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Research Article

Storms on the Western Black Sea shore: an opportunity for long-distance dispersal of coastal dunes psammophytes?

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Abstract

Coastal areas are among the planet's most dynamic yet vulnerable environments, the shape of sand dunes being typically modeled by two sets of forces: the relatively constant action of wind and waves and the disturbances caused by infrequent but powerful storms. These meteorological events affect the plant communities, but can also play an important role in the long-distance dispersal of some psammophytes. Plants can be exposed, uprooted, broke apart and spread by wind and waves. Then, depending on the resilience of the plant to seawater temperature, the duration of immersion, the type of fragment transported and the suitability of the new habitat, new population can be established. This study aimed to assess the resilience to seawater immersion and the ability of long-distance dispersal of three psammophytes: *Convolvulus persicus*, *Alyssum borzaeanum*, and *Silene thymifolia*. The experiments tested the viability of vegetative fragments and the reproductive capacity of seeds under different exposure periods (5, 10, and 15 days) and temperature conditions: 4 °C (average surface seawater temperature during winter and early spring when storms on the Black Sea Coast occur), 13 °C (average surface seawater temperature), and 23 °C (average summer surface seawater temperature). The increase of both seawater temperature and the exposure time had a significant negative influence on the viability of the vegetative fragments and on germination capacity. After exposure to seawater, *Convolvulus persicus*, and *Silene thymifolia* are able to establish new populations through vegetative fragments or seeds, whilst in case of *Alyssum borzaeanum*, no vegetative regeneration was observed.

Keywords: coastal areas; reproductive capacity; seawater immersion; vegetative propagation; vulnerability assessment

Introduction

Coastal dunes are ecosystems of high environmental heterogeneity that support diverse and dynamic plant communities with various functional adaptations (Acosta *et al*., 2009; Miller *et al*., 2010; Álvarez-Molina *et al*., 2012; Agır *et al*., 2016; Strat and Holobiuc, 2018). Dune formation is typically influenced by two sets of forces: the relatively constant action of wind and waves and the disturbances caused by infrequent but powerful

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storms (Miller *et al.*, 2010). Due to these factors, the coastal zones are considered to be among the planet's most dynamic yet vulnerable environments (Strat, 2016; Du and Hesp, 2020). These ecosystems globally encounter numerous challenges, increasing their vulnerability to more frequent and intense storms (Cordazzo and Davy, 1999; Sigren *et al.*, 2014; Vergiev, 2018). While wave and wind action generally act to gradually build foredunes into regular dunes, storms, accompanied by heavy rainfall and overwash, lead to erosion, sediment deposition over interdune regions and partial or total destruction of vegetation (Nicholls *et al.*, 2007; Miller *et al.*, 2010; Bernardino *et al.*, 2023). In Romania, these meteorological events have caused total or partial losses of some marine and coastal habitats and substantial damage to a large number of the coastline protected areas (Anton and Rusu, 2019). In the context of global climate change, it is expected that the intensity and frequency of strong storms will increase and with it the vulnerability of coastal areas (Temmerman *et al.*, 2013; Hoggart *et al.*, 2014; Rusu, 2018).

Certain rare and endangered psammophilous species rely heavily on these areas for survival (Strat and Holobiuc, 2018). Apart from providing habitat, coastal dunes also deliver several ecosystem services such as protection against storm waves, storm surges and high tides, acting as a buffer for low-lying inland (Vergiev, 2017a; Vergiev *et al.*, 2019). Plant communities are believed to play a crucial part in these processes by absorbing storm wave energy, supporting and strengthening the dune sediment using root and rhizome systems, thus preventing erosion (Sigren *et al.*, 2014; Vergiev, 2022). Whilst psammophytes are able to cope with some degree of exposure to salinity from seawater, they are vulnerable to the impact of flooding (Vergiev, 2017a).

Therefore, the extreme and unusual storms give rise to an increasing vulnerability for coastal areas and the plant communities related to this type of habitat. On the other hand, these meteorological events, whether extreme or not, can also play an important role in the long-distance dispersal of some psammophytes. This can be achieved through the action of waves and wind which can expose, uproot, break apart into pieces and spread plant fragments, whole plants or seeds through sea waves from one coastal area to another. Long-term flooding may also initiate the decomposition processes of plant fragments (Vergiev *et al.*, 2013). From there, depending on the resilience of the plant to seawater immersion, the time of immersion, the temperature of the water, the type of fragment transported and the suitability of the new habitat, new beaches can be colonized.

Apart from seed production, many herbaceous perennials reproduce asexually by means of vegetative organs (Fenner and Thompson, 2005), also known as clonal growth, that results in the production of vegetative descendants (ramets) that are genetically identical to the parent but with the potential to become independent of the mother organism (Barrett and Eckert, 1990; Klimeš *et al.*, 1997; Vallejo-Marín *et al.*, 2010). The majority of plants combine these two modes of reproduction, and trade-offs between these types are predicted to regulate overall fitness (Van Drunen and Dorken, 2012). Perennial dune plants primarily depend on special reproductive strategies; when it comes to the establishment of plant species on active sand dunes, vegetative propagation is thought to be more significant than sexual reproduction (Li *et al.*, 2005; Yan *et al.*, 2007). In the case of coastal dunes, water currents are the ones involved in dispersing the propagules of numerous plant species (Maun, 2009).

This study aimed to assess the resilience to seawater and the ability of long-distance propagation by seawater of three psammophytes: *Convolvulus persicus* L., *Alyssum borzaeanum* Nyár., and *Silene thymifolia* Sibth. et Sm. We investigated the viability of vegetative buds and seed reproduction capacity after various immersion periods and under different temperature conditions. Thus, three temperatures were selected to indicate 1) average surface seawater temperature during winter, when more wave storms develop in the Western Black Sea (Bernardino *et al.*, 2021), 2) average surface seawater temperature and 3) average summer surface seawater temperature. The ability of wetland plants to withstand freshwater flooding has been thoroughly investigated. However, despite the concerns regarding the impact of storm events on coastal areas, the response of individual terrestrial coastal plants to seawater flooding is poorly understood and the information is insufficient (White *et al.*, 2014), making the prediction of ecological effects of anthropogenic changes in storms surge events difficult (Hanley *et al.*, 2013). Understanding the response of these three rare and threatened psammophilous plant species from the Western coast of the Black Sea to seawater flooding is very important for clarifying their long-distance dispersal mechanisms and reproductive strategies. This knowledge can play an essential role in developing effective management strategies to conserve these species and their habitats.

Materials and Methods

Study species

Convolvulus persicus L.

C. persicus is a pioneer perennial hemicryptophyte plant of sandy shores, with elongated woody rhizomes that are deeply buried in sand and spread laterally, helping the plant grow through the sand (Strat and Holobiuc, 2018). Stems and leaves are densely wooly-tomentose, stems of 20-50 cm and elliptic-oblong and short petiolate leaves (Grințescu, 1960; Ciocârlan, 2009; Dihoru and Negrean, 2009). The flowers are single, white, bisexual, funnel-shaped (Strat and Holobiuc, 2018). Fruits are ovoid capsules (10 mm in diameter) with two–four dormant seeds (Grințescu, 1960; Strat and Holobiuc, 2018). It is considerd a clonal species, where vegetative reproduction predominates (Strat and Holobiuc, 2018). *C. persicus* is considered as a rare and endangered species in the National Red List of vascular plants (Oltean *et al*., 1994) and critically endangered in the Red Book of vascular plants in Romania (Dihoru and Negrean, 2009).

Alyssum borzaeanum Nyár.

A. borzaeanum is a perennial chamaephyte plant with a woody root, numerous erect stems of 10-25 (30) cm, at the base with many sterile shoots, covered with a white dense layer of stellate hairs. Flowers are little and yellow, disposed in corymbs and fruits are silicle (3-3.6 mm in length) with little stellate hairs, each loculus containing one-two not winged seeds (Nyárády, 1955). *A. borzaeanum* is considered as a rare species in the National Red List of vascular plants (Oltean *et al*., 1994) and critically endangered in the Red Book of vascular plants in Romania (Dihoru and Negrean, 2009). Additionally, it is listed as Data Deficient in the European Red List of Vascular Plants.

Silene thymifolia Sibth. et Sm.

S. thymifolia is a perennial hemicryptophyte plant with branched, woody stock and numerous scabrid-puberulent stems that can reach up to 20 cm (Jianu and Făgăraș, 2015). Leaves are little, ovate and pubescent and flowers are white, bisexual, with bifid petals, disposed in dichasial inflorescence. Fruits are capsules (8-9 mm) and contain reniform seeds (Gușuleac, 1953; Dihoru and Negrean, 2009; Jianu *et al*., 2021). *S. thymifolia* is considered as a rare species in the National Red List of vascular plants (Oltean *et al*., 1994) and vulnerable in the Red Book of vascular plants in Romania (Dihoru and Negrean, 2009), although the species is extremely rare in Romania, the only confirmed location currently being the one from Agigea Natural Reserve (Făgăraș *et al*., 2016). The current conservation status of *S. thymifolia* is totally inadequate and unfavourable in Romania (Jianu *et al*., 2021).

Plant material

Plant material from the three analyzed psammophytes, *C. persicus, A. borzaeanum*, and *S. thymifolia* were collected from Romania and Bulgaria. Whole plants, rhizomes, and seeds of *C. persicus*, along with sand samples, were obtained from Perișor Beach in Romania in January 2023. The plant material for *A. borzaeanum* and *S. thymifolia* was collected from Durankulak North Beach in Bulgaria in November 2022. The plant material was transported to the laboratory, where it underwent cutting and division into segments of similar length. Subsequently, all tests were conducted at the Plant Physiology Laboratory of „Alexandru Ioan Cuza" University of Iași.

Vegetative regeneration experiment

For this experiment, we adapted the methodology proposed by Vergiev (2018, 2021) to assess the vulnerability of psammophilous species, including *S. thymifolia*, to flooding along the Bulgarian Black Sea Coast caused by unusual storms.

All plant fragments from analyzed species underwent submersion in containers filled with seawater, maintained at three constant controlled temperatures (4 °C, 13 °C, and 23 °C) for 5, 10, or 15 days, depending on the treatment group, within a growth chamber. These temperature regimes simulate natural events, such as average seawater temperatures during various seasons, allowing us to investigate the relationship between temperature and species' resilience. The first temperature treatment at 4 °C corresponds to the average surface seawater temperature during winter and early spring, coinciding with storms on the Black Sea Coast. The second treatment at 13 °C represents the average surface seawater temperature, while the third treatment, at 23 °C, corresponds to the average summer surface seawater temperature (Vergiev, 2021).

The water from the containers was changed twice per day in order to prevent water putrefactive processes. Every fifth day, two-four fragments from each species per temperature treatment group (4 °C, 13 °C, and 23 °C) were removed from the seawater containers and planted in plastic pots with sand collected from their natural habitat. Simultaneously, plant fragments were inspected every fifth day for visible morphological changes and each change was recorded. After transplantation into sand, the fragments were watered daily and kept at 20 °C for one month before assessing the number of viable buds capable of generating new plants.

The initiation of decomposition for plant fragments was defined as the point where observable decay exceeded 15% of the entire vegetative organ surface. Complete decomposition was recorded when visible decay encompassed more than 50% of an organ's surface (Vergiev, 2021).

Germination experiment

To assess the influence of different temperature conditions on seed reproductive capacity, seawater immersion simulations were conducted with the three temperature variations considered for the previous experiments (4 °C, 13 °C, and 23 °C) for 15 days prior to the germination test. Following immersion, the seeds were distributed in Petri dishes lined with filter paper moistened with distilled water. The dishes were placed in a growth chamber at a constant temperature of 24 °C, corresponding to a 12/12 hours photoperiod, for 12 days. To maintain a constant humidity level, the Petri dishes were watered regularly with distilled water throughout this period. Daily recordings were made to monitor the progress of the experiment. The germination rate was calculated as the final percentage of germinated seeds.

Statistical analysis

Our investigations on plant responses to seawater conditions involved a series of statistical analyses. Non-parametric tests and regression analyses were employed to comprehensively assess various aspects of the data, identifying key factors influencing bud viability, decomposition, and germination capacity under different seawater conditions. The Wilcoxon signed-rank test was first applied to compare the initial number of buds with the number of buds after seawater immersion for *C. persicus* and *S. thymifolia*. Then, it was used to compare the number of buds after immersion with the number of final viable buds, for each species. The Kendall rank correlation test was used to examine the relationship between seawater temperature and the number of viable buds after immersion. Differences in viable buds across different seawater immersion duration (5 days, 10 days, and 15 days) were assessed using the Kruskal-Wallis test. Additionally, the correlation between seawater temperature and the degree of plant fragment decomposition was examined using the Kendall rank correlation test. The Kruskal-Wallis test was also applied to identify differences in vegetative fragments' decomposition across experimental conditions.

To understand the joint influence of temperature and immersion duration on viable buds, a simple Generalized Linear Model (GLM) with a Gaussian distribution was employed. Similarly, the impact of seawater temperature on germination rate was evaluated using another simple GLM with a Gaussian distribution. All of the statistical analyses were performed in R Software. Mean bud viability (MBV) for each treatment group was recorded as the percentage of vegetative buds that produced new vegetative shoots and roots after immersion in seawater (Vergiev, 2018).

Results

For *C. persicus*, the highest Mean Bud Viability (MBV) values were consistently observed at 13 °C across all immersion periods. Similarly, for *S. thymifolia*, the highest MBV values were also recorded at 13 °C for two treatment groups (10 and 15 days), while the highest value after 5 days of immersion was recorded at 4 °C. Throughout the seawater submersion, plant fragments from both species exhibited the highest regenerative capacity (highest MBV values) at the temperature of 13 °C. In both cases, MBV percentages appear to be notably decreasing as temperatures increase from 13 °C to 23 °C (Table 1).

Table 1. Mean bud viability (MBV) and fragments' decomposition rate across the analyzed species, temperature and immersion duration

Species	Temperature (°C)	Analysed parameters	Time of immersion		
			5 days	10 days	15 days
Convolvulus persicus	4	Mean bud viability (%)	15.15	0	0
		Mean percentage of fragments' decomposition rate (%)	2	5	10
	13	Mean bud viability (%)	26.08	10	6.25
		Mean percentage of fragments' decomposition rate (%)	5	10	20
	23	Mean bud viability (%)	25	0	0
		Mean percentage of fragments' decomposition rate (%)	10	20	60
Silene thymifolia	4	Mean bud viability (%)	100	21.21	0
		Mean percentage of fragments' decomposition rate (%)	5	10	25
	13	Mean bud viability (%)	14.28	42.85	6.66
		Mean percentage of fragments' decomposition rate (%)	8	20	45
	23	Mean bud viability (%)	0	0	0
		Mean percentage of fragments' decomposition rate (%)	10	35	60
Alyssum borzaeanum	4	Mean bud viability (%)	0	0	0
		Mean percentage of fragments' decomposition rate (%)	13	23	>70
	13	Mean bud viability (%)	0	0	0
		Mean percentage of fragments' decomposition rate (%)	24	57	>90
	23	Mean bud viability (%)	0	0	0
		Mean percentage of fragments' decomposition rate (%)	38	>90	>90

In all temperature treatment groups (4 °C, 13 °C, 23 °C), we observed a gradual decline in MBV values with the increase of immersion period. The only exception is represented by *S. thymifolia* fragments immersed at 13 °C, reaching maximum MBV after 10 days and decreasing in the following 5 days of exposure to seawater (day 15). *C. persicus* exhibited the highest MBV values, for all temperature conditions, after 5 days of submergence (Table 1).

In the case of *A. borzaeanum*, immersion in seawater doesn't seem to have stimulated, nor inhibited the viability of the buds, as the collected samples did not show any visible buds at the moment of counting. The highest values of fragments' decomposition rate were observed for plant fragments exposed to higher temperatures for all species. Out of all species, *A. borzaeanum* fragments recorded the mean percentage of total decomposition earliest, specifically on the 10^{th} day at 13 °C and 23 °C, and on the 15^{th} day at 4 °C. Both *C. persicus* and *S. thymifolia* performed better at lower temperatures; the mean percentage of complete decomposition was noted on the 15^{th} day at 23 °C (Table 1). The Wilcoxon signed-rank test revealed no significant difference between the initial number of buds and number of buds after seawater immersion for *C. persicus* ($V = 3$, $P = 0.3711$) and *S. thymifolia* ($V = 56.5$, $P = 0.1798$). The test was not assessed for *A. borzaeanum* since the mean bud viability (%) was 0 in all scenarios of duration of seawater immersion. Comparing the number of buds after immersion with the number of final viable buds showed significant results for both *C. persicus* ($V = 171$, $P = 0.0002$) and *S. thymifolia* ($V = 45$, $P = 0.0090$). These findings indicate a significant difference between the number of buds after immersion and the eventual viable buds for both analysed species. The Kendall rank correlation test showed a significant negative correlation ($z = -1.9824$, $P = 0.0474$, $tau = -0.221$) between the number of viable buds after immersion and temperature. This indicate that as seawater temperature increases, the number of viable buds decreases. The Kruskal-Wallis test examining the difference in viable buds after different seawater immersion durations (5 days, 10 days, and 15 days), showed a significant difference ($P = 0.0118$) between treatment groups. Subsequent pairwise comparison indicated a significant decline in the number of viable buds after 15 days of seawater immersion, compared to 5 days (Table 2).

Table 2. Pairwise comparison of viable buds after seawater immersion duration

Treatment groups	5 days of immersion	10 days of immersion
10 days of immersion	0.067	-
15 days of immersion	**0.025**	0.561

*Notes: The significant values are in bold (P value < 0.05)

The Kendall rank correlation test investigating the relationship between seawater temperature and plant fragment decomposition showed a significant influence ($z = 5.4598$, P value < 0.05, $tau = 0.3607$), indicating a correlation between the degree of plants fragments' decomposition and temperature. As anticipated, higher water temperatures accelerated decomposition processes (Figure 1). The Kruskal-Wallis Test assessed the statistical differences between groups and demonstrated significant variation in fragment decomposition across the 3 experimental conditions (4 °C, 13 °C, 23 °C).

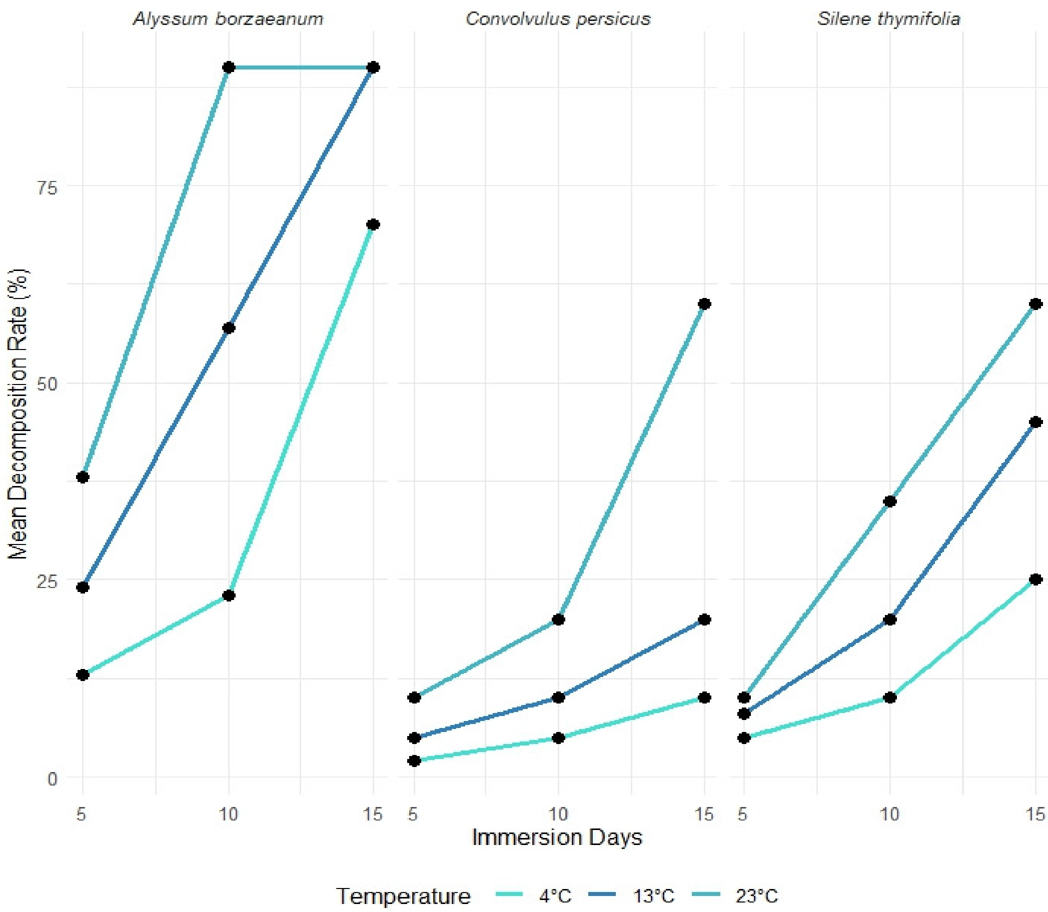

Figure 1. Mean percentage of fragments' decomposition rate across different seawater immersion days and temperature treatments for the three analysed psammophytes

Although previous tests indicated a significant correlation between seawater temperature and the number of viable buds, also revealing significant differences in the number of viable buds according to immersion time, we further tested the combined influence of temperature and immersion period on the number of viable buds. The results showed that both temperature and immersion duration significantly negatively influence the number of viable buds, which means that the number of viable buds decreases with the increase of temperature and the number of days of seawater immersion (Figure 2). The observed negative influence was recorded only for two experimental groups (10 days and 15 immersion days) (Table 3).

Table 3. Influence of temperature and seawater immersion period on number of viable buds for *C. persicus* and *S. thymifolia*

Variables	Number of viable buds	
	t value	*P* value
Temperature	-2.272	**0.0265**
10 days of immersion	-2.455	**0.01691**
15 days of immersion	-3.204	**0.0021**

*Notes: The significant values are in bold (*P* value < 0.05)

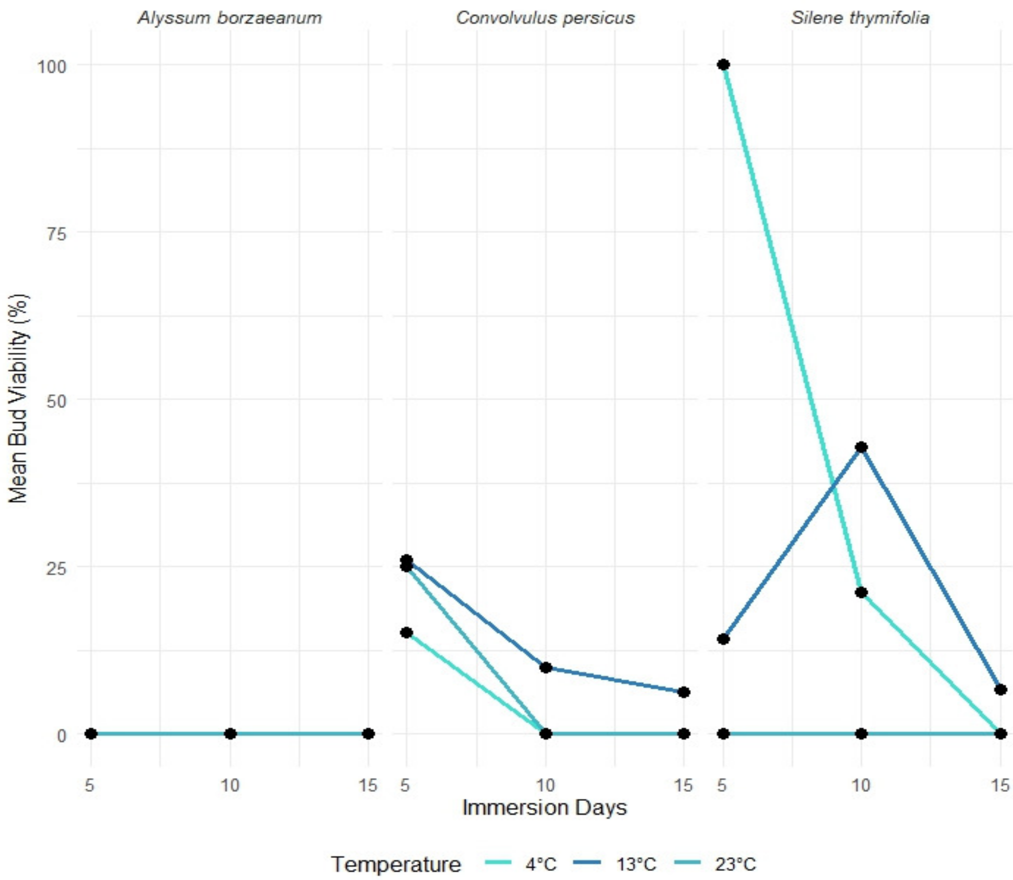

Figure 2. Mean bud viability (MBV) (%) over immersion days for the three analysed psammophytes under different seawater temperatures (4 °C, 13 °C, 23 °C)

In the germination experiment, we assessed the potential impact of seawater temperature on germination rates. The results indicated that, even though the seawater temperature did not exhibit a statistically significant effect on germination (t value = -1.679, P = 0.1370), it did have a negative influence, suggesting a decrease in germination percentage with the increase in seawater temperature.

A. borzaeanum showed the earliest germination, occurring on the first day of the experiment at 13 °C and 23 °C, and on the second day at 4 °C. *C. persicus* seeds-initiated germination on the 7th day, only under the 4 °C experimental condition. No seeds were recorded germinating at 13 °C and 23 °C for this species. For *S. thymifolia*, germination initiated on the fourth day across all temperature treatments. Out of all species, the highest germination rate was recorded for *A. borzaeanum*, for all seawater temperatures (

Table 4).

Table 4. Germination rate of the analysed plant species under different temperature treatments

Species	Temperature (°C)	Germination rate (%)
Convolvulus persicus	4	20
	13	0
	23	0
Silene thymifolia	4	19.44
	13	25
	23	11.11
Alyssum borzaeanum	4	43.33
	13	43.33
	23	13.33

Discussion

Although the differences between the overall initial number of buds pre- and post-immersion were not significant, there were cases in which the number of buds increased (of *S. thymifolia* fragments) after exposure to seawater. This may be due to the fact that the buds were not visible at the moment of initial counting and can indicate that seawater immersion does not inhibit the development of buds in the case of *S. thymifolia*. These results do not take into account the period of time the buds spent in the water. The difference however becomes apparent between the number of buds after submergence and the final number of buds that have spent one month planted in sand.

The number of viable buds after immersion and seawater temperature were found to be negatively correlated, indicating a reduction in the number of viable buds as seawater temperature rises. Plant fragments from both *C. persicus* and *S. thymifolia* demonstrated their peak regenerative potential (as indicated by the highest MBV values) at the temperature of 13 °C. We can say that both species retained bud viability for longer at cooler temperatures corresponding to winter and early spring months (4 °C) and average seawater temperature (13 °C) compared to the temperature of summer months (23 °C), the number of viable buds decreasing with the increase of temperature. These findings are in contrast with those of Vergiev (2017a), Vergiev (2017b), Vergiev (2018), Vergiev (2019a), Vergiev (2019b) and Vergiev (2022), in which different temperatures influenced rhizome viability of species that grow on sand (*Carex colchica* J.Gay, *Galilea mucronata* Parl., *Leymus racemosus* (Lam.) Tzvelev subsp. *sabulosus* (M. Bieb.), *Ammophila arenaria* (L.) Link, *Elymus elongatus* (Host) Runemark, *Thinopyrum ponticum* (Podp.) Barkworth & D.R.Dewey, and *Cynodon dactylon* (L.) Pers) in similar levels and average differences between coolest and warmest temperatures were statistically insignificant. In other words, the studies suggest that the presence of water is a more crucial factor for viability, rather than the temperature of the water itself. However, our results are in agreement with those of the study of *Ammophila arenaria* rhizomes (Konlechner and Hilton, 2009), that withstand longer immersion during winter months because rhizome retained viability for longer in cooler water. In the studies of van Eck *et al*. (2005 and 2006) that focused on the impact of flooding events on grassland plant species, similar seasonal effects were observed. The authors discovered that all species survived longer when they underwent submersion in winter simulated conditions compared to summer conditions.

The duration of seawater exposure for propagules is another important factor affecting the number of viable buds. Significant differences were observed among the immersion period treatment groups. Generally, MBV recorded the highest percentages following 5 days of seawater submergence for both *C. persicus* and *S.*

thymifolia fragments, with a visible decline as the immersion period increased. This is in agreement with the study of *Ammophila arenaria* and *Leymus mollis* (Trin.) Pilg. rhizomes (Aptekar and Rejmánek, 2000), in which there was also a significant interaction between species and submergence duration. Contrary to our results, the duration of immersion had no significant effect in the study of *Galilea mucronata*, *Carex colchica*, *Leymus racemosus*, *Ammophila arenaria*, and *Elymus elongatus* (Vergiev, 2017a; Vergiev, 2017b; Vergiev, 2018; Vergiev, 2019a), *Thinopyrum ponticum* (Vergiev, 2019b) and *Cynodon dactylon* (Vergiev, 2022).

The contrasting situation of *A. borzaeanum* (no buds observed) may be attributed to the timing of sampling, which occurred at the end of November. As observed for other chamaephytes developing on sands, such species have a short vegetative period which may be correlated with a separation between root growth (in autumn) and shoot growth (in spring) (Martínez *et al.*, 1998; Palacio and Montserrat-Martí, 2007). Leaf primordia may accumulate during or at the end of winter, then expanding quickly in spring (Montserrat-Martí *et al.*, 2011). Therefore, it may be plausible that the absence of vegetative buds in the collected material could be attributed to the sampling timing not aligning with the period of vegetative bud development.

The results also indicated a correlation between the degree of plants fragments decomposition and temperature and significant differences between the degree of fragment decomposition between the three temperature conditions (4 °C, 13 °C, 23 °C). This is in contrast with studies of Vergiev (2017a), Vergiev (2017b), Vergiev (2018), Vergiev (2019a), Vergiev (2019b), Vergiev (2022), in which a significant relationship between resilience and water temperature was not found. As we expected, decaying processes were enhanced by higher water temperatures and the highest decomposition percentages were recorded for plants submerged at the highest temperatures for all species. These results are consistent with some previous studies, in which it was found that the parameter concerning the decomposition of the leaves of *Crambe maritima* L., *Eryngium maritimum* L., and *Xanthium strumarium* L. (Vergiev *et al*., 2019) and *Centaurea arenaria* M. Bieb. ex Willd., *Crambe tataria* Sebeok, *Aurinia uechtritziana* (Bornm.) Cullen & Dudley, *Stachys maritima* Gouan, and *Silene thymifolia* (Vergiev, 2021) was accelerated by higher water temperatures. After transplantation on sand from the natural habitat, the fragments were permitted to grow for one month before checking for regeneration signs and establishing bud viability. Up until the end of the experiment, the newly developed stems and roots maintained their viability.

The direction and speed of Black Sea currents play a major role in determining the region's size and extent that is susceptible to the studied species colonization. After 5 days of exposure to seawater, both *S. thymifolia* and *C. persicus* retained relatively high MBV values. *S. thymifolia* maintained a higher MBV when exposed to cooler temperatures (≤ 42.85%) after 10 days of submergence than *C. persicus* (≤ 10%). Both species maintained the capacity to regenerate from buds following 15 days of immersion at average seawater temperature (13 °C), even though the MBV appears to be low (< 6.7%). Assuming near-shore current speeds of 0.2 m s^{-1} (Miladinova et al., 2020) and transportation in a constant direction, we can estimate that both *C. persicus* and *S. thymifolia* fragments may possibly be transported 17.28 km day^{-1} and up to 259.2 km within 15 days while still remaining viable. Even if this is a pure basic calculation, the possibility of dispersal via seawater is supported.

In regard to the germination capacity of the psammophilous species, the results revealed that seawater temperature negatively influenced the germination process, resulting in a decline in germination rate as seawater temperature rose. When the seeds were immersed in seawater, it was observed that the majority of them floated. This is an extremely important factor to take into consideration, since buoyancy is necessary for seed dispersal by currents (Guja *et al*., 2010). Although seeds did float, successful dispersal requires that they remain viable after exposure to seawater. Similar to the vegetative regeneration responses, seeds subjected to seawater temperatures that simulate average seawater temperature (13 °C) and that of winter and early spring months (4 °C) possess a higher germination rate, as opposed to those submerged at the temperature of summer months (23 °C).

Two out of the three studied plants seem to be guided by a compensatory rule, where vegetative reproduction occurs in response to stressful environmental factors, replacing, at least temporarily, seed reproduction (as seen in the cases of *C. persicus* and *S. thymifolia*). From our field observations, *C. persicus* seems to rely primarily on vegetative growth as its main survival strategy in nature. This is clearly observed for the populations along the Black Sea shore (including those from the Danube Delta, on dunes from Cardon – Sfiștofca area), even though the plant produces fruits with viable seeds. In Agigea, the population of *C. persicus* appears to face an entirely different issue, where sexual reproduction has been entirely replaced by vegetative growth, with the plant no longer producing any seeds at all. Even if the installation of new individuals of *S. thymifolia* as a result of vegetative propagation was not yet confirmed by our *in-situ* observations, this process may be possible, as shown by the viability of buds during the experiments and of the newly formed shoots after the transplantation. *A. borzaeanum* does not appear to exhibit compensatory potential, and this may be due to its high reproductive capacity through seeds, even after immersion in seawater; this species recorded the highest germination rate values, as observed in this experiment.

The majority of perennial plants present a combination of clonal propagation with sexual reproduction and very seldom clonal plants become entirely asexual. A balance is kept between reproductive modes, and biotic or abiotic factors are assumed to have an impact on this balance (Dorken and Eckert, 2001; Silvertown, 2008). In response to environmental stressors, plants either invest more in reproductive growth, which maintains population diversity, or in vegetative growth, which ensures sufficient population expansion (Chen *et al*., 2023; Ba *et al*., 2024). In certain challenging environments where the process of reproduction through seeds becomes more difficult, clonal integration has been shown to be an efficient method for ramets to endure these environmental stresses and survive sand burial (Yu *et al*., 2004; Herben *et al*., 2015).

Conclusions

The most important factors in the assessment of reproductive strategies of the studied species when subjected to the stress of seawater immersion considered in this experiment are the temperature and the exposure time. Both parameters influenced the buds viability and the resistence of plants to decomposition, while the germination of seeds was influenced only by the water temperature. Dispersal during cooler months coressponding to winter and spring seems to be more favourable than during warmer months corresponding to summer, due to lower water temperature and reduced decomposition of plants.

Our study proved that storms, although stressful events, may be an opportunity for long-distance dispersal of plants from the sand dune sea shore. Some species (such as *C. persicus* and *S. thymifolia*) may be able to use both vegetative and seed propagation, whilst others (*A. borzaeanum*) seem to rely mainly on seed reproduction. Depending on the distance from the shore, some plants can be uprooted, while others can just be flooded. Our observations showed that all the analysed species have some resilience to seawater immersion, being able to withstand flooding and maintaining their viability for some time. It is worth noting that the hemicryptophytes without leaves during winter (*C. persicus*, *S. thymifolia*) proved to be more resistant to the decomposition effect of seawater immersion than the chamaephytes (*A. borzaeanum*) with persistant, hairy leaves. Based on this idea, future research on marine dispersal could be conducted taking into account different field collection periods of vegetative fragments (during spring months); this might extend the explanations of the reproductive strategies of these types of sand dune plant species.

Authors' Contributions

Conceptualization: AS and CCM; Data curation: AS, CCM and AGC; Formal analysis: AS and AGC; Investigation: AS and CCM; Methodology: AS and CCM; Resources: AS and CCM; Supervision CCM and MMZ; Validation: AGC and CCM; Visualization: AGC and MMZ; Writing - original draft: AS, AGC and CCM; Writing - review and editing: AS, AGC and CCM. All authors read and approved the final manuscript.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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