the plant cells, and the water in the body of the plant in this environment is easy to seep out of the body, resulting in water shortage; salt stress breaks the dynamic balance between the ions in the soil, resulting in unbalanced ion uptake in the plant [4]. Salt stress inhibits plant seed germination and early seedling growth, but for some plants with strong salt tolerance, lower salt concentration has a certain promotion effect on their seed germination and seedling growth, such as Chinese *T. austromongolica* [3], salt pawpaw, salt spicebush [5].

Characteristics of the mother tree (age, diameter class size) are important factors affecting seed quality and seed germination [6]. Seed germination is key to population renewal and species continuation in the life history of seed plants [7-9]. The age of the mother tree is one of the main reasons for the differentiation of seed characteristics within populations, and the reproductive age of the mother tree affects the maturity of the mother tree itself and the efficiency of its utilization of resources; although the intraspecific habitats are relatively uniform, there are subtle differences in the spatial positions actually occupied by different plants, which affects the plants' access to resources, and consequently, their growth and reproduction [10, 11]. Seeds of mother tree individuals of different ages (young individuals, middle-aged and old individuals) showed differences in their adaptive capacity, and with the increase of mother tree diameter at breast height (DBH), the seed mass of A. Tetracentron sinense showed a tendency to increase firstly and then decrease later [12]. It has also been shown that the seed germination rate of Emmenopterys henryi is not related to the size of the mother tree [13]. Overall, the effect of mother tree size on seed quality is not consistent and is affected by a combination of tree species, growth environment and other aspects.

Tamarix austromongolica belongs to the genus Tamarix in the family Tamarixaceae, a deciduous shrub or small tree, which is a dominant species in the Yellow River Delta region, with the ecological functions of windbreak and sand fixation, soil and water conservation and saline-alkaline land improvement [14]. Studies have found that salinity is a key factor affecting its growth and distribution in the region [15, 16]. Two-season flowering is a phenomenon in which a plant has two independent flowering periods within a single growing season, or a long-term sustained flowering period with two peak flowering periods, and can produce mature seeds throughout the flowering period [17]. *T. austromongolic*a is a two-season flowering plant with a spring and summer flowering period and both flowering periods produce viable seeds, with flowering extending from mid-May to October and seed maturation from mid-June to October [18, 19]. In this paper, we investigated the germination process of spring and summer flowering seeds of *T. austromongolica* with different mother tree sizes under different salt concentrations, to clarify the effect of mother tree size on seed salt tolerance, to increase the knowledge of the germination ability of two-season flowering seeds, to reveal the mechanism of *T. austromongolica*'s adaptability, and to provide scientific basis for the protection and rational utilization of the forest stand.

2. Materials and Methods

2.1. Overview of the Study Area

The seeds used in the experiment were collected from the natural forest of *T. austromongolica* distributed around the Wetland City Construction Promotion Center in Dongying City, Shandong Province. The area has a continental monsoon climate, with rain and heat in the same season and four distinct seasons. The average annual temperature ranges from 11.7 to 12.6 ℃, the annual precipitation is 550-600 mm, mostly concentrated in the summer, the precipitation varies greatly from year to year, which makes it easy to form droughts and floods, and the frost-free period lasts 206 d. The average annual temperature ranges from 11.7 to 12.6 ℃.

2.2. Seed Collection

The seeds used in the experiment were randomly collected on July 23 and September 21, 2019 in the natural forest of *T. austromongolica* at three ground diameter classes, and 10 plants were collected at each diameter class. The ground diameter of diameter class I was 3.4-9.9 cm, that of diameter class II was 10-19.9 cm, and that of diameter class III was 20-35 cm. Seeds from sunny branches in the middle of the plant were taken and brought back to the laboratory to be dried in the shade for the germination test.

2.3. Experimental Design

Set the concentration of NaCl solution were control 0‰, 3‰, 5‰, 7‰, 9‰, 11‰, 13‰, 15‰ of eight gradients; according to the mother tree of *T. austromongolica* ground diameter is divided into I, II, III, the three grades of *T. austromongolica* have the seeds of spring and summer flowering period, a total of 48 treatments. Seeds were randomly selected (without going to seed hairs), 50 seeds in 1 group, 4 replications. The seeds were placed in Petri dishes with a diameter of 90 mm padded with 2 layers of filter paper and kept the filter paper moist, and the Petri dishes were placed in a light incubator sterilized with 75% alcohol, with the temperature set at 25 ℃, 12 h of light and 12 h of darkness, and incubated for 8 d. During the process of seed germination, the number of seed germination was observed and recorded every 24 h. The number of seed germination was recorded by weighing and replenishment of seed loss per day, and the number of seed loss per day was recorded by weighing and replenishment of seed loss per day. Weighing method was used to replenish the water lost every day to keep the filter paper and seeds moist, and to maintain the relative stability of the concentration of each treatment during the experiment. At the end of the experiment, five representative seedlings with moderate growth were selected for each treatment to measure seedling height and root length; 10 seedlings were selected for each treatment and weighed separately for fresh weight using a 1-in-10,000 balance. Then germination rate (GR), mean germination time (MGT), germination index (GI) and vigor index (VI) were calculated.

GR (germination rate) = $G1/N\times100\%$, where: G1 is the number of germination; N is the total number of seeds for test.

MGT (mean germination time) = $(\sum D\times n)/\sum n$, where: n is the number of normal germinated grains on each corresponding day; D is the number of days from seed placement.

GI (germination index) = $\sum (Gt/Dt)$, where: Gt is the number of net germination on each day; Dt is the corresponding germination day.

VI (vigor index) = $S \times GI$, where: S is the fresh weight of seedlings; GI is the germination index.

2.4. Data Processing and Analysis

Data were organized and statistically analyzed using Excel with SPSS 19.0 software. Two-factor analysis of variance (ANOVA) was performed on germination index, root length and seedling height of spring flowering seeds and summer flowering seeds using SPSS 19.0. If the differences were significant, multiple comparisons were then performed using the least significant difference (LSD) method, and the level of significant difference test for analysis was $P < 0.05$.

3. Results and Analysis

3.1. Effects of Mother Tree Size and Salt Stress on Seed Germination of Flowering Seeds in Two Seasons of T. Austromongolica

For spring flowering seeds, mother tree size had a significant effect on germination rate, germination index and vigor index, and a non-significant effect on average germination time; salt stress had a highly significant effect on germination rate and germination index, and a significant effect on average germination time and vigor index (Table 1). The mother tree size was II > III > I in germination rate, germination index and vigor index ($P<0.05$), and the average germination time was shortest in diameter class I. Germination rate and germination index did not differ significantly between individual salt concentrations below 9‰ but were significantly higher than 11-15‰ treatments.

For summer-flowering seeds, mother tree size had highly significant ($P < 0.01$) effects on germination rate, mean germination time, germination index and vigor index, while salt stress had no significant ($P > 0.05$) effects on germination rate, mean germination time, germination index and vigor index (Table 1). The performance of mother tree size in germination rate, mean germination time, germination index and vigor index was I > II > III.

The differences in germination rate ($F=425.29$, $P<0.01$), mean germination time ($F=8.34$, $P<0.01$), germination index ($F=973.43$, $P<0.01$), and vigor index ($F=79.59$, $P<0.01$) between the two seasons of flowering seeds were highly significant. Summer flowering seeds had high germination percentage, mean germination time, germination index and vigor index compared to spring flowering seeds.

3.2. Effects of Mother Tree Size and Salt Stress on Early Seedling Growth of T. Austromongolica

For spring-flowering seeds, the effect of parent tree diameter class on root length, seedling height, and seedling biomass was not significant ($P > 0.05$), and the effect of salt stress on root length, seedling height, and seedling weight was highly significant ($P < 0.01$) (Table 1). Root length and seedling height decreased gradually with the increase of salt stress, and salt concentration below 9‰ was significantly higher than that of 11-15‰ treatment ($P < 0.05$). Seedling biomass CK was significantly greater than other salt concentrations ($P < 0.05$), 9‰ was greater than 13‰ and 15% ($P < 0.05$), and the differences among other salt concentrations were not significant (P > 0.05).

For summer-flowering seeds, parent tree diameter class had a highly significant effect on root length and a non-significant effect on seedling height and seedling weight; salt stress had a highly significant effect on root length, a significant effect on seedling height, and a non-significant effect on seedling weight (Table 1). Between different diameter classes, the difference in root length between diameter class I and III was not significant (P>0.05) and was highly significant lower than that of diameter class II (P<0.01). Root length and seedling height increased and then decreased with increasing salt concentration, and were highest (P < 0.05) at 3‰.

The differences in root length (F=72.34, P<0.01) and seedling weight (F=55.15, P<0.01) were highly significant between the two seasons of flowering seeds, but not in seedling height (F=1.36, P>0.05). Summer flowering seeds had high root length and seedling weight compared to spring flowering seeds.

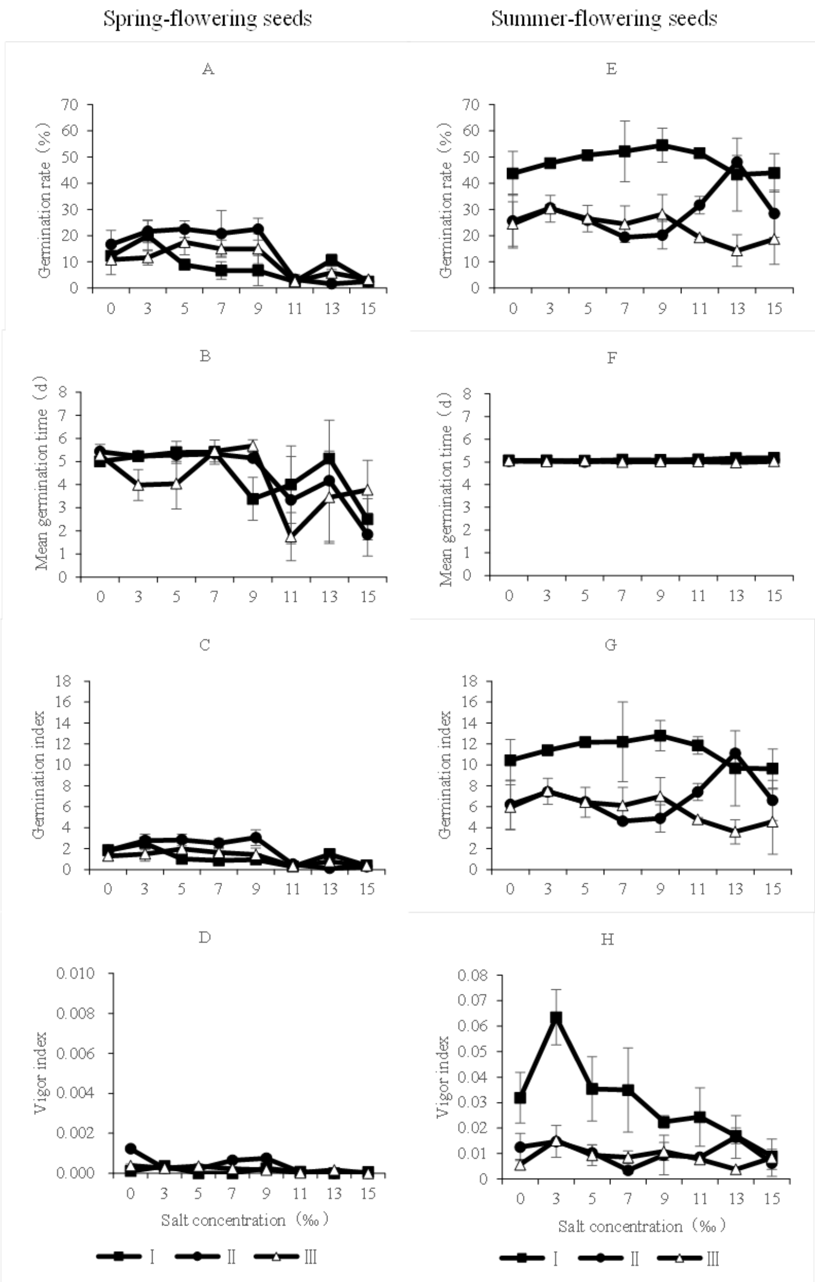

Figure 1. *Effect of parent tree size and salt stress on seed germination of T. austromongolica in two flowering seasons.*

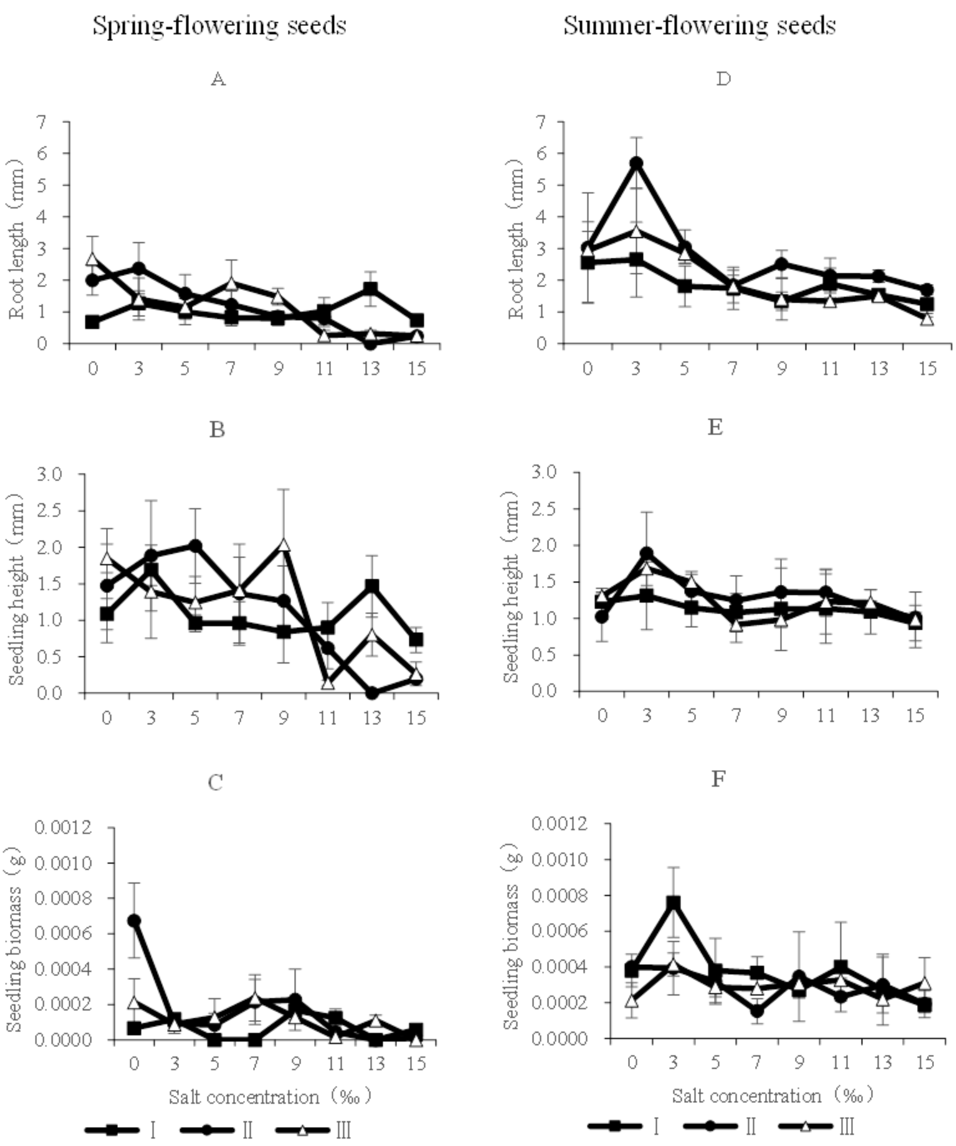

Figure 2. Effects of mother tree size and salt stress on flowering seedlings of T. austromongolica in two seasons.

Table 1. Two-factor ANOVA analysis of seed germination of T. austromongolica by mother tree size and salt stress.

Florescence	Indexes	Mother tree size		Salt stress		Mother tree size × salt stress	
		F-value	P-value	F-value	P-value	F-value	P-value
Spring-flowering seeds	Germination rate	4.91	<0.05	9.44	<0.01	1.92	<0.05
	Mean germination time	0.24	0.79	2.68	<0.05	0.67	0.80
	Germination index	5.00	<0.05	9.03	<0.01	2.22	<0.05
	Vogor index	4.61	<0.05	2.79	<0.05	1.61	<0.05
	Root length	0.33	0.72	3.80	<0.01	2.23	<0.05
	Seedling height	0.06	0.95	4.39	<0.01	1.67	0.09
	Seedling biomass	2.77	0.07	3.82	<0.01	2.24	<0.05
Summer-flowering seeds	Germination rate	79.35	<0.01	0.71	0.67	3.29	<0.01
	Mean germination time	12.02	<0.01	1.22	0.31	1.15	0.34

Florescence	Indexes	Mother tree size		Salt stress		Mother tree size × salt stress	
		F-value	P-value	F-value	P-value	F-value	P-value
	Germination index	65.87	<0.01	0.92	0.50	3.00	<0.01
	Vogor index	11.72	<0.01	1.47	0.20	0.85	0.61
	Root length	8.87	<0.01	10.777	<0.01	1.34	0.22
	Seedling height	1.46	0.24	3.04	<0.05	0.63	0.83
	Seedling biomass	1.65	0.20	2.10	0.06	0.80	0.67

4. Discussion

4.1. The Effect of Parent Tree Diameter Level on Seed Germination in Two Flowering Seasons of Salt-stressed T. Austromongolica

Seed germination is a key stage of plant natural regeneration and can directly reflect the seed quality. The difference in the diameter level of the mother tree affects seed quality and thus seed germination. In this paper, we found that the germination rate, germination index, and vigor index of the spring flowering seeds of T. austromongolica were II>III>I, and II was the largest in the diameter class. Studies on Tetracentron sinense [12], Scaphium wallichii [11], and Camellia rosthorniana [20] also found that seed germination was strongest in the middle of the radial class of the mother tree, which was thought to be related to the increased reproductive allocation at that stage. In a study of lateral cypress seeds of different parent tree sizes, It was found that the germination rate and germination index of Platycladus orientalis seeds increased with the increase of parent tree size [21]. It was shown similar results for Pinus thunbergii seeds, which were related to the increase of nutrient accumulation in the parent tree [22]. For the spring flowering seeds of T. austromongolica, the germination rate, germination index, and vigor index were I>II>III, and the highest quality of seeds was found in the diameter class I. For herbaceous plants Ligularia virgaurea [23], and Gentianopsis paludosa [24], it was found that the reproductive allocation decreased with the increase of individual size, but it was obviously affected by the habitat. Therefore, the variation of flowering seeds with parent tree size in both seasons of T. austromongolica may be the result of the joint influence of its own nutrient accumulation and habitat.

In this study, we found that the germination rate and germination index of T. austromongolica seeds in the spring flowering season at NaCl concentrations of 3‰-9‰ were higher than those of the control and other salt concentration treatments, and the seed germination in the summer flowering season was not affected by the salt concentration, which indicated that its seeds had a strong salt tolerance ability. In terms of salt tolerance, the summer flowering seeds of T. austromongolica were stronger than those of spring flowering, which was consistent with the results of a study on the spring and summer flowering seeds of Tamarix arceuthoides [25]. However, the study on the seeds of Tamarix ramosissima in two seasons found that the seeds in spring flowering stage were more salt-tolerant [26]. In this paper, we found that the average germination time of spring-flowering seeds of T. austromongolica was lower than that of summer-flowering seeds, and the germination rate was high, which was consistent with the previous findings [25]. However, it was found that spring flowering seeds germinated slower than summer flowering seeds in his study on the germination of Tamarix arceuthoides and Tamarix ramosissima under salt treatment [27]. T. austromongolica spring flowering seed formation in the Yellow River Delta region at the time of low rainfall and high soil salinity, while the summer flowering seed formation into the rainy season, the habitat improved significantly, so the seed quality is obviously affected by environmental factors.

4.2. Effect of Mother Tree Diameter Class on Early Growth of Seedlings of Offspring

In this paper, it was found that there was no significant effect of mother tree diameter class on root length, seedling height and seedling weight of spring flowering seeds, and there was also no significant effect on seedling height and seedling weight of summer flowering seeds, but the effect on root length was highly significant. In contrast, a study on Pinus thunbergii seedlings found that both seedling biomass and growth rate increased with increasing diameter class [22], and a similar finding was found for Platycladus orientalis seedlings [21]. This suggests a complex effect of parent tree size on offspring seedlings. With increasing salt concentration, seedling height and root length of both spring-flowering seed and summer-flowering seed progeny seedlings were significantly suppressed. Thus, salt stress plays a major role in seedling growth.

The level of root development determines the growth and development of plants [28]. In this paper, it was found that summer flowering seeds under salt stress had higher root length than spring flowering seeds progeny seedlings. Studies on Morus alba L. found that the degree of root development under salt stress was related to its salt resistance [29]. Therefore, summer flowering seedling progeny were more salt-adapted than spring ones. High seedling growth and seedling size favor seedlings to obtain more resources and improve competitiveness with surrounding plants [22, 30]. The biomass of seedling offspring of T. austromongolica was higher in the summer flowering season than in the spring, suggesting a greater adaptive capacity.

5. Conclusion

In this paper, it was found that *T. austromongolica* spring flowering seeds were the most salt-tolerant in the intermediate diameter class II of the mother tree; while the summer flowering seeds' salt tolerance decreased with the increase of the diameter class of the mother tree. For seedling growth, mother tree size had no significant effect. Therefore, mother tree size was an important factor affecting seed germination, but was influenced by flowering stage. Salt stress had a significant effect on seed germination during flowering in spring, but not in summer; it had a significant effect on seedling size, and growth was first promoted and then inhibited with increasing salt concentration. Therefore, the effect of soil salinity on seed germination was related to flowering stage, but was an important determinant of seedling growth. Compared with spring-flowering seeds, summer-flowering seeds had high salt tolerance and seedling size with greater salt tolerance. Therefore, the mother tree size and flowering period together determine the salt tolerance ability of *T. austromongolica* seeds, and the regulation of seed salt tolerance is an important strategy for the natural regeneration of its population adapted to the Yellow River Delta region, which is important for predicting the evolution and conservation of this population.

Abbreviations

DBH	Diameter at Breast Height
ANOVA	Analysis of Variance
GR	Germination Rat
MGT	Mean Germination Time
GI	Germination Index
VI	Vigor Index
LSD	Least Significant Difference

Author Contributions

The Manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript.

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Conflicts of Interest

The authors declare that they have no competing interests.

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