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Research Article

Trade-off strategies of individual clonal reproductive traits in Chinese wildrye (*Leymus chinensis* (Trin.) Tzvel) under NaCl stress

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Abstract

Soil salinity is a serious abiotic stress negatively affecting crop productivity and threatening the global food security. The extent of salt affected soils is continuously increasing due to poor irrigation, improper agricultural practices, and over-fertilization. This study was conducted to investigate the adaptive characteristics of individual growth and the changes of clonal components against salinity stress. The experiment was comprised of different treatments; control, 100 and 200 mmol L^{-1} salt stress. The results indicate that increasing concentration of salt stress decreased the individual plant biomass, leaf biomass and underground biomass, and increased stem biomass and plant height. Moreover, at 200 mmol L^{-1} the diameter and volume of the underground parts showed a small reduction whilst length and area of the underground part increased significantly. Besides this *Leymus chinensis* reduced the input of leaves and various seed plants, allocated more energy to the underground roots, and adopted the root configuration strategy of fine root extension to increase the length, area and volume of the roots. Under high concentration of salt stress; the underground bud reservoir expanded continuously, but the upward output was slightly decreased. This indicates that bud bank was expanded to prepare for reproduction, but the continuous salt stress seriously interfered with the underground clonal components of *L. chinensis*, and the nutrients and energy synthesized by cells were insufficient to support the continuous extension of rhizostems and the development of daughter plants. In conclusion, salt stress is majorly contributing to decrease the growth and biomass production of *L. chinensis,* however, salt had more pronounced negative impacts on upward growth as compared to underground growth.

Keywords: clone (asexual) propagation; *Leymus chinensis*; leaf traits; root architecture; salt stress

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Introduction

Saline-alkali is widely distributed problem across the globe with an area equivalent to about 10% of the arable land (Liu and Wang, 2021). In China around 12 million hectares are salt affected which accounts for 15% of the total cultivated area (Jiang *et al.,* 2023). The extent of salinity stress is continuously increasing which is restricting the sustainable crop production and destroying the ecological environment (Bogunovic *et al.*, 2019). Sheepgrass (*Leymus chinensis*) is one of the most important perennial grasses in China and it is an important source of feed of animals (Chen *et al.,* 2023). Generally, the distribution order of total biomass of sheep grass in whole growing seasons is remained as: vegetative growth > rhizomes > reproductive growth (Chen *et al.,* 2023). Under suboptimal environmental conditions, this grass allocates more nutrients to reproductive tillers to ensure population sustainability. Additionally, there exists an antagonistic relationship between sexual reproductive growth and vegetative growth (Guo *et al.,* 2023). The survival and development of *L. chinensis* indicating that it adapted to changing soil habitat conditions (Des Forges, 2016; Liu *et al.*, 2019). The distribution of roots in the soil directly reflects the growth of the underground part (rhizomes and roots) of the clone plant. In contrast to seedlings, high storage capacity of rhizome caused more resilient seedling development under stress conditions. Study showed that the total length of rhizomes and the length of rhizomes internode depicted an increasing sequence with increase in altitude, while the distance between adjacent branches did not show significant altitude difference (Banik, 2015).

The in-depth research of underground part makes more researchers to focus on bud reservoir. Compared with the seed bank, under extreme conditions, the perennial herb of *L. chinensis* is more likely to retain the bud bank, and the possibility of output to the next generation is much higher than that of the sexual reproduction bank represented by the seed bank. The bud bank and the seed bank, constitute the plant reproduction bank, and they represent two different reproductive strategies of plants (Chen *et al.,* 2022). Earlier study found that daughter shoot exported from underground bud bank in autumn (August to October) entered the stage of young spike differentiation (Zhang *et al.*, 2009), and the number of daughter shoot was closely related to the number of headings in the following year. In addition, studies have found that under natural habitats, production of new rhizomes; its length and density was considerably reduced in the year of high sexual reproductive resource input. Nonetheless, the density of tiller buds and tiller nodes is not reduced but significantly increased, resulting in a significant decrease in total bud density and total seed plant density. This signifies that elevated regenerative distribution influences not only the quantity of distinct cloned progeny in the *L. chinensis* population but also significantly impacts the dispersal distance of cloned progeny. Additionally, *L. chinensis* employs a comparatively more vigorous growth strategy during reproduction (Sun *et al.,* 2021). Our hypothesis was that salt stress could inhibit the overall growth and performance of *L. chinensis* however, it can have more toxic effects on above ground growth as compared to underground growth. This experiment was executed to determine the effect of different salinity levels on growth, physiological characteristics and nutrient concentration in plant parts of sheep-grass.

Materials and Methods

Experimental material

The experiment was executed in greenhouse condition, Northeast Normal University (43°51 'N, 125°19' E, 236 m a. s. l.), China. The greenhouse environment was maintained with daytime temperatures of 25 - 30 °C (max 35 °C under natural sunlight) and nighttime temperatures of 18 – 22 °C. Relative humidity ranged between 50% and 70%. For experiment, wild seedlings of *Leymus chinensis* were collected from Changling, Songyuan City, Jilin Province. Later, green and vigorous seedlings of uniform length were selected to transplant into plastic pot with a side length of 30 cm and a height of 0.5 m. We used fine sand as culture medium and 20 seedlings were planted in each pot by maintaining equal distance.

Stress conditions and treatment

Between May 15 and June 28, seedlings in the early growth stage (14 - 30 days). received the similar treatment, which included watering them with distilled water. After every four days, water containing half-strength Hoagland nutritional solution was applied. After designing two concentration gradients, salt stress treatment was started in July and it was continued until the late growth season. The study included three distinct salt stress levels: control, 100, and 200 mmol L^{-1}. The 100, and 200 mmol L^{-1} treatments were prepared by mixing half-strength Hoagland-nutritional solution with NaCl. The control group received 0.5 times the Hoagland nutrition solution. The seedlings (seedling stage) of *L. chinensis* were treated against stress between June 29 and August 24. The experiment contained 15 pots which were divided into five groups and each group had three pots. Treatments were applied on a daily basis from 17:00 to 18:00, following a 4-day cycle. The experiment was replicated three times. On the initial day, each pot received 1 L of treatment solution as per set treatments, followed by 0.5 L of water for the subsequent three days. Every experimental unit remained under shed for 8 weeks until treatment applications were completed.

Plant growth and photosynthetic related parameters

Fifteen seedlings of *L. chinensis* per pot were labelled to assess the height as well as leaf length weekly, and logged dynamic data throughout the growing season. By following the Blackman's formula, the relative height growth rate (RHGR) of *L. chinensis* was determined:

$$RHGR\ (H_i) = \frac{\ln(H_{i+1}) - \ln(H_i)}{T_{i+1} - T_i}$$

In above equation, H_i and H_{i+1} denotes plant height at Ti and T_{i+1}, respectively. The photosynthetic indices (photosynthesis rate (Pn), transpiration rate (Tr), stomatal conductance (Gs), intercellular CO_2 (Ci) and water use efficiency (WUE) of sheep-grass were determined using CIRAS-3 portable photosynthesizing apparatus (PP SYSTEMS, USA) once during the middle and late growing season (July and late August). For this, the leaf that was fully unfolded and located above the plant was chosen.

Leymus chinensis leaf and plant sample collection

Following a stress treatment that lasted for eight weeks, fresh samples of ten leaves were collected from every treatment and subjected to determine the fresh weight, and leaf area of each fresh leaf was measured by YMJ-B. Finally, leaves were oven-dried at 65 °C for 48 h to determine leaf dry weight. Destructive sampling was done to take plant samples. The plants were properly cleansed to remove root sand. The quantity of rhizomes, internode buds, terminal buds, and seed plants (tillers and rhizomes) was counted manually. Fresh root samples of 15 sheep-grass plants were selected and marked to measure to root length, diameter, area, volume and tip count by a root system scanner (Epson 11000 XL scanner). Different plant parts were separated manually and each part was used to determine its dry weight after placing at 65 °C in oven for 48 h.

Determination of total carbon (C), total nitrogen (N) and total phosphorus (P) content

For this, collected plants from all pots were subjected to separate the different plant parts (leaves, stems, rhizomes and roots) and grounded. Total C and total N was measured using Elementary various EL III, UK (Muñoz-Huerta *et al.,* 2013). Whereas, total P content was calculated using molybdenum-antimony resistance colorimetric method (721 spectrophotometer) (Wieczorek *et al*., 2022).

Data Analysis

Data pertaining the diverse traits was analyzed by analysis of variance technique and prior to analysis, the data was subjected to test homogeneity of recorded observations. The impact of salinity on the collected data was examined by two-factor ANOVA, and the differences among treatment means were analyzed by Tukey's multiple comparison test (Steel *et al.,* 1997).

Results

Effects of NaCl stress on individual characteristics of Leymus chinensis

In the presence of nutritional solution, *L. chinensis* exhibited a comparatively rapid rate of growth *(RHGR)* (Figure 1). After reaching the peak growth rate in mid-June RHGR continued to decline and the plant grew slowly. After salt stress (25 days) the RHGR of *L. chinensis* began to be slightly smaller than that of the control group. We found no difference among RHGR in control as well as treated with 100 mmol L^{-1}salinity level. The RHGR under 200 mmol L^{-1} salinity showed a significant downward trend after the stress treatment was started although it fluctuated slightly and it was significantly higher in control.

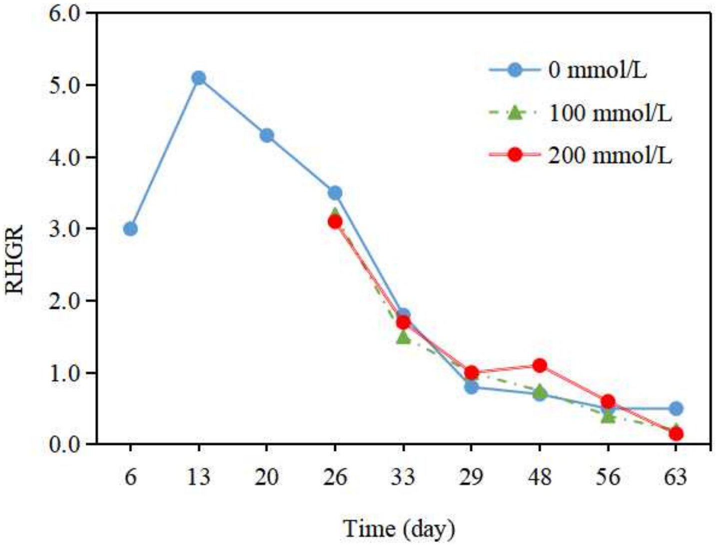

Figure 1. Effects of salt stress on relative height growth rate of *L. chinensis*

Note: The first measurement from June 5 is recorded as the first day of dynamic tracking. The stress treatment began on 29 June (day 24 of tracking)

Single plant height and biomass of sheepgrass

The results showed that under salt stress concentration; plant height was significantly increased. The plant height increased by 4.66% and 4.38% at 100 and 200 mmol L^{-1} than that of control treatment (Figure 2A).

With the increase of salt stress concentration, the plant biomass of sheepgrass decreased. The biomass per plant of *L. chinensis* decreased by 8.7% at 100 mmol L^{-1} and 19.62% at 200 mmol L^{-1} salinity than that of control treatment (Figure 2B).

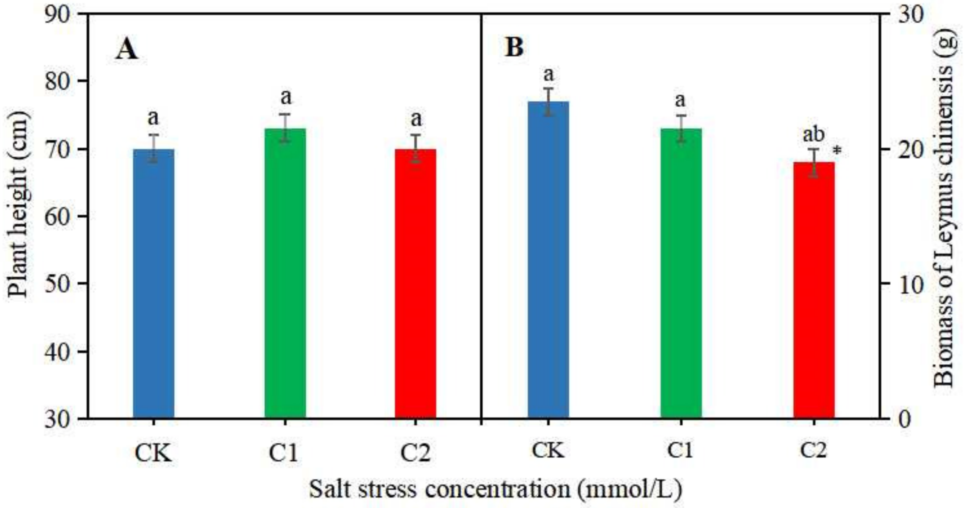

Figure 2. Effects of salt stress on plant height (A) and biomass (B) of *Leymus chinensis*

Note: CK stands for 0 mmol L^{-1}; C1 represents salt concentration of 100 mmol L^{-1}.C2 represents the salt concentration of 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

Effect of salt stress on biomass of aboveground and underground parts of sheepgrass

The above and below ground plant biomass decreased with increasing salinity stress There was no significant difference between the biomass of each plant, where the leaf biomass was decreased by 3.72% and 19.09% at 100 and 200 mmol L^{-1} salinity while stem biomass was decreased by 9.51% and 2.28% at 100 and 200 mmol L^{-1} salt stress (Figure 3, Table 1).

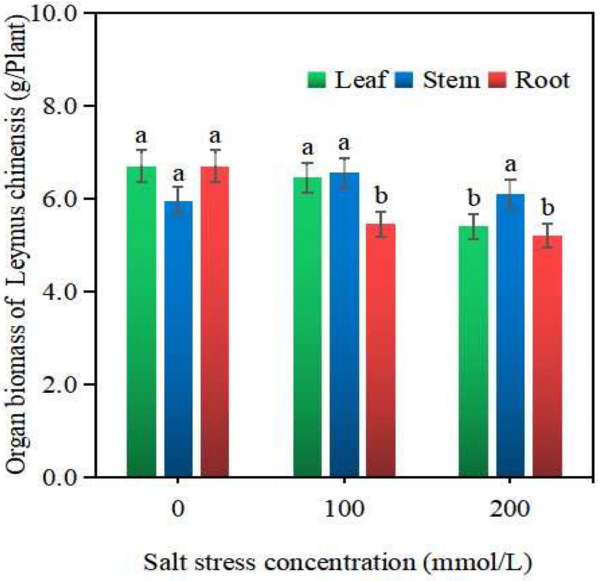

Figure 3. Effects of salt stress on organ's biomass of *L. chinensis*

Note: CK stands for 0 mmol L^{-1}; C1 represents salt concentration of 100 mmol L^{-1}.C2 represents the salt concentration of 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

Table 1. Correlation analysis between aboveground and underground part of *L. chinensis* with salt stress

Variables	Leaf	Stem	Root
Leaf	1	0.586^{**}	0.393^{**}
Stem	0.586^{**}	1	0.431^{**}
Root	0.393^{**}	0.431^{**}	1
Salt treatment	-0.122	0.018	-0.113

Note: * indicates a significant association at the 0.05 level. *Correlation is significant at the 0.05 level. ** indicates a significant correlation at the 0.01 level. **Correlation is significant at the 0.01 level

Effect of salt stress on C, N and P content in organs of L. chinensis

The carbon content in all organs was higher at 100 mmol L^{-1} salinity stress as compared to control (Figure 4A).

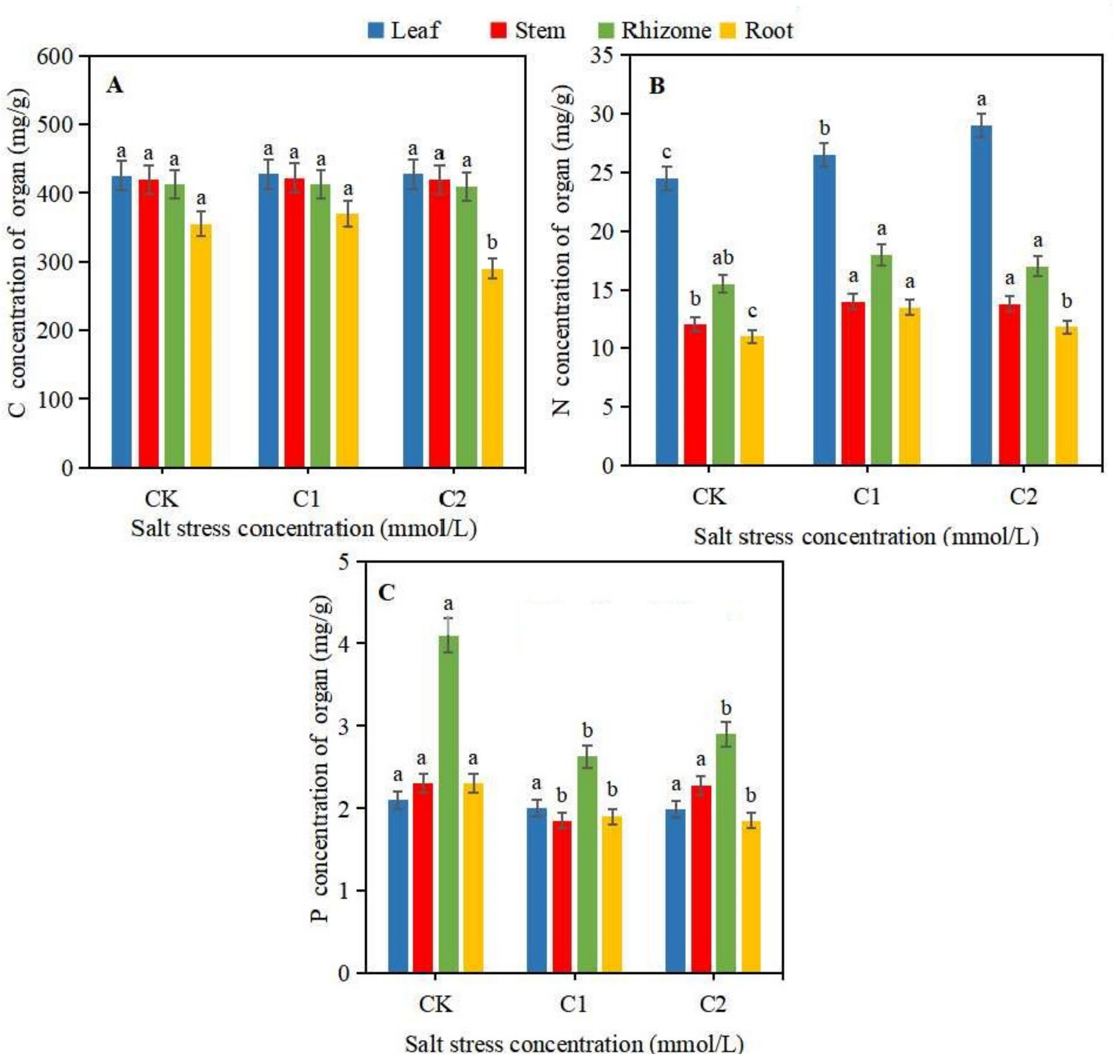

Figure 4. Effects of salt stress on carbon content (A), nitrogen content (B) and phosphorus content in organs of *L. chinensis*

Note: CK stands for 0 mmol L^{-1}; C1 represents salt concentration of 100 mmol L^{-1}.C2 represents the salt concentration of 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

There was no different among treatments for N contents. Under salt stress, N content in four organs of *L. chinensis* was increasingly noticed under the treatment comprising high salinity level (Figure 4B). Non-significant effect of different treatments was noticed for phosphorous, however, rhizomes had more P contents. By increasing salinity level, P concentration in different (leaf, roots) plant parts showed a decreasing trend. In addition (Figure 4C), C:N:P ratio was differently recorded in leaves. The increasing salt stress concentration led to diminish C/N ratio and maximize the N/P and C/P ratio (Figure 5A, B). The C/N ratio was decreased with increase in salts concentration and N/P and C/P ratio was maximum at 100 mmol L^{-1} salt stress. Moreover, C/N also decreased with increasing salt contents and ratio of N/P was maximum at 100 mmol L^{-1} (Figure 5C, D).

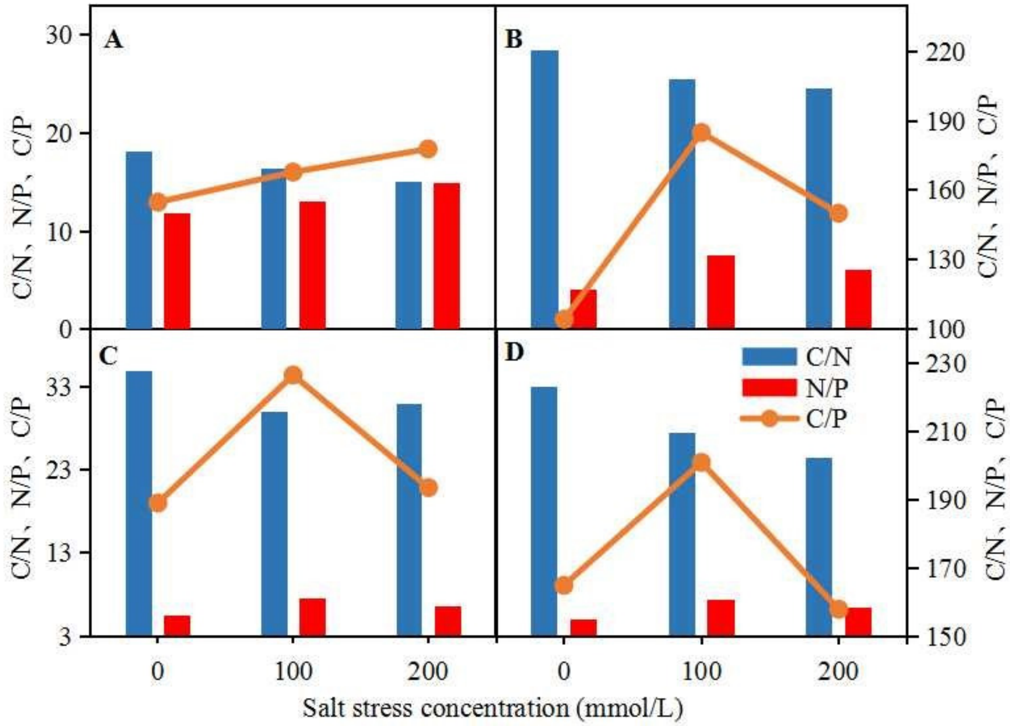

Figure 5. Effects of salt stress on the ratio of carbon, nitrogen and phosphorus in leaves (A), rhizomes (B), stems (C), and roots (D) of *Leymus chinensis*

Note: CK stands for 0 mmol L^{-1}; C1 represents salt concentration of 100 mmol L^{-1}; C2 represents the salt concentration of 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

Effect of salt stress on leaf traits

High leaf relative water content was recorded with increasing salinity level. The maximum relative water content was noted at 200 mmol L^{-1} salt stress followed after 100 mmol L^{-1} salt stress and lowest relative water content was noted in control (Figure 6A).

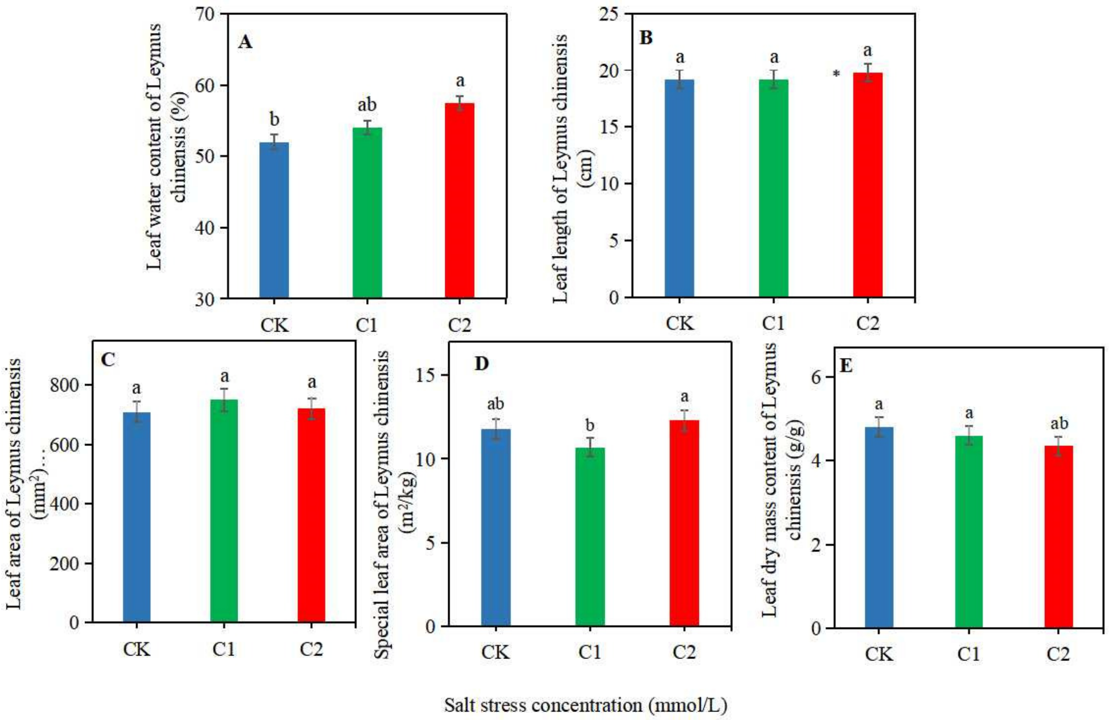

Figure 6. Effect of salt stress on *L. chinensis* leaf's water content (A), length (B), area (C), special area (D) and dry mass (E)

Note: CK represents 0 mmol L^{-1}; C1 represents salt concentration 100 mmol L^{-1}; C2 represents a salt concentration of 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

The leaf length of the herb increased, while, non-significant effect of salinity stress was recorded on leaf length (Figure 6B). The maximum leaf area was recorded under the 100 mmol L^{-1} salt concentration, as compared 200 mmol L^{-1} (Figure 6C). Moreover, salinity also reduced the specific leaf area and a reduction of 9.41% in specific leaf area was observed with 200 mmol L^{-1} salinity level (Figure 6D). Additionally, LDMC contents showed a marked reduction under increasing salts concentration. The concentration LDMC was decreased by 3.34% and 9.25% and 100 and 200 mmol L^{-1} salinity level (Figure 6E).

Effect of salt stress on photosynthetic index of L. chinensis

Salt stress showed a negative impact on gas exchange characteristics. The Tr and stomatal conductance were decreased by 9.24% and 43.89% at 100 mmol L^{-1} concentration of salinity than control. Water use efficiency (WUE) increased significantly with increase in salt concentration, and it showed an increase of 8.63% and 21.72% at 100 mmol L^{-1} and 200 mmol L^{-1} than control group, respectively. Intercellular CO_2 concentration decreased by 3.93% and 5.99% at 100 mmol L^{-1} and 200 mmol L^{-1}, respectively, compared to control treatment (Figure 7).

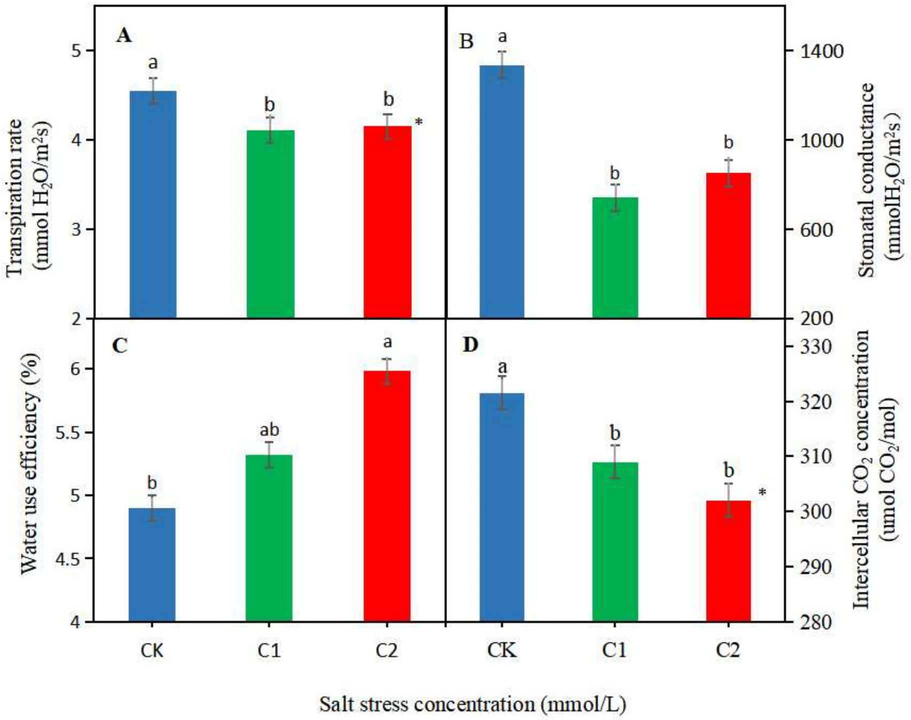

Figure 7. Effects of mid-growth season (July 20) Effects of salt stress on photosynthetic indexes of *L. chinensis:* traspiration rate (A), stomatal conductance (B), water use efficiency (D) and intercellular Co^2 concentration

Note: CK stands for 0 mmol L^{-1}; C1 represents the salt concentration of 100 mmol L^{-1}; C2 represents the salt concentration of 200 mmol L^{-1}. Different letters indicate significant difference between treatments (p<0.05), and * indicates significant difference between different treatments ($p < 0.05$)

Net photosynthesis (Pn) under salt treatment at 100 mmol L^{-1} was decreased compared to the control group. The results indicated that Pn of sheep grass was lower in July and net Pn also showed a marked reduction with increasing concentrations of salts stress. The net photosynthetic rate at 100 mmol L^{-1} and 200 mmol L^{-1} salt concentration was 14.27% and 14.37% lower than that of control group, respectively (Figure 8).

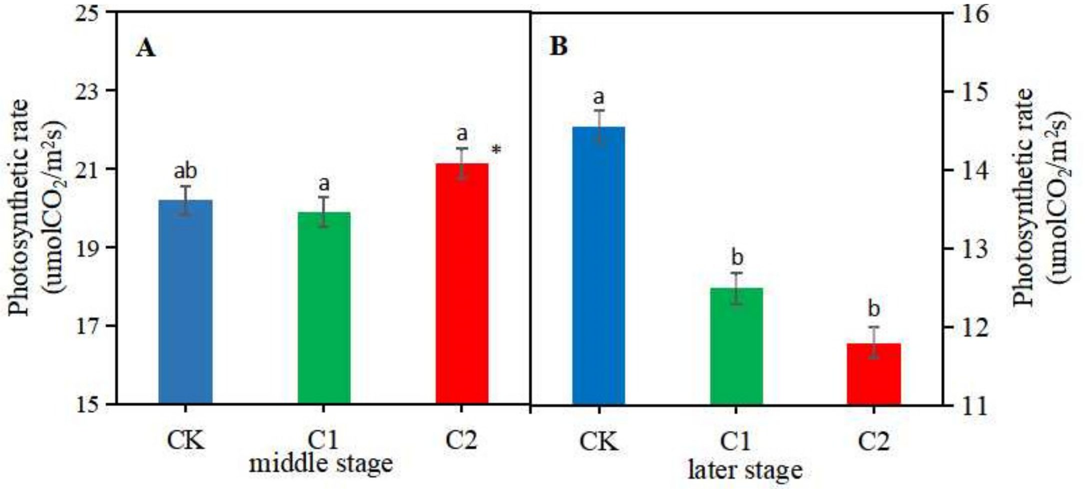

Figure 8. Effects of mid-late salt stress on photosynthetic indexes of *L. chinensis* at middle age (A) and at later age (B)

Effects of salt tress on the subsurface configuration of L. chinensis

Salt stress also imposed a negative impact on underground plant parts. The underground length was the smallest at 100 mmol L^{-1} and it was 10.67% less as compared to control (Figure 9A). Salinity decreased the lower diameter of the grass by 16.31% at 100 mmol L^{-1} as compared to control (Figure 9B). Additionally, the underground area was decreased by 21.58% at 100 mmol L^{-1} and 19.67% at 200 mmol L^{-1} (Figure 9C, D).

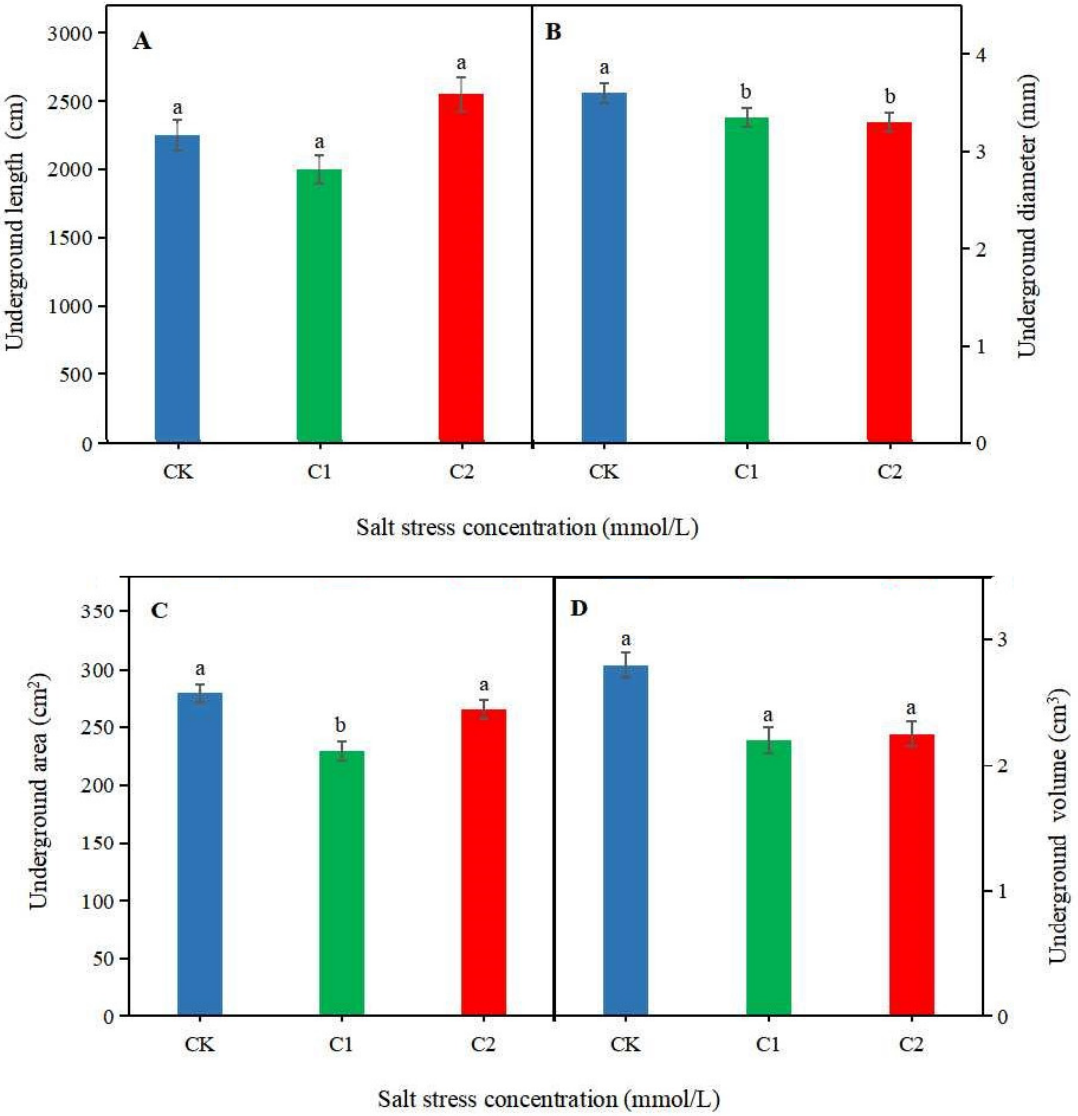

Figure 9. Effects of salt stress on the length (A), diameter (B), area (C) and volume (D) of *L. chinensis* underground part

Note: CK represents 0 mmol L^{-1}; C1 represents salt concentration 100 mmol L^{-1}; C2 represents salt concentration 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

Correlation analysis of various indicators in the underground part

The results indicate that salt stress reduced diameter, length, area and volume of *L. chinensis* and there was significant positive linking among these four traits. The positive relationship between the diameter of the lower part and salt treatment was not recorded (Table 2).

Table 2. Correlation analysis of underground part of *L. chinensis* with salt stress

Variables	Diameter	Length	Area	Volume
Diameter	1	-	-	-
Length	0.556**	1	-	-
Area	0.702**	0.948**	1	-
Volume	0.762**	0.876**	0.979**	1
Salt treatment	0.046	-0.177	-0.046	-0.117

Note: ** indicates a significant correlation at the 0.05 level. **Correlation is significant at the 0.05 level

Effect of salt stress on the clonal growth of L. chinensis

Salt stress treatment had a differential effect on the number of total daughter strains. The total daughter strains were reduced by 21.63% and 24.44% at 100 mmol L^{-1} and 200 mmol L^{-1} as compared to control (Figure 10A). Salt stress also reduced the total buds per mother plant and it was decreased by 14% and 3% at 100 mmol L^{-1} and 200 mmol L^{-1} (Figure 10B). In salinity, the clonal index was decreased by 13.08% at 100 mmol L^{-1} and by 1.41% at 200 mmol L^{-1} The rhizomes shoot number were also reduced by 20.75% and 15.46% (Table 3).

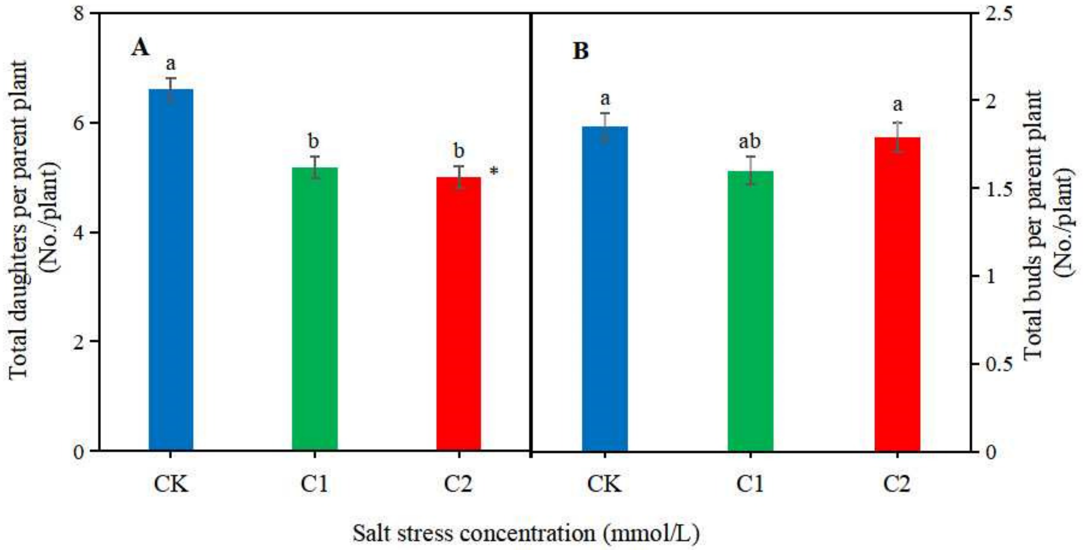

Figure 10. Effects of salt stress on total plant number (A) and total buds' number (B) of *L. chinensis*

Note: CK stands for 0 mmol L^{-1}; C1 represents the salt concentration of 100 mmol L^{-1}; C2 represents the salt concentration of 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

Table 3. Clonal growth traits of *L. chinensis* under salt stress

Clonal growth	**Stress concentration**		
	CK	**C1**	**C2**
The tillering plant	1.9722 ± 0.19a	1.7143 ± 0.21a	2.00 ± 0.24a
Rhizome shoot plants	1.6944 ± 0.45a	1.3429 ± 0.36a	1.4324 ± 0.29a
Tiller bud	0.1667 ± 0.09a	0.0857 ± 0.05a	0.1081 ± 0.05a
Internode buds of the rhizome	2.0278 ± 0.34a	1.6571 ± 0.36a	2.1622 ± 0.37a
Root tip bud	0.5833 ± 0.12a	0.6286 ± 0.15a	0.3514 ± 0.09a

Note: CK represents 0 mmol L^{-1}; C1 represents salt concentration of 100 mmol L^{-1}; C2 represents salt concentration of 200 mmol L^{-1}

Effect of salt stress on contents of Na$^+$, K$^+$ and Na$^+$ / K$^+$ in underground buds

As salt stress increased, the Na$^+$ content in the bud was increased by 88.94% and 138.13% at 100 mmol L^{-1} and 200 mmol L^{-1} over control treatment (Figure 11A). This highlight the non-significant impact of salinity on shoot K$^+$ concentration and there was no significant change in K$^+$ concentration at 100 mmol L^{-1} and 200 mmol L^{-1} salinity level (Figure 11B). Further, salinity showed significant impact for Na$^+$/K$^+$, and Na$^+$/K$^+$. Na$^+$/K$^+$ was maximized by 85.7% and 146.98% over the control group, respectively, at 100 mmol L^{-1} and 200 mmol L^{-1} salt stress (Figure 11C).

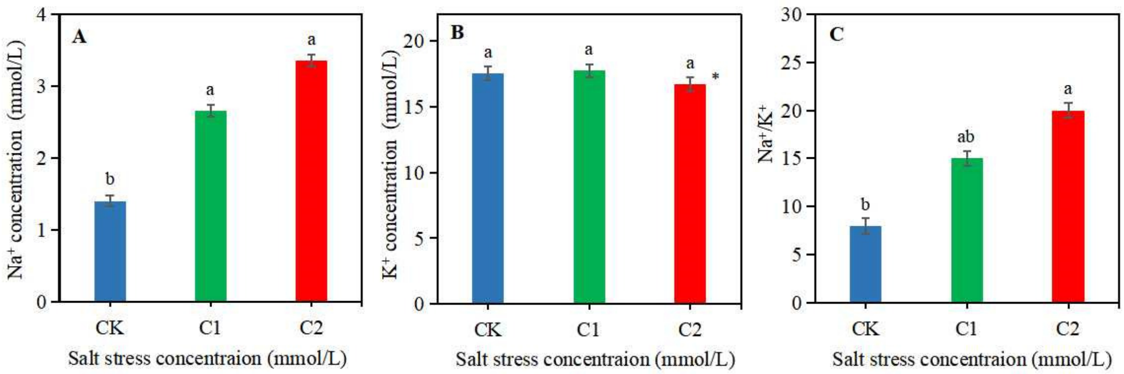

Figure 11. Effects of salt stress on Na$^+$ content (A), K$^+$ content (B) and Na$^+$ /K$^+$ ratio (C) of *L. chinensis*

Note: CK represents 0 mmol L^{-1}; C1 represents salt concentration 100 mmol L^{-1}; C2 represents salt concentration 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

Effect of salt stress on anions content of the underground bud

Data showed a significant effect of different salinity levels compared with control group for Cl$^-$ concentration. The concentration of Cl$^-$ was increased by 33.33% at 100 mmol L^{-1} and 44.73% at 200 mmol L^{-1} than control treatment (Figure 12A). Increasing concentration of salts decreased the concentration of NO$_3^-$ in plant shoots (Figure 12B). Conversely, $H_2PO_4^-$ concentration in underground shoots was increased with increasing salinity level (Figure 12C). Moreover, SO_4^{2-} content of the underground shoots at 100 mmol L^{-1} and 200 mmol L^{-1} were decreased by 4.48% and 4.76%, respectively, when compared to control treatment (Figure 12D).

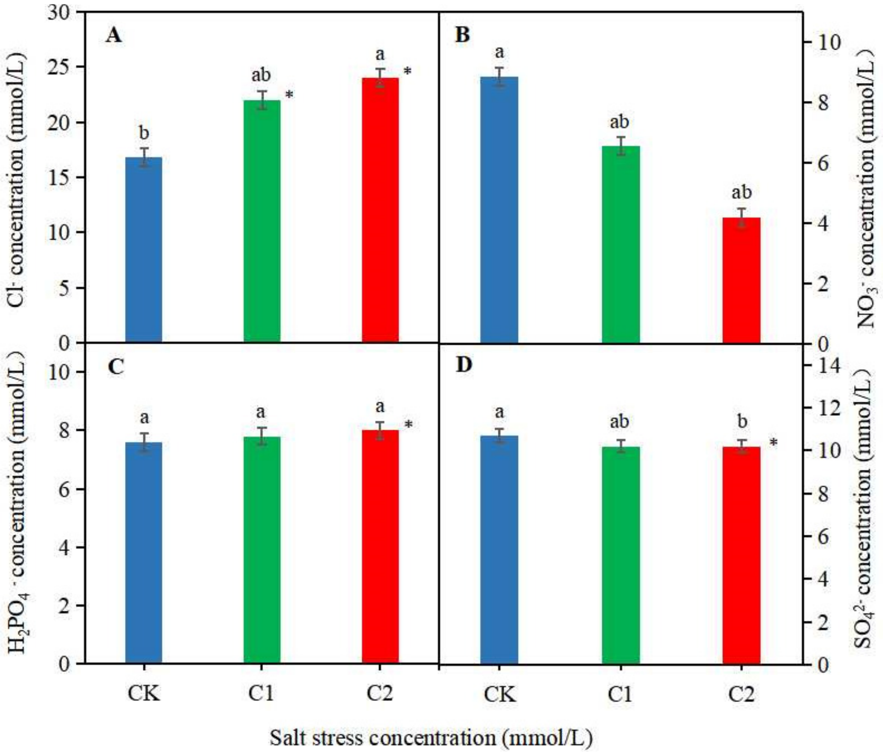

Figure 12. Effects of salt stress on Cl^- content (A), NO_3^- content (B), $H_2PO_4^-$ content (C) and SO_4^{2-} content (D) of *L. chinensis* buds

Note: CK represents 0 mmol L^{-1}; C1 represents salt concentration 100 mmol L^{-1}; C2 represents salt concentration 200 mmol L^{-1}. Different letters indicate significant difference between treatments ($p < 0.05$)

Discussion

Effects of salt stress on individual characteristics of Leymus chinensis

The results indicated that salt stress has a negative impact on plant growth, however, increasing salts concentration increased plant height and stem indicating that salt stress has a promoting effect on *L. chinensis* growth (Zheng *et al.*, 2004). Salt stress had a non-significant impact on carbon, nitrogen, and phosphorus concentration in different organs. Recent advancements in ecological stoichiometric studies have established the growth rate hypothesis as a prominent theory (Min *et al.*, 2007; Sardans *et al.*, 2021). Biological organisms exhibiting rapid growth demonstrate elevated nitrogen (N) and phosphorus (P) levels. The adjustment of an organism's life history can lead to alterations in the C:N:P ratio of the organism (Aerts and Chapin III, 1999; Weider *et al.*, 2005). In this investigation, the lowest relative height growth rate and highest C/P and N/P ratios occurred at 100 mmol L^{-1} salt concentration. This was consistent with the typical "growth rate theory" indicating that plant height, stem biomass per plant and C/N ratio were the largest under this concentration (Chen *et al.*, 2021).

Effects of salt stress on leaf traits of L. chinensis

Leaf biomass and dry matter production was decreased with increasing salts concentration. The net photosynthetic rate was significantly different in the middle and late growing season, showed the salt treatment decreased the leaves of *L. chinensis*. This aligns with earlier studies of Li *et al.* (2016), and Li *et al.* (2017). *L. chinensis* acclimatizes to different degrees of soil salinization by changing and adjusting the ecological stoichiometric ratio from inside the body (Lu *et al.*, 2018). Salt stressed in sheep-grass leaves could be adjusted for specific duration, which was reflected in the photosynthetic indexes such as net photosynthetic rate and stomatal conductance of leaves. The increase of stomatal conductance increases the CO_2 content in the leaves, thus increasing the photosynthetic rate (Ma *et al.*, 2008). However, if the stomatal regulation is too large, it will accelerate the water loss of the leaves and increase the transpiration rate (Lawson and Blatt, 2014; Liu *et al.*, 2014). Under soil conditions with different salt stress levels, various physiological indexes of *L. chinensis* produced a series of adaptive responses, and it could grow normally in moderate and severe salt environments. However, after continuous salt stress, the water regulation system in the leaves was disturbed, and the water status of the leaves substantially improved (Belay *et al.*, 2002; Liu *et al.*, 2014; Acosta-Motos *et al.*, 2017; Yang *et al.*, 2021).

Effects of salt stress on subsoil configuration of L. chinensis

The underground biomass production of *L. chinensis* (rhizomes and roots) showed a decreasing trend with increasing salts concentration. Under no salinity, the contents of C and P in the roots and stems were very high, and the values of N/P and C/P were very small. The underground part of *L. chinensis* was in a good development condition, and the space was constantly expanding to absorb more nutrients. When 100 mmol L^{-1} salt was imposed the underground biomass decreased slightly when compared to control treatment, and the C and N contents in the roots and stems were both higher, while the C/N value was small (Wen *et al.*, 2022). According to the growth rate theory, the underground part could adapt to the salt treatment at this time, and the underground part could protect itself while continuously output to the above-ground part, especially in the growth of the stem (plant height). These results indicated that the "economic" investment of *L. chinensis* gave priority to rhizomes, maintained the growth of coarse rhizomes, reduced the expansion degree and development of fibrous roots, and mainly adopted the strategy of reducing root development and ensuring the normal development of rhizomes. Under high salinity (200 mmol L^{-1}), the aboveground and underground biomass decreased significantly which was linked with the fact that C/N, N/P and C/P ratios of the roots and stems were not large.

Effects of salt stress on clonal growth of L. chinensis

The low salt treatment concentration (100 mmol L^{-1}) limited the development of underground bud bank and the output of aboveground seed. However, under high concentration (200 mmol L^{-1}) of salt treatment, the underground bud reservoir expanded continuously which aligns with previous studies (Hohmann *et al.*, 2017). The results indicated that the bud bank was expanded to meet the needs of breeding due to salt stress, but the nutrients and energy were insufficient to support the continued development of daughter plants, which was a trade-off strategy for the growth and reproduction of *L. chinensis* under limited and harsh environment. And there are trade-offs between different kinds of seed and different kinds of buds (Zhang *et al.*, 2009). The maximum value was reached in late August, which was the sample recovery time for this experiment (Altig *et al.*, 2020). By the end of October, the proportion of tillering buds increased dramatically, so it is inferred that the tillering buds in this experiment are at the peak of growth and development. The similar results were found, indicating that Na^+, is a harmful ion, requires less energy and is taken in first when *L. chinensis* absorbs and accumulates organic solutes. Furthermore, the absorption of K^+ is inhibited, and the proportion balance of Na^+/K^+ is destroyed, resulting in changes in the penetration potential of the cell, and abnormal accumulation of NO_3^-, $H_2PO_4^-$, SO_4^{2-} and other anions (Hossen, 2017).

Effect of NaCl stress on individual characteristics of sheepgrass

The increasing concentration of salts decreased the plant biomass which aligns with previous studies reporting that increasing NaCl level decreased the leaf biomass per plant and dry matter content (Shao *et al.*, 2016). The increase of C/P and N/P ratios in leaves and the significant difference of net photosynthetic rate in the middle and late growing season indicated that *L. chinensis* adapted to different degrees of soil salinization by changing and adjusting its ecological stoichiometric ratio, water content and osmotic pressure. The underground biomass of *L. chinensis* showed a slow decline at first and then a sharp decline under increasing salt stress. During this process, the contents and ratios of chemical elements (carbon, nitrogen and phosphorus) in rhizomes and roots were regulated to a certain extent. Under 100 mmol L^{-1} salt treatment, the underground biomass decreased, and the C and N contents in rhizomes and roots were both higher, while the C/N value was small, indicating that the underground part of *L. chinensis* could still guarantee itself while continuously output to the above-ground part. In terms of resource allocation, the rhizomes were preferentially selected to reduce the expansion degree and reduce the development of fibrous roots. When treated with 200 mmol L^{-1} salt, the diameter and volume of the underground part decreased little, while the length and area of the underground part increased significantly, indicating that the underground part of *L. chinensis* tried to seek favorable resources by expanding the space to escape the stress of the current environment. These results indicated that the bud bank was expanded to prepare for reproduction, but the nutrients and energy were insufficient to support the continued development of daughter plants, which was a trade-off strategy for the growth and reproduction of *L. chinensis* in a limited and harsh environment (Tozer *et al.*, 2021).

Conclusion

Salinity stress imposed a negative impact on growth and physiological traits of sheep-grass. However, sheep-grass showed an increase of C/P and N/P ratios in leaves and the significant difference in net photosynthetic rate in the middle and late growing season to adapt to different degrees of soil salinization by changing and adjusting its ecological stoichiometric ratio, water content and osmotic pressure. Moreover, bud bank of *L. chinensis* was also increased for reproduction however, continuous salinity stress inferred and reduced the underground clonal components of *L. chinensis*. Therefore, individual *L. chinensis* makes trade-offs between and within organs to deal with salinity stress.

Authors' Contributions

Conceptualization: ZWG, YHC; Data curation: MC, LJH; Formal analysis: YQQ, MYB, XNL; Funding acquisition: ZWG; Investigation: ZWG, YHC; Methodology: ZWG, YHC; Project administration: ZWG; Resources: ZWG, JYL, QL, GG; Software: ZWG, GG, YHC; Supervision: ZWG, YHC, CSM; Validation: YGM, ZWG; Visualization: YGM, CMS; Roles/Writing - original draft: ZWG, YHC; and Writing - review & editing: ZWG, YHC, CSM.

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.

References

Acosta-Motos JR, Ortuño MF, Bernal-Vicente A, Diaz-Vivancos P, Sanchez-Blanco MJ, Hernandez JA (2017). Plant responses to salt stress: adaptive mechanisms. Agronomy 7(1):18. *https://doi.org/10.3390/agronomy7010018*

Aerts R, Chapin III FS (1999). The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. Advances in Ecological Research. Academic Press 30:1-67. *https://doi.org/10.1016/s0065-2504(08)60016-1*

Altig D, Baker S, Barrero JM, Bloom N, Bunn P, Chen S, … Thwaites G (2020). Economic uncertainty before and during the COVID-19 pandemic. Journal of Public Economics 191:104274. *https://doi.org/10.1016/j.jpubeco.2020.104274*

Banik RL (2015). Morphology and Growth. In Liese W, Köhl M (Eds) Bamboo Vol 10. Tropical Forestry. Springer Cham pp 43-89. *https://doi.org/10.1007/978-3-319-14133-6_3*

Belay A, Claassens A, Wehner F (2002). Effect of direct nitrogen and potassium and residual phosphorus fertilizers on soil chemical properties, microbial components and maize yield under long-term crop rotation. Biology and Fertility of Soils 35:420-427. *https://doi.org/10.1007/s00374-002-0489-x*

Bogunovic I, Fernández MP, Kisic I, Marimón MB (2019). Agriculture and grazing environments. In Pereira P (Ed). Advances in Chemical Pollution, Environmental Management and Protection. Elsevier pp 23-70. *https://doi.org/10.1016/bs.apmp.2019.07.005*

Chen YD, Moles A, Bu ZJ, Zhang MM, Wang ZC, Zhao HY (2021). Induced defense and its cost in two bryophyte species. American Journal of Botany 108: 777-787. *https://doi.org/10.1002/ajb2.1654*

Chen XS, Huang Y, Cai YH, Hou ZY, Deng ZM, Li F, … Xie YH (2022). Belowground seed and bud banks play complementary roles in the potential recruitment of dominant macrophyte communities in a Yangtze River-connected floodplain wetland. Frontiers in Plant Science 13:1075496. *https://doi.org/10.3389/fpls.2022.1075496*

Chen H, Xiong F, Wu Q, Wang W, Cui Z, Zhang F,… Zhang L (2023). Estimation of energy value and digestibility and prediction equations for sheep fed with diets containing *Leymus chinensis* hay. Agriculture 13(6):1213. *https://doi.org/10.3390/agriculture13061213*

Des Forges R (2016). China's roles in world history and historiography. Frontiers of history in China 11(2):117-246.

Guo J, Li H, Yang Y, Yang X (2023). Clonal dominant grass Leymus chinensis benefits more from physiological integration in sexual reproduction than its main companions in a meadow. Frontiers in Plant Science 14:1205166. *https://doi.org/10.3389/fpls.2023.1205166*

Hohmann HP, van Dijl JM, Krishnappa L, Prágai Z (2017). Host organisms: *Bacillus subtilis.* In: Wittmann C, Liao JC (Eds). Industrial Biotechnology: Microorganisms Vol 2. Wiley pp 221-297. *https://doi.org/10.1002/9783527807796.ch7*

Hossen MS (2017). Comparative physiology of salinity and drought stress tolerance in indica and japonica rice seedlings, MS Thesis. Sher-e-Bangla Agricultural University Dhaka, Bangladesh.

Jiang L, Qiu G, Yu X (2023). Identification and spatial analysis of land salinity in China's Yellow River delta using a land salinity monitoring index from harmonized UAV-landsat imagery. Sensors 23(17):7584. *https://doi:10.3390/s23177584*

Lawson T, Blatt MR (2014). Stomatal size, speed, and responsiveness impact on photosynthesis and water use efficiency. Plant Physiology 164:1556-1570. *https://doi.org/10.1104/pp.114.237107*

Li Q, Li Y, Wang D, Guo D, Zhao Y (2016). Research on nutrient responses of soil-plant system in community succession of degraded grassland based on ecological stoichiometry. Journal of Jilin Agricultural University 38:693-702.

Li Y, Liang S, Zhao Y, Li W, Wang Y (2017). Machine learning for the prediction of *Leymus chinensis* carbon, nitrogen and phosphorus contents and understanding of mechanisms underlying grassland degradation. Journal of Environmental Management 192:116-123. *https://doi.org/10.1016/j.jenvman.2017.01.047*

Liu B, Kang C, Wang X, Bao G (2014). Physiological and biochemical response characteristics of *Leymus chinensis* to saline-alkali stress. Transactions of the Chinese Society of Agricultural Engineering 30:166-173.

Liu G, Qi D, Dong X, Liu H, Liu S (2019). Basic knowledge of sheepgrass (*Leymus chinensis*). In: Liu G, Li X, Zhan, Q (Eds). An environmentally friendly native grass for animals. Springer, Singapore 1-51. *https://doi.org/10.1007/978-981-13-8633-6_1*

Liu L, Wang B (2021). Protection of halophytes and their uses for cultivation of saline-alkali soil in China. Biology 10:353. *https://doi.org/10.3390/biology10050353*

Lu Q, Bai J, Zhang G, Zhao Q, Wu J (2018). Spatial and seasonal distribution of carbon, nitrogen, phosphorus, and sulfur and their ecological stoichiometry in wetland soils along a water and salt gradient in the Yellow River Delta, China. Physics and Chemistry of the Earth, Parts A/B/C 104:9-17. *https://doi.org/10.1016/j.pce.2018.04.001*

Ma H, Liang Z, Kong X, Yan C, Chen Y (2008). Effects of salinity, temperature and their interaction on the germination percentage and seedling growth of *Leymus chinensis* (Trin.) Tzvel.(Poaceae). Acta Ecologica Sinica 27:4710-7.

Min WMW, Li XLX, Yan LYL (2007). Review of adaptation mechanism of plants to salt stress. Scientia Silvae Sinicae 43:111-117.

Muñoz-Huerta RF, Guevara-Gonzalez RG, Contreras-Medina LM, Torres-Pacheco I, Prado-Olivarez J, Ocampo-Velazquez RV (2013). A review of methods for sensing the nitrogen status in plants: advantages, disadvantages and recent advances. Sensors 13(8):10823-10843. *https://doi.org/10.3390/s130810823*

Sardans J, Janssens IA, Ciais P, Obersteiner M, Peñuelas J (2021). Recent advances and future research in ecological stoichiometry. Perspectives in Plant Ecology, Evolution and Systematics 50:125611. *https://doi.org/10.1016/j.ppees.2021.125611*

Shao T, Li L, Wu Y, Chen M, Long X, Shao H, Liu Z, Rengel Z (2016). Balance between salt stress and endogenous hormones influence dry matter accumulation in Jerusalem artichoke. Science of the Total Environment 568:891-898. *https://doi.10.1016/j.scitotenv.2016.06.076*

Steel RGD, Torrie JH, Dickey D (1997). Principles and Procedures of statistics: a biometric approach (3rd ed). McGraw-Hill Book Co., New York, USA pp 663-666.

Sun H, Zheng C, Chen T, Postma JA, Gao Y (2021). Motherly care: how *Leymus chinensis* ramets support their offspring exposed to saline-alkali and clipping stresses. Science of the Total Environment 801:149675. *https://doi.org/10.1016/j.scitotenv.2021.149675*

Tozer K, Douglas G, Dodd M, Müller K (2021). Vegetation options for increasing resilience in pastoral hill country. Frontiers in Sustainable Food Systems 5:550334. *https://doi.org/10.3389/fsufs.2021.550334*

Weider LJ, Elser JJ, Crease TJ, Mateos M, Cotner JB, Markow TA (2005). The functional significance of ribosomal (r) DNA variation: impacts on the evolutionary ecology of organisms. Annual Review of Ecology Evolution and Systematics 36:219-242. *https://doi.org/10.1146/annurev.ecolsys.36.102003.152620*

Wieczorek D, Żyszka-Haberecht B, Kafka A, Lipok J (2022). Determination of phosphorus compounds in plant tissues: from colourimetry to advanced instrumental analytical chemistry. Plant Methods 18:22. *https://doi.org/10.1186/s13007-022-00854-6*

Wen S, Liu B, Long S, Gao S, Liu Q, Liu T, Xu Y (2022). Low nitrogen level improves low-light tolerance in tall fescue by regulating carbon and nitrogen metabolism. Environmental and Experimental Botany 194:104749. *https://doi.org/10.1016/j.envexpbot.2021.104749*

Yang X, Lu M, Wang Y, Wang Y, Liu Z, Chen S (2021). Response mechanism of plants to drought stress. Horticulturae 7:50. *https://doi.org/10.3390/horticulturae7030050*

Zhang J, Xu A, Mu C, Wang J (2009). Occurrence and output of all types of belowground buds of *Leymus chinensis* and the dynamics of formation and maintenance of aboveground shoots. Acta Prataculturae Sinica 18:54-60.

Zheng QS, Liu ZP, Liu YL, Liu L (2004). Effects of iso-osmotic salt and water stresses on growth and ionic distribution in Aloe seedlings. Chinese Journal of Plant Ecology 28:823. *https://doi.10.17521/cjpe.2004.0107*

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