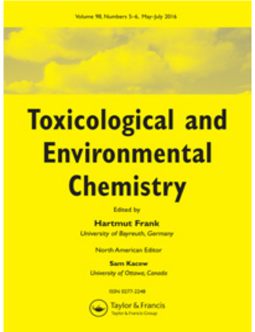

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Effects of organic matter on *Leymus chinensis* germination, growth, and urease activity and available nitrogen in coastal saline soil

Tao Zhang[a,b], Jiaqi Zhang[a], Ting Wang[a], Xiuping Tian[a], Huaina Ge[a], Yan Ma[a] and Kun Wang[b]

[a]College of Animal Science and Veterinary Medicine, Tianjin Agricultural University, Tianjin, China; [b]College of Animal Science and Technology, China Agricultural University, Beijing, China

ABSTRACT

The present study was carried out to investigate the effect of three organic matters (stalk powder, microbial fertilizer, and manure) on *Leymus chinensis* germination, growth, and urease activity and available nitrogen (N) in coastal saline soil. The study was conducted in a completely randomized design with eight treatments: $J_0V_0Y_0$, $J_1V_0Y_0$, $J_0V_1Y_0$, $J_0V_0Y_1$, $J_1V_1Y_0$, $J_1V_0Y_1$, $J_0V_1Y_1$, $J_1V_1Y_1$. The notations were based on the quantities of each agent added to 1 kg of coastal saline soil: J_0 – no straw powder, J_1 – 0.2 kg straw powder, Y_0 – no manure, Y_1 – 0.3 kg manure, V_0 – no microbial fertilizer, V_1 – 0.2 L microbial fertilizer, each in quantic repeat. *L. chinensis* was sown as 50 seeds per pot. Results indicated that addition of organic agents exerted a significantly enhanced germination, increase in fresh weight and elevated soil urease activity. Soil available N levels were significantly positively correlated with soil urease activity and fresh weight, but not with germination rate. It is noteworthy that the halophyte *L. chinensis* showed improved characteristics when grown in coastal saline soil with addition of organic amendments.

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Coastal saline soil; available nitrogen; *Leymus chinensis*; germination rate; fresh weight; organic matters

Introduction

There is 1.4×10^8 ha of severe saline abandoned and low-yielding land distributed in coastal zone of China (Wang 1993). Soil salinity that inhibits plant growth and decreases herbage yield is a serious and chronic problem for graziers. Thus, utilization of coastal saline soil resources is meaningful for sustainable agricultural development.

As an effective remediation of saline soil, plant-improved measures of saline soil focus on utilizing halophytes to improve the quality of saline-alkaline soil (Wang et al. 2011). As a saline-alkali-tolerant grass of the Eurasian Steppe, the halophyte *Leymus chinensis* is a (1) rhizomatous perennial herbage of Gramineae, and (2) has a great economic, ecological, and nutritional importance for its extensive ecological adaptability. However, germination and early seedling growth are the most critical periods for plant establishment in

CONTACT Kun Wang wangkuncaudk@163.com

coastal saline soil (Lin et al. 2014). Adaptation to coastal saline environment during seed germination and early seedling stages is crucial for plant establishment (Qu et al. 2008). More effective means of germination and early seedling processes may thus facilitate utilization of this species under salt and alkali environments (Lin et al. 2014).

Thus far, most of the studies exploring the influence of improvement measures on germination and seedling establishment of plants in coastal saline soil focused on utilizing salt-tolerant natural halophytes, but only little attention was paid to external factors such as changes in soil properties by addition of organic matter (OM). In fact, addition of OM is essential for plant to increase germination ratio and to enhance fresh weight. It is noteworthy that germination ratio and fresh weight are significant for forage production and graziers. Keeping in mind *L. chinensis* germination, growth, and properties of heavy coastal saline soil, the objectives of this study were to examine the influence of OM on *L. chinensis* germination, growth, and properties of heavy coastal saline soil, and correlation among these parameters.

Materials and methods

The soils used in this experiment were collected from the National Coastal Forage-grass Cultivational Centre (Figure 1), located at N38°50′54″, E117°10′27″ at the edge of coast of Bohai Bay. The climate is temperate continental monsoon with apparent seasonal variation. The annual mean air temperature is 11.4 °C−12.9 °C, and annual mean precipitation and evaporation are 520−660 mm and 1900 mm, respectively (Wang et al. 2012). Soil samples were freed of large organic debris, air-dried, passed through a 2-mm sieve, and then placed in 10-kg pots, 50 cm in diameter.

Some chemical and physical characters of the initial soil used in the study prior to the application of amendments were as follows: clay 46%, silt 40%, sand 14%, pH 7.66, electrical conductivity (EC) 3.25 d/Sm, Ca^{2+} 1.31 mmol/kg, Mg^{2+} 1.38 mmol/kg, Na^{+} and K^{+} 43.5 mmol/kg, OM 1.56 g/kg, sodium adsorption ratio 20.25 $[\text{meq L}^{-1}]^{0.5}$, cation exchange capacity 343.3 mmol/kg, exchangeable sodium % 38.2, total N 0.045%, and absorbable P 9.28 mg/kg.

The experiment work was conducted with eight soil treatments: $J_0V_0Y_0$, $J_1V_0Y_0$, $J_0V_1Y_0$, $J_0V_0Y_1$, $J_1V_1Y_0$, $J_1V_0Y_1$, $J_0V_1Y_1$, $J_1V_1Y_1$. Quantities of each OM added to 1 kg soil were J_0 − no straw powder, J_1 − 0.2 kg straw powder, Y_0 − no manure, Y_1 − 0.3 kg manure, V_0 − no microbial fertilizer, V_1 − 0.2 L microbial fertilizer. The amount of living microbe in the microbial fertilizer was 4.9×10^{10} colony forming units (cfu)/mL.

Figure 1. Location (A) and overview of study area (B).

Fifty healthy seeds of *L. chinensis* were sown in each pot, three replicates for each treatment. To compare the effects of these agents, the % germination and growth condition of seedling of *L. chinensis* were measured together with soil properties (soil EC and pH). *L. chinensis* was selected because of its high saline-alkali tolerance and sensitivity to changes in saline-alkali soil properties (Ju et al. 2011). *L. chinensis* seeds obtained from the National Grassland Ecosystem Observation and Research Station were first selected by floating on water, and then soaked at 25 °C in distilled water for 25 min. Seeds were then disinfected in 1% $KMnO_4$ solution for 15 min and rinsed with distilled water prior to sowing. Seedlings were germinated in a greenhouse with temperatures between 20 °C and 25 °C and relative humidity (RH) from 40% to 70%. The plants were watered daily and germinating seeds counted. Seeds were considered to have germinated when the first tow vanes expanded fully and roots were established. Germination % was calculated 31 days after sowing (Jiang et al. 2012), and fresh biomass of seedling was calculated (Jiang et al. 2012). Soil urease activity was determined with the method of indophenol blue colorimetry (Zhou 1987) and available N was determined with the alkali solution diffusion method (Bao 2011).

Data for germination %, fresh biomass of seedlings, soil urease activity, and available N were subjected to ANOVA utilizing a completely randomized design by using SPSS Statistics and Origin9 (Analytical software, 2005). The criterion for significance was set at $p < 0.05$.

Results and discussion

Addition of organic agents exerted a significant effect on germination % (Table 1). Germination % in $J_0V_0Y_0$, $J_1V_0Y_0$, $J_0V_1Y_0$, $J_0V_0Y_1$, $J_1V_1Y_0$, $J_1V_0Y_1$, $J_0V_1Y_1$, $J_1V_1Y_1$ were 40%, 75%, 60%, 87%, 84%, 80%, 85%, and 91%, respectively. The results of the present study were supported by Lv et al. (2013) who reported that organic fertilizer significantly enhanced castor bean seedlings germination %. Liu, Yang, et al. (2011) reported that straw powder and straw mulching binding structure conditioner displayed the optimal effect on increasing yield, which was related to seed %. Similarly, Wang et al. (2011) found that applying 2%−8% bio-organic fertilizers alleviated the inhibitory effects of salt-alkali stress on seed emergence, seedling growth, and leaf physiological indices of sunflower, and after 7 days under salt-

Table 1. Effects of organic agents on *L. chinensis* germination percentage, fresh weight, soil urease activity, and available nitrogen mass fraction.

Treatment	Germination percentage (%)	Fresh weight (g)	Urease activity (mg/kg)	Available nitrogen mass fraction (mg/kg)
$J_0V_0Y_0$ (CK)	40 ± 1.87^F	2.01 ± 0.30^H	30.34 ± 0.78^H	45.33 ± 1.99^F
$J_1V_0Y_0$	75 ± 1.58^D	9.54 ± 0.21^F	90.49 ± 0.84^D	118.97 ± 2.21^E
$J_0V_1Y_0$	60 ± 1.58^E	5.346 ± 0.27^G	46.44 ± 0.78^G	102.985 ± 1.98^E
$J_0V_0Y_1$	87 ± 1.59^B	9.776 ± 0.18^E	55.75 ± 0.65^F	96.745 ± 3.02^E
$J_1V_1Y_0$	84 ± 1.41^B	10.382 ± 0.15^D	94.55 ± 0.47^C	340.865 ± 2.03^B
$J_1V_0Y_1$	80 ± 1.58^C	12.532 ± 0.14^B	97.5 ± 0.93^B	264.38 ± 3.07^C
$J_0V_1Y_1$	85 ± 1.57^B	11.828 ± 0.15^C	64.37 ± 0.79^E	156.96 ± 3.97^D
$J_1V_1Y_1$	91 ± 2.00^A	13.062 ± 0.13^A	123.72 ± 0.38^A	414.92 ± 2.57^A

Note: Different letters mean significant differences at $p < 0.05$.
Each agent added to 1 kg soil: J_0 − no straw powder, J_1 − 0.2 kg straw powder, Y_0 − no manure, Y_1 − 0.3 kg manure, V_0 − no microbial fertilizer, V_1 − 0.2 kg microbial fertilizer.

alkali stress, the seedling growth in treatments 2%, 4%, and 8% bio-organic fertilizer was markedly increased. Wang, Sun, et al. (2014) also reported of seven soil improvement treatments that enhanced the growth rate of forage grass.

Addition of organic agents significantly increased fresh weight as shown in Table 1. The results of present study showed a positive correlation between addition of organic agents and fresh weight. Lu and Shen (2011) found a rise in plant height, fresh and dry weight of single plant, content of acid detergent fiber and neutral detergent fiber under similar N content with increasing organic fertilizer application. Furthermore, both dry matter yield above-ground and digestible dry matter yield rose with increasing organic fertilizer application. Lv et al. (2013) reported that both vermicompost and bacterial manure elevated the weight ratio of castor bean root, and organic fertilizer, in particular vermicompost, exerted a more positive effect on the regulation of castor bean seedlings under saline-alkali soil, and was advantageous for incubating strong castor bean seedlings. The bacterial manure produced a similar effect on seedling growth. Similarly, Li, Shen, et al. (2012) reported that organic microbe fertilizer application, especially at planting and transplanting stages, significantly promoted watermelon growth. Application of organic fertilizer significantly elevated plant height, leaf number, chlorophyll content, total uptake of N, P, and K, and quality and yield of watermelon. Liu, Cui, et al. (2011) noted that application of organic fertilizer markedly alleviated the inhibitory effect of salt on wheat seedlings treated with 150 mmol/L NaCl, and plant growth was less reduced than that of seedlings in the control (no fertilizer) under salt stress. Data confirmed the findings of the above-cited studies that fresh weight increased significantly with the addition of organic agents.

Addition of organic agents markedly elevated soil urease activity (Table 1). He et al. (1997) reported that the effect of organic fertilizer on soil urease characteristics was far more than that of signal chemical fertilizer. Song et al. (2014) observed that a close correlation existed between urease activity and soil labile N fractions content, which was enhanced by applying organic fertilizers alone. Li, Wang, et al. (2012) noted that organic fertilizer significantly increased soil urease, reduced soil pH, total salt content, and soil bulk density to some extent. There was a positive correlation between urease activity and added OM. Gu et al. (2014) reported an apparent enhancement in activity of urease by applying bio-organic fertilizer. Zhao et al. (2014) found that a long-term application of organic manure significantly increased urease, and correlation analysis showed that the activity of soil urease was positively related to OM. Perucci (1990, 1992) reported a rise of microbial biomass and urease activity after addition of organic agent such as compost vegetable residues into the soil. Mandel et al. (2007) demonstrated that organic fertilizer significantly elevated soil enzyme activity and the number of soil enzymes by enhancing soil physical properties. However, findings of this study were not in agreement with Kulandaivelu et al. (2013) who reported that urease activity was negatively correlated with sand, fine clay, bulk density, and other physical properties mutable by added agents.

Soil urease activity was significantly correlated with germination % (Table 2). Soil available N was significantly positively correlated with soil urease activity, and fresh weight (the correlation coefficients were 0.887 and 0.712, respectively), but not markedly associated with germination rate. The greater the available N mass fraction, the higher was the soil urease activity and greater *L. chinensis* fresh weight. Germination rate and fresh weight were significantly positively related; the higher the urease activity, the higher the germination rate and greater the fresh weight.

Table 2. Analysis of correlation among germination percentage, fresh weight, soil urease activity, and available nitrogen mass fraction.

	Germination %	Fresh weight	Soil urease activity	Available N mass fraction
Germination percentage	1	0.943*	0.723*	0.644
Fresh weight	0.943*	1	0.824*	0.712*
Soil urease activity	0.723*	0.824*	1	0.887*
Available nitrogen	0.644	0.712*	0.887*	1

$n = 8$, *significant at $p < 0.05$ level.

Huang et al. (2015) reported that nitrogen fertilizer application improved herbage yield from 2 tonnes/ha without fertilizer to more than 10.7 tonnes/ha with 180 kg/ha fertilizer in severe saline-sodic grassland. Zhang, Zai, et al. (2014) observed that the effects of inoculation with fungus fertilizer on the physico-chemical and biological characteristics of coastal saline soil were positive, and OM content in the soil was significantly enhanced; however, available N content increased only in June and July. Inoculation with fungus fertilizer induced significantly stimulating effects on urease activity in July and August. Zhang, Wan, et al. (2014) noted that soil OM yielded greater positive indirect path coefficients through pH and total N, and after cultivation of soil, biological activities and fertility level increased. Tejada et al. (2006) reported that added organic wastes exerted a positive effect on physical, chemical, and biological properties of the soil, soil urease activities rose significantly, % plant cover was >50% in all treated plots while only 8% in the control soil. Liang et al. (2003) found that incorporation of organic manure into soil significantly increased soil urease activity and dry weight of plant compared to the control. The incorporation of organic manure into salt soil seems to be a practical and effective way to improve soil fertility, and enhance salt tolerance and growth of crops. Wang, Cang, et al. (2014) found that a combination of organic amendments, including green waste compost, sedge peat, and furfural residue, had substantial potential for ameliorating saline soils in coastal areas and promoting plant growth.

Conclusions

Data showed that, in order to understand the effects of OM on properties of coastal saline soil used for graziery and *L. chinensis* germination, growth needs to be considered. Organic agents exerted a positive effect as shown by enhanced germination % and fresh weight of *L. chinensis*. On the other hand, with the addition of organic amendments, *L. chinensis* showed an enhanced effect on coastal saline soil. It was found that the activities of urease and available N mass fraction were associated with fresh weight and germination %, and consequently with saline tolerance of *L. chinensis*. Measuring the correlation among germination %, fresh weight, soil urease activity, and available N mass fraction may shed light on the role of OM on planting grass and coast salinity soil amendment processes involved in microbial response to multiple stressors.

Disclosure statement

No potential conflict of interest was reported by the authors.

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