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SUITABLE FLOW VELOCITY FOR GERMINATION AND ESTABLISHMENT OF *SUAEDA SALSA* IN LIAOHE ESTUARY

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Abstract

The germination and establishment of *Suaeda salsa* are different with the varying flow velocitied and seed burial depths. This study focused on the seeds and seedlings of *Suaeda salsa* in the Liaohe Estuary, through outdoor simulation experiments and indoor analysis, the germination rate and seedling growth traits at different burial depths under different flow velocity conditions were determined. The main findings were as follows: Within the burial depth of 0-2cm, and the flow velocity range of 0.059-0.073 m/s was suitable for germination and establishment, at 2-3 cm depth, the suitable flow velocity range was 0.059-0.069 m/s, at 3-4 cm depth, it remained 0.059-0.069 m/s, and at 4-5 cm depth, the suitable flow velocity range extended to 0.059-0.076 m/s. These results provide theoretical support for the conservation and restoration of *Suaeda salsa* in the Liaohe Estuary and the sustainable development of wetlands, holding significant scientific and practical value.

Key words: Suitable flow velocity; Germination; Establishment; *Sueda salsa*; Liaohe Estuary.

Introduction

The *Suaeda salsa* in the Liaohe Estuary, located at the intersection of land and sea, plays an irreplaceable role in protecting the Bohai Sea coastline. It serves as a habitat for multiple endangered species and is renowned globally for its red beach landscape, making it of significant ecological importance to the region (Cao *et al*., 2022). However, in recent years, the area covered by *Suaeda salsa* has drastically decreased, leading to severe degradation of the wetland, which has directly impacted the habitat safety of endangered species (Dong *et al*., 2024). Urgent protection and restoration of the *Suaeda salsa* in the Liaohe Estuary wetland are needed. The most direct manifestation of wetland degradation is the sharp reduction in *Suaeda salsa* coverage, with changes in hydrological conditions being the primary driver of its degradation (Ma *et al*., 2024a). For the protection and restoration of the *Suaeda salsa* wetland ecosystem, the germination status of its seeds, such as germination rate, is essential for the entire growth stage and the development of community (Zhang *et al*., 2024). Therefore, during the protection and restoration of *Suaeda salsa* in the Liaohe Estuary, improving its germination status through changes in hydrological conditions is particularly important for the recovery of the *Suaeda salsa* community.

Current research on *Suaeda salsa* primarily focuses on the physiological and biochemical characteristics of its various organs or tissues, as well as their responses to salt stress (Guo *et al*., 2020; Zhang *et al*., 2021; Guo *et al*., 2022; Adichirattle *et al*., 2024). However, studies on *Suaeda salsa* seeds under salt stress mainly concentrate on germination under single alkali (Na_2CO_3, $NaHCO_3$) or single salt ($NaCl$, Na_2SO_4) stress conditions (Li *et al*., 2022; Qi *et al*., 2023; Sun *et al*., 2023). Hydrological disturbance is one of the common disturbances in wetland ecosystems, and most plant seeds are sensitive to such disturbances (Liang & Shi, 2021). Li *et al*. (Li *et al*., 2010) found that after hydrological disturbance, the germination rate remained unchanged for two species, increased for three species, and decreased for sixteen species among twenty-one herbaceous plant species. This indicates that hydrological disturbance can hinder seed germination for most species. The natural regeneration process of plants includes seed dispersal, seed bank dynamics, seed germination, and seedling establishment (Harden & Scruggs, 2003; Kim *et al*., 2022), with seed germination and early seedling establishment being crucial stages (Jiang *et al*., 2003; Wainwright & Parsons, 2010), Moreover the germination and establishment stages are sensitive periods (Kirkby & Chorley, 1967), during which germination and survival rates are highly susceptible to external environmental disturbances (Ortiz-Martínez *et al*., 2021). In coastal wetland environments, the greatest threat to *Suaeda salsa* seed germination and establishment is flow velocity disturbance (Leip *et al*., 2011). Studying the suitable mechanisms for *Suaeda salsa* germination is crucial for its conservation and restoration. Although some progress has been made in this area, there is still a lack of in-depth research considering the influence of eco-hydrological dynamic factors on *Suaeda salsa* germination. Our hypotheses were: (i) as an annual herbaceous plant, the germination and establishment of *Suaeda salsa* play a decisive role in its community development, and (ii) the flow velocity has a crucial impact on its germination and establishment stages by affecting the retention germination rate of seeds and the height and fresh weight of seedlings.

Material and Method

Test materials

① ***Suaeda salsa* seeds:** The tested seeds were collected from *Suaeda salsa* wetland in Liaohe estuary (121°30′-122°00′E, 40°50′-41°20′N). Soak the retrieved *Suaeda salsa* seeds in a 500mg/L $KMnO_4$ solution for 30 minutes, then select the seeds that have sunk to the bottom of the beaker and rinse them with distilled water. After drying in a cool shade, select the black seeds and store them in refrigeration for the experiment.

② **Soil:** The tested soil was collected from the Liaohe estuary, it was transported to the experimental site for original soil restoration and laid in the experimental field. The soil pH is 7.1~7.2, and the water content is 69.32%~72.97%, reaching the field capacity state.

③ **Water:** The test water was groundwater from the comprehensive experimental base of the Shenyang Agricultural University, Shenyang City, Liaoning Province.

Experiment setting

The simulation experiment was carried out in the tidal flat simulation pool of Shenyang Agricultural University. The tidal flat simulation pool was divided into 25 simulated scour channels using stainless steel plates(Fig. 1). This setup was then used to simulate five different flow velocities and five different seed burial depths, observing changes in the germination of *Suaeda salsa* seeds and the growth of seedlings under these conditions.

Based on field monitoring data from the past five years, the duration and frequency of inundation have been determined, the maximum flow velocity, the minimum flow velocity, the average flow velocity, the average flow velocity during high tide, and the average flow velocity during low tide were used as experimental velocities and named as V_1 = 0.226 m/s, V_2 = 0.156 m/s, V_3 = 0.086 m/s, V_4 = 0.056 m/s, and V_5 = 0.025 m/s in the study. Five seed burial depths for each flow velocity condition were set: H_0=0cm; H_1=1~2cm; H_2=2~3cm; H_3=3~4cm; H_4=4~5cm. Each experiment was repeated 5 times and set the control group (CK).

The selected seeds were evenly sown in the standard quadrats (1m×0.5m) separated by stainless steel. After uniformly sowing 1,000 black seeds in each quadrat, a two-day scouring experiment was conducted with 3.2 hours per day. After seven days, the seedlings and ungerminated seeds were excavated for detection and analysis. In this study, the standard quadrats in each scouring channel were named A_1, A_2, A_3 and A_4.

Data analysis

Data recording, organization, and statistics were conducted using Office, while analysis was performed with SPSS 22.0, including Pearson correlation analysis($P<0.05$) for seed germination and growth traits. Origin 2021 was utilized for figures creation.

Results and Discussion

Effect of flow velocity on the germination rate of *Suaeda salsa* seeds: For *Suaeda salsa*, the ability of seeds to germinate is one of the critical factors affecting the survival, reproduction, and renewal of the community in harsh environments (Guan *et al*., 2011; Czortek & Pielech, 2020). Germination rate is one of the key indicators for assessing seed quality. Therefore, determining the impact of flow velocities on the germination rates of *Suaeda salsa* seeds at various burial depths, measuring the germination rate is essential.

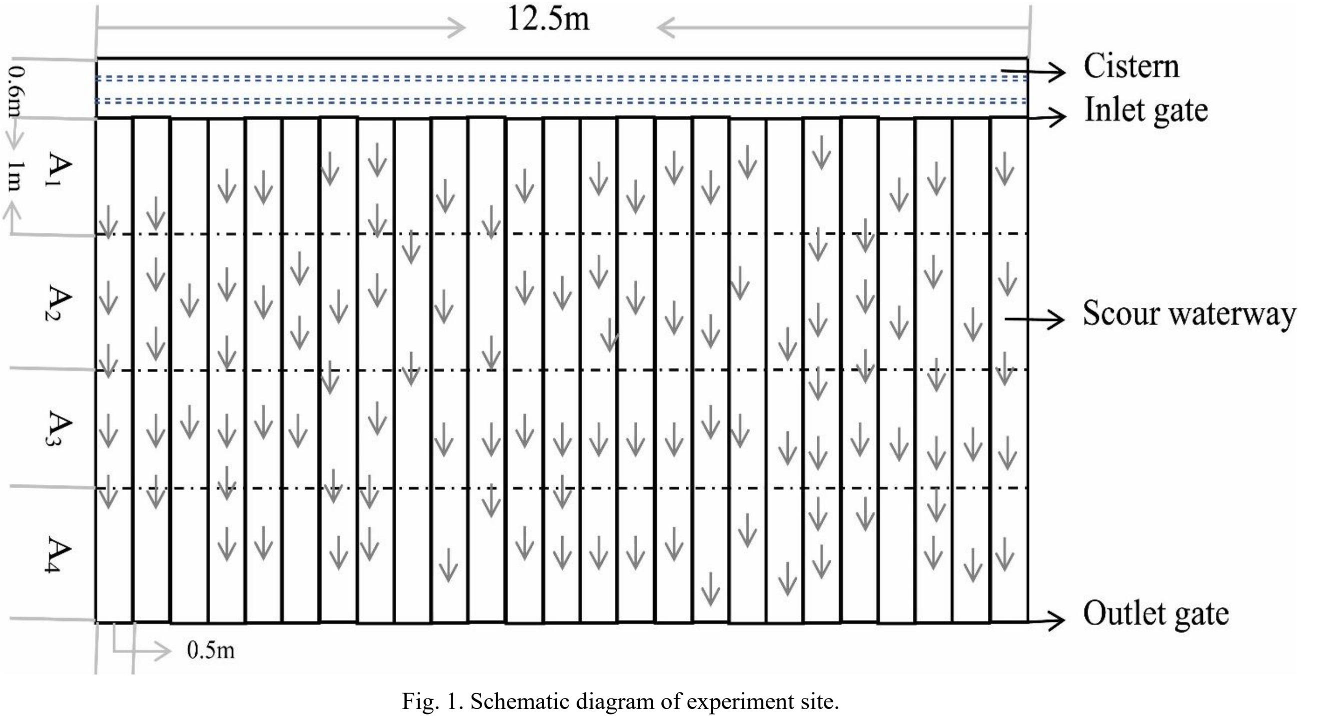

Fig. 1. Schematic diagram of experiment site.

As shown in Fig. 2, at the H_0(0cm) burial depth, the germination rate of *Suaeda salsa* seeds shows an initial increase followed by a decrease as flow velocity reduces. when the flow velocity changes from V_1(0.226m/s) to V_2(0.156m/s), the germination rate increases, and then significantly increases further as the flow velocity changes from V_2(0.156m/s) to V_3(0.086m/s) and V_4(0.056m/s), reaching its maximum germination range, when the flow velocity shifts from V_4(0.056m/s) to V_5(0.026m/s), the germination rate gradually decreases. Compared to the control group, the germination rates under flow velocities from V_1(0.226m/s) to V_5(0.025m/s) are all significantly lower. It is due to the light weight and low density of *Suaeda salsa* seeds, they are easily lost with runoff when entering the scour channel, leading to reduction of seeds available for germination and thus decreasing in germination rate, this is consistent with the research results of Ma *et al*., (Ma *et al*., 2024b). At H_1(1-2cm) burial depth, the trend of changes in the germination rate of *Suaeda salsa* seeds under different flow velocity gradients is largely similar to that at H_0(0cm) burial depth. When the flow velocity gradient changes from V_1(0.226m/s) to V_2(0.156m/s), there is a significant increase in seed germination rate, which differs considerably from that at H_0(0cm) burial depth. This is due to the soil covering effect on the seeds, which reduces the seed loss rate. When the flow velocity gradient reaches V_3(0.086m/s) to V_4(0.056m/s), the seed germination rate reaches its maximum range. The difference in germination rates between the various flow velocity gradients and the static water condition is significantly reduced compared to that at H_0(0cm) burial depth, indicating that seed burial depth significantly influences germination rates across different flow velocities. At H_2(2-3cm) burial depth, the germination rates of *Suaeda salsa* seeds under V_1(0.226m/s) and V_2(0.156m/s) flow velocity gradients are significantly lower than those under V_3(0.086m/s) to V_5(0.025m/s) gradients and the control group. The main reason is that the soil erosion depth caused by higher flow velocity and stronger water flow shear force is greater than that of V_3(0.086m/s)-V_5(0.025m/s), making it more difficult for seeds that already rely on soil roughness to germinate (Wang *et al*., 2022). At H_3(3-4cm) burial depth, the germination rates of *Suaeda salsa* seeds under all flow velocity gradients show a significant increase, reaching their maximum range. The differences in germination rates among the various flow velocity gradients are lower than that at other burial depths, and the changes in germination rates under different flow velocity gradients become less pronounced, although the maximum germination range remains at V_3(0.086m/s) to V_4(0.056m/s). Compared to the control group, the germination rates under V_1(0.226m/s), V_2(0.156m/s), and V_5(0.025m/s) flow velocity gradients are lower than under static water conditions, while those under V_3(0.086m/s) and V_4(0.056m/s) flow velocity gradients are higher. This indicates that at a seed burial depth of 3-4 cm, an appropriate range of flow velocity can promote the germination of *Suaeda salsa* seeds by replenishing water (Wang *et al*., 2022). At H_4(4-5cm) burial depth, the trend of changes in the germination rates of *Suaeda salsa* seeds under different flow velocity gradients is similar to that at H_3(3-4cm) burial depth. However, when the flow velocity changes from V_1(0.226m/s) to V_2(0.156m/s), the seed germination rate changes from increasing to decreasing. This is because the soil erosion depth has not reached 4-5cm, and due to the decrease in soil layer thickness and the increase in infiltration water, the seed germination rate under the V_1(0.226m/s) is higher than that under the V_2(0.156m/s).

Based on the above analysis, it can be found that under H_0(0cm), H_1(1-2cm), and H_3(3-4cm) depths, the germination rate of *Suaeda salsa* seeds is relatively higher when the flow velocity is within 0.036-0.086m/s. Under H_2(2-3cm) and H_4(4-5cm) depths, the germination rate is relatively higher when the flow velocity is within 0.059-0.086m/s. Under the same flow velocity condition, the germination rate of *Suaeda salsa* seeds at different depths, from high to low was H_3(3-4cm) > H_2(2-3cm) > H_4(4-5cm) > H_1(1-2cm) > H_0(0cm).

Effect of flow velocity on the height of *Suaeda salsa* seedlings: Height is one of the most basic indicators in plant observation, which can intuitively reflect the growth rate and uniformity of plants. At the same time, plant height can extent reflect the fertility of soil to a certain (Jing *et al*., 2024).

As shown in Fig. 3, at the H_0(0cm) burial depth, the height of *Suaeda salsa* seedlings first increases and then decreases with flow velocity changes from V_1(0.226m/s) to V_5(0.025m/s). When the flow velocity changes from V_1(0.226m/s) and V_2(0.156m/s) to V_3(0.086m/s), the height of *Suaeda salsa* seedlings significantly increases reaching its maximum range. Subsequently, as the flow velocity changes from V_3(0.086m/s) to V_4(0.056m/s) and V_5(0.025m/s), the height of *Suaeda salsa* seedlings gradually decreases. Compared to the control group, under the flow velocity gradients from V_1(0.226m/s) to V_5(0.025m/s), the heights of *Suaeda salsa* seedlings are significantly lower, indicating that at a sowing depth of 0cm, flow velocity has a strong inhibitory effect on the growth height of *Suaeda salsa* seedlings. At H_1(1-2cm) and H_2(2-3cm) burial depths, with changes in the flow velocity gradient, the height of *Suaeda salsa* seedlings also firstly increases and then decreases, reaching higher ranges under the flow velocity gradients of V_3(0.086m/s) and V_4(0.056m/s), similar to the trend at H_0(0cm) burial depth. Compared to the control group, at H_1(1-2cm) and H_2(2-3cm) burial depths, the heights of *Suaeda salsa* seedlings under flow velocity gradients of V_1(0.226m/s), V_2(0.156m/s), and V_5(0.025m/s) are lower than those under static water conditions, but the heights under gradients of V_3(0.086m/s) and V_4(0.056m/s) are higher, suggesting that within the sowing depths of 1-3cm, flow velocities in the range of 0.036-0.086m/s have a certain promoting effect on the growth of *Suaeda salsa* seedlings. Analysis reveals that flow velocities in the range of 0.036-0.086m/s have high water replenishment efficiency for soil at depths of 1-3cm, thus promoting the growth of *Suaeda salsa* seedlings. At H_3(3-4cm) and H_4(4-5cm) burial depths, the change in the height of *Suaeda salsa* seedlings is relatively mild. At H_3(3-4cm) burial depth, there is low variability in the height of *Suaeda salsa* seedlings between different flow velocity gradients, and the maximum height range is achieved under the flow velocity gradients of V_3(0.086m/s) and V_4(0.056m/s), where the heights of *Suaeda salsa* seedlings under these gradients are higher than that in the control group. At H_4(4-5cm) burial depth, the trend of the *Suaeda salsa* seedlings height is similar to that at H_3(3-4cm) burial depth; however, under the flow velocity V_1(0.226m/s), the heights of *Suaeda salsa* seedlings in plots A_1 and A_2 are higher than that in A_3 and A_4, which is due to soil erosion reducing the thickness of the soil layer and increasing infiltration water, leading to an increase in the height of *Suaeda salsa* seedlings (Wang *et al*., 2021). Compared to the control group, under the flow velocity gradients from V_1(0.226m/s) to V_5(0.025m/s), the heights of *Suaeda salsa* seedlings are higher, and the maximum height range is reached under the flow velocity V_3(0.086m/s).

Based on the above analysis, it can be found that at H_0(0cm) burial depth, when the flow velocity is within 0.059-0.086m/s, the height of the *Suaeda salsa* seedlings is relatively higher; at H_1(1-2cm), H_2(2-3cm), H_3(3-4cm), and H_4(4-5cm) burial depths, when the flow velocity is within 0.036-0.086m/s, the height of the *Suaeda salsa* seedlings is relatively higher. Under the same flow velocity gradient, the height of the *Suaeda salsa* seedlings at each burial depth, from high to low was H_3(3-4cm) > H_4(4-5cm) > H_2(2-3cm) > H_1(1-2cm) > H_0(0cm).

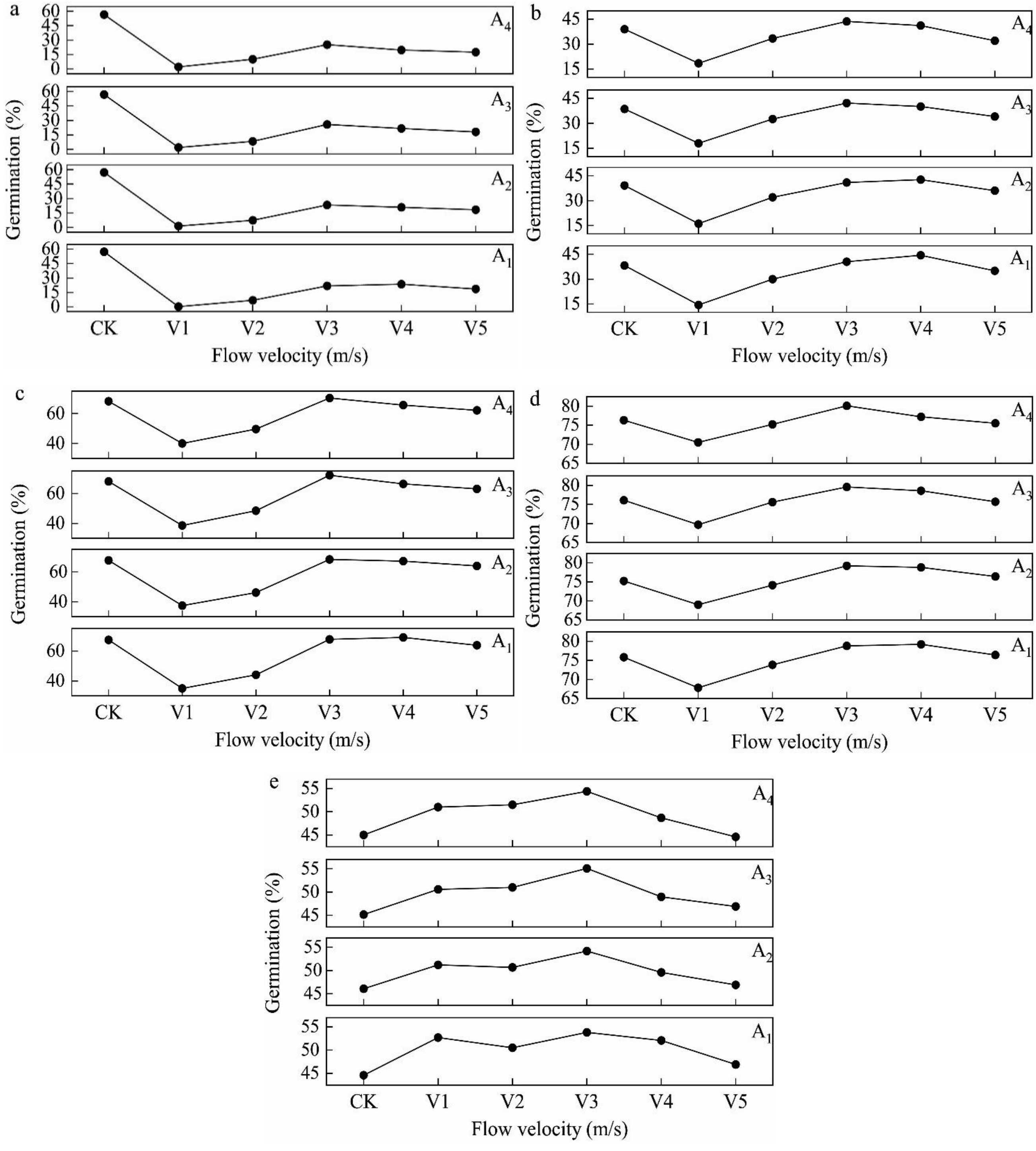

Fig. 2. Germination status of *Suaeda salsa* seed from Liaohe estuary under different flow velocity conditions. Note: when seed burial depth is H_0(a), when seed burial depth is H_1(b), when seed burial depth is H_2(c), when seed burial depth is H_3(d), when seed burial depth is H_4(e).

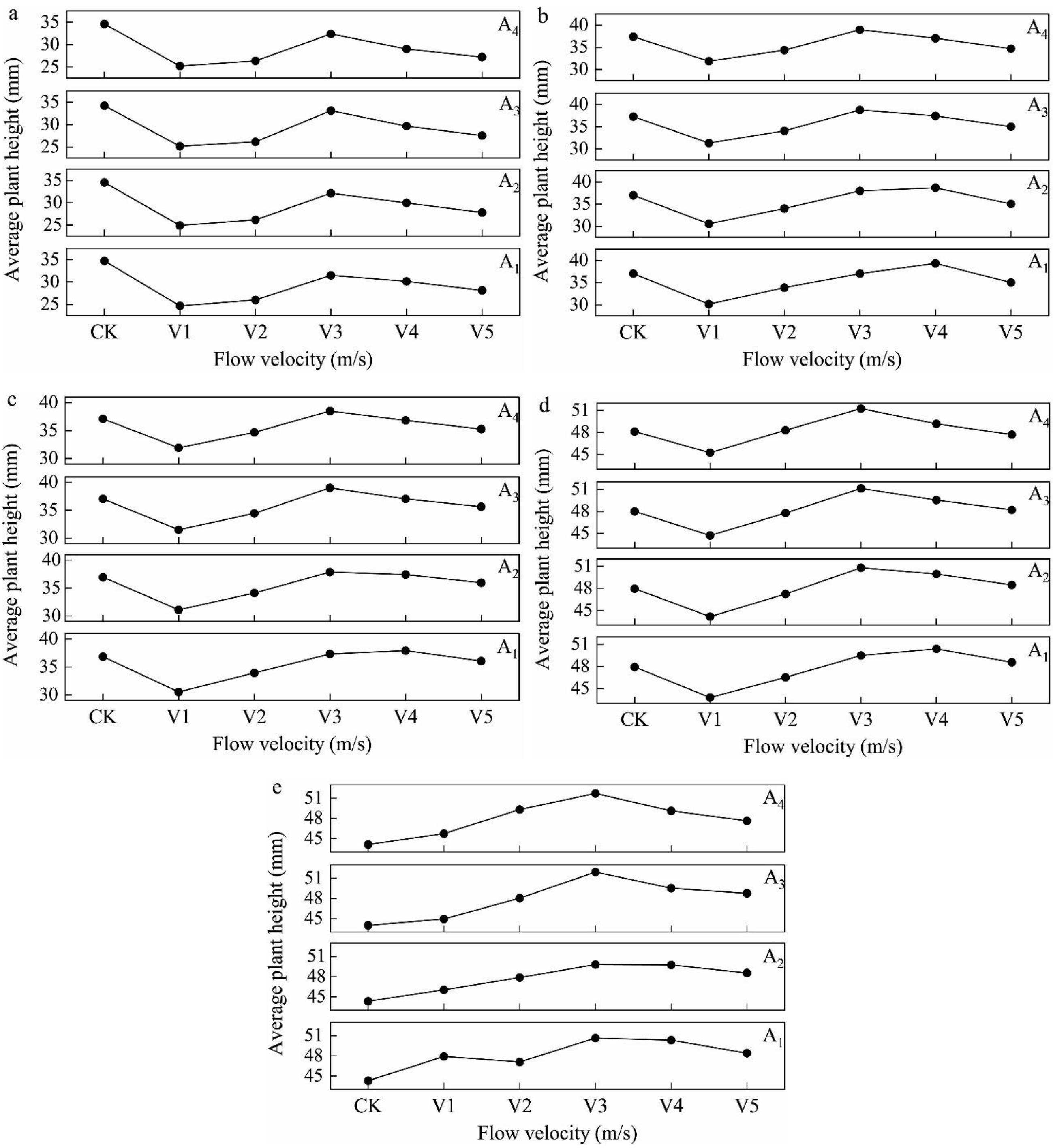

Fig. 3. The influence of flow velocity on vegetation height at different depths. Note: when seed burial depth is H_0(a), when seed burial depth is H_1(b), when seed burial depth is H_2(c), when seed burial depth is H_3(d), when seed burial depth is H_4(e).

Effect of flow velocity on *Suaeda salsa* seedlings fresh weight: Fresh weight refers to plants' mass in the natural state (Hosoda *et al*., 2024). Through Fig. 4 it can be seen that the trend of fresh weight of *Suaeda salsa* seedlings with flow velocity at different depths is similar to that of seedling height. At H_0(0cm) burial depth, as the flow velocity gradually decreases, the fresh weight initially increases then decreases. When the flow velocity changes from V_1(0.226m/s) to V_3(0.086m/s), the fresh weight of *Suaeda salsa* seedlings significantly increases, reaching its maximum range under V_3(0.086m/s). Following this, as the flow velocity changes from V_3(0.086m/s) to V_5(0.025m/s), the fresh weight of the seedlings decreases. Compared to the control group, the fresh weight under flow velocity gradients from V_1(0.226m/s) to V_5(0.025m/s) is significantly lower, consistent with the germination rate characteristics. At H_1(1-2cm) and H_2(2-3cm) burial depths, the trend of changes in the fresh weight of *Suaeda salsa* seedlings with varying flow velocities is largely consistent with that at H_0(0cm) burial depth, both showing an initial increase then decrease, reaching their maximum ranges under V_3(0.086m/s). However, compared to the control group, the fresh weight at H_1(1-2cm) and H_2(2-3cm) burial depths under flow velocity gradients V_3(0.086m/s), V_4(0.056m/s), and V_5(0.025m/s) is greater. At H_3(3-4cm) burial depth, as the flow velocity decreases, the change in the fresh weight still shows an initial increase then a decrease, but the differences between different flow velocity gradients are weaker than those between H_0(0cm)-H_2(2-3cm) burial depths, and reach the maximum range under V_3(0.086m/s)-V_4(0.056m/s) flow velocity gradients. It was found that the flow velocity range of the fresh weight of *Suaeda salsa* seedlings reached the maximum value, the flow velocity range of the height of *Suaeda salsa* seedlings reached the maximum value and the flow velocity range of the germination rate of *Suaeda salsa* reached the maximum value were basically coincident, indicating that flow velocity not only affects germination rate but also further affects the growth characteristics of *Suaeda salsa* seedlings. At H_4(4-5cm) burial depth, the differences in fresh weight changes of *Suaeda salsa* seedlings between different flow velocity gradients are further reduced, indicating that the effect of flow velocity on *Suaeda salsa* seedlings' fresh weight is relatively small at the burial depth of 4-5cm. It was found that the fresh weight of *Suaeda salsa* seedlings reached the maximum range under the flow velocity gradient of V_3(0.086m/s)-V_4(0.056m/s).

Through the above analysis, it was found that at H_0(0cm), H_1(1-2cm), and H_2(2-3cm) burial depths, the fresh weight of *Suaeda salsa* seedlings is relatively higher when the flow velocity is within 0.059-0.086m/s. At H_3(3-4cm) and H_4(4-5cm) burial depths, the fresh weight of the seedlings is relatively higher when the flow velocity is within 0.036-0.086m/s. Under the same flow velocity, the fresh weight of *Suaeda salsa* seedlings at different burial depths showed H_3(3-4cm)>H_2(2-3cm)>H_4(4-5cm)>H_1(1-2cm)>H_0(0cm).

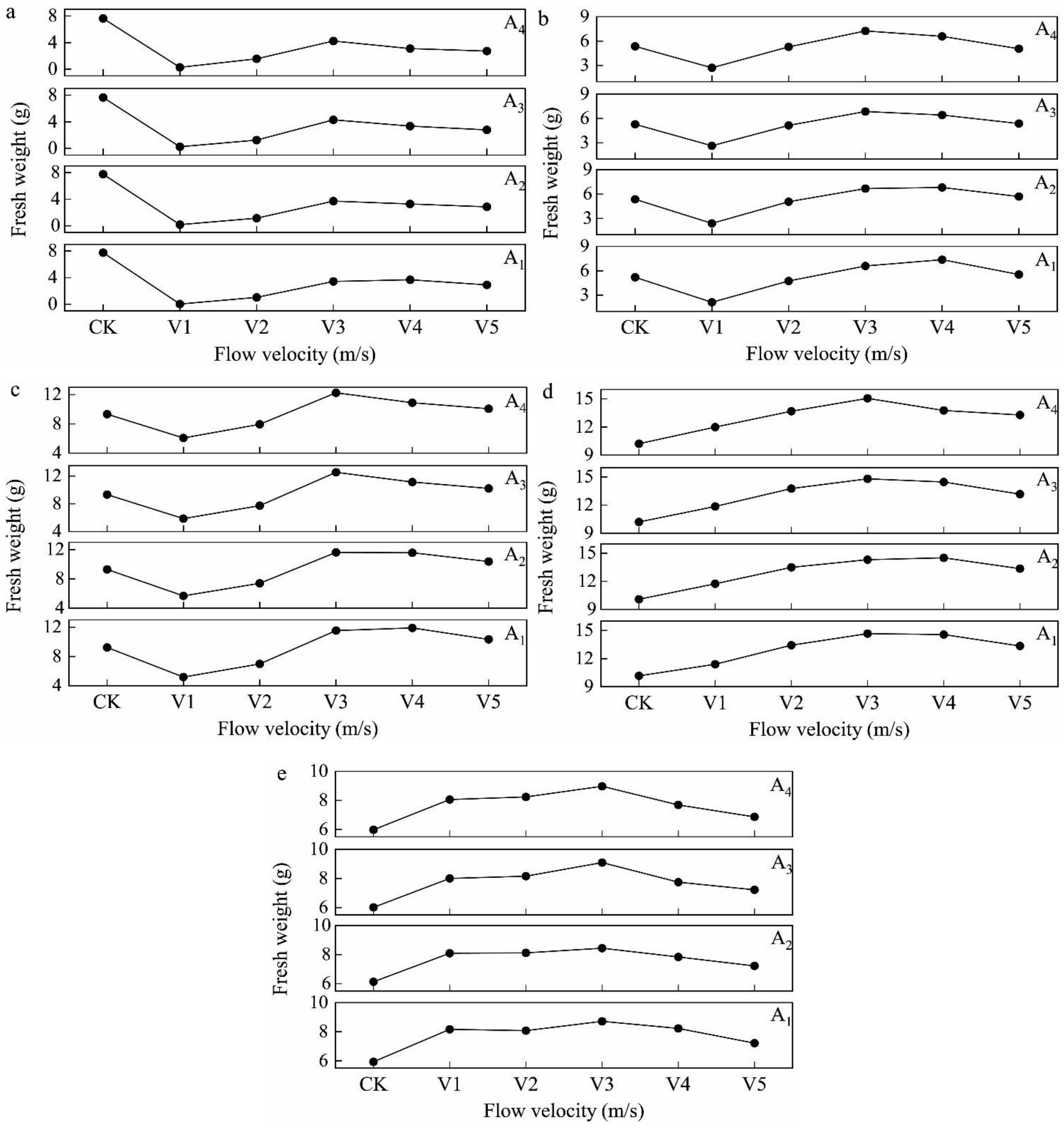

Fig. 4. The influence of flow velocity on vegetation fresh weight under different depth conditions. Note: when seed burial depth is H_0(a), when seed burial depth is H_1(b), when seed burial depth is H_2(c), when seed burial depth is H_3(d), when seed burial depth is H_4(e).

Conclusion

Under the same flow velocity, the overall trend of seed germination rate, plant height, and fresh weight at different burial depths was basically consistent. The seed germination rate, plant height, and fresh weight at H_3(3-4cm) burial depth conditions were higher than that at the other four burial depth conditions. At the same burial depth conditions, the response characteristics of seed germination rate, plant height, and fresh weight to flow velocity were basically consistent. In the range of 0.059-0.086m/s, the seed germination rate, plant height and fresh weight of each burial depth could reach the maximum range.

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