Response of Enzyme Activity of Cultivated Saline Soil to Sand Covering

Jian Wang1, 2, 3, 4, *, Chenxi Yang1, 2, 3, 4 and Zhen Guo1, 2, 3, 4

1Shaanxi Provincial Land Engineering Construction Group Co., Ltd, Xi’an, China
2Institute of Land Engineering and Technology, Shaanxi Provincial Land Engineering Construction Group Co., Ltd, Xi’an, China
3Key Laboratory of Degraded and Unused Land Consolidation Engineering, the Ministry of Nature and Resources, Xi’an, China
4Shaanxi Provincial Land Consolidation Engineering Technology Research Center, Xi’an, China

*Corresponding author: jian_wang2020@shanxidichan.com

Abstract. In order to clarify the response of soil enzyme activities of cultivated saline-alkali soil to sand cover in the Loess Plateau-Mu Us Sandy ecotone. In this study, the soil enzyme activities of the saline-alkali soils before and after sand-covering (CK) and after sand-covering for three consecutive years were taken as the research objects. The results showed that the activities of four soil enzymes (urease, catalase, protease, and alkaline phosphatase) all increased to varying degrees after covering with sand. Among them, the effect of covering with sand on soil urease and alkaline phosphatase was significant. In general, the activities of the four soil enzymes are negatively correlated with soil physical properties (except porosity) and positively correlated with soil chemical properties. This study provides data support for revealing the long-term effects of sand-covering on soil enzyme activities of cultivated saline-alkali soils, and provides a reasonable idea and theoretical reference for the feasibility of using sand-covered soil organic restructuring technology in saline-alkali soil management.

Keywords: Sand-covered, saline soil, soil enzyme activity, correlation analysis.

1. Introduction

Soil salinization is one of the most difficult environmental problems in the world, which seriously restricts the sustainable development of agriculture. The total area of salinized land in the world has exceeded 1.1×10^9 hectares, accounting for about 7% of the total land area of the world [1]. However, what is more serious is that due to unreasonable farming and management, the area of global salinized land is increasing at a rate of 1% to 2% per year [2]. In the future, global food security will increase. It is a serious problem. Expanding arable land space is one of the important ways to ensure food security.

Soil enzyme activity is closely related to soil properties, soil types and environmental conditions, and is currently widely used as an important indicator of soil quality and soil biological activity [3-4]. Among them, urease, alkaline phosphatase, catalase and protease activities are more sensitive to environmental changes. Urease promotes the hydrolysis of nitrogen-containing organic matter, and is
closely related to the formation and formation of inorganic nitrogen in the soil [5]. Urine enzyme activity is sensitive to high salt and high salt stress [6]. This shows that urease activity can be used as an indicator of soil quality. Phosphorus in soil mainly exists in organic form. Alkaline phosphatase is the main enzyme involved in the phosphorus cycle, because it can convert organic phosphorus into inorganic phosphorus, which is a nutrient available for plants [7]. Alkaline phosphatase is sensitive to the external environment and is an indicator of soil organic phosphorus mineralization and biological activity [16-17]. Catalase can decompose peroxide produced in the metabolic process, thereby preventing its toxic effects on organisms [8]. These enzymes play an important role in the cycle of soil carbon, nitrogen, and phosphorus, participate in a large number of soil biochemical processes, and directly participate in various biochemical reactions in the soil [9]. However, the current researches on soil enzyme activity are mostly concentrated on cultivated soil and grassland soil, and there are few studies on the distribution of soil enzyme activity and influencing factors after the change of saline soil enzyme activity to sand.

We conducted an experimental study on the change characteristics of soil enzyme activity over time in the saline-alkali land under sand-covered conditions in the Loess Plateau-Mu Us Sandy Farming and Pastoral Zone, and used correlation analysis methods to study the relationship between soil enzyme activity and soil physical and chemical properties. It reveals the influence of sand-covering on soil enzyme activity in saline-alkali land, and the soil environmental factors that affect the change and distribution of soil enzyme activity under sand-covering conditions, and provides theoretical basis for the sustainable development and utilization of sand-to-saline-alkali technology.

2. Materials and Method

2.1. Sampling Design
The study area is located in Wangtanzi Village (108°20′E, 37°54′N), Duiziliang Town, Dingbian County (Figure 1). The terrain is flat and the soil is uniform. The sand reform was implemented in 2015, that is, about 30 cm of sand was covered on the surface of the saline-alkali soil, and the soil was shallowly mixed to loosen the cultivated layer. It was idle that year, and began farming on April 30, 2016. The planted crop is corn, which is one season per year. 6 quadrats (10m × 10m) were set up in the study area, with a distance of about 15 meters. Respectively in 2015 (before the sand reform, CK), 2016 (one year after the sand reform, T1), 2017 (two years after the sand reform, T2), 2018 (three years after the sand reform, T3). After the corn was harvested in late September, a five-point sampling method was used to collect soil samples of the cultivated layer at the same location of the test field, and mixed them as one sample, totaling 18 samples. Take some fresh soil samples for the determination of available nitrogen and soil enzyme activity; after the remaining soil samples are air-dried, pass through a 2mm sieve for the determination of soil physical and chemical properties.

2.2. Soil Analysis
The soil pH value is determined by the water-soil ratio 2.5:1 water leaching method, the soluble salt content is determined by the drying method; the bulk density and total porosity are determined by the ring knife method; the soil organic carbon is determined by the combustion oxidation-non-dispersive infrared method; Nitrogen is determined by semi-micro Kjeldahl method; available nitrogen is determined by alkaline solution diffusion method; available phosphorus is determined by NaHCO₃ extraction-molybdenum antimony colorimetric method; available potassium is determined by ammonium acid extraction-atomic absorption spectrophotometry [10].

The soil enzyme activity is measured by a micro-method under a microplate reader, and the enzyme activity is calculated based on the absorption peak of the product produced per gram of soil per hour at a specific wavelength. The indophenol blue colorimetric method was used to determine the urease activity. The NH₄-N produced by the hydrolysis of urea was measured at a wavelength of 578nm. The alkaline phosphatase activity was measured by the phenyl disodium phosphate colorimetric method, and the absorbance of the product phenol was measured at a wavelength of
660nm. The catalase activity was measured by potassium permanganate titration, and the absorbance of hydrogen peroxide was measured at a wavelength of 240nm. The ninhydrin colorimetric method was used to determine the protease activity, and the absorbance of the product pertyrosine was determined at a wavelength of 680nm [11].

2.3. Statistical Analysis
Microsoft Excel 2013 software was used to organize the data. All experimental data were tested for normal distribution and homogeneity of variance by Shapiro-Wilk and Levene's tests. Secondly, ANOVA was used to test the difference in soil enzyme activity between different years, and Duncan's multiple comparison method was used to compare the difference in soil enzyme activity between different years. Finally, the R language "PerformanceAnalytics", "corrplot" and "agricolae" packages are used for Pearson correlation analysis and path analysis between soil physical and chemical properties [12].

3. Results and analysis

3.1. Analysis of differences in soil enzyme activities
It can be seen from Fig.1 that sand covering significantly affected soil urease and alkaline phosphatase activities ($P_{\text{urease}} < 0.05$, $P_{\text{alkaline phosphatase}} < 0.05$). After being covered with sand, with the extension of farming years, soil urease activity increased by 42.26%, 42.26% and 53.61%, and alkaline phosphatase activity increased by 23.17%, 31.71% and 31.71%, respectively. Soil catalase and protease activity after sand reformation showed an increasing trend with the extension of farming years, but did not reach a statistically significant level ($P_{\text{catalase}} > 0.05$, $P_{\text{protease}} > 0.05$).

Figure 1. Effect of Sand Covering on Soil Enzyme Activity. Different lowercase letters represent significant differences between groups.

3.2. Cross section comparison of Rehmannia glutinosa
The correlation analysis between soil enzyme activity and soil physical and chemical properties (pH, salinity, bulk density, porosity, alkalinity, organic matter, total nitrogen, available nitrogen, available phosphorus and available potassium) is shown in Fig.2. The data for correlation analysis is based on all processing and all levels. Urease has a significant negative correlation with soil pH and bulk density, and a significant positive correlation with soil available potassium; catalase has a significant...
negative correlation with salt content, and a significant positive correlation with available nitrogen; protease is related to pH, bulk density, and alkalinity. It is significantly positively correlated with organic matter; alkaline phosphatase is significantly negatively correlated with pH, bulk density, and alkalinity, and is significantly positively correlated with total nitrogen, available phosphorus and available potassium. In addition, the study also shows that the activities of urease, alkaline phosphatase, protease and catalase are all positively correlated, indicating that any kind of soil enzyme activity can reflect the other two soil enzyme activities to a large extent.

**Figure 2.** Pearson correlation analysis between enzyme activities and soil properties. Degree of alkalinity, DOA; Salt content, SC; Porosity, POR; Soil bulk density, SBD; Soil organic matter, SOM; Total nitrogen, TN; Available nitrogen, AN; Available phosphorus, AP; Available potassium, AK; Urease, UE; Catalase, CAT; Protease, PRO; Alkaline phosphatase, ALP. *, P < 0.05; **, P < 0.01; ***, P < 0.001.

### 4. Discussion and conclusion

Soil enzymes are biocatalysts that produce specific biochemical reactions in the soil. They always participate in the biochemical cycle of the soil, the conversion process of organic matter and minerals, and their activity has a significant effect on soil metabolism, the conversion of soil nutrient forms, and the improvement of soil fertility. Significant significance [13]. Compared with soil organic matter, after land consolidation, soil enzyme activity is often used as an indicator of soil fertility changes due to its fast, sensitive, and convenient detection characteristics [14]. In recent years, there have been many research reports on soil enzyme activity, most of which are concentrated on soil urease, phosphatase, invertase activity and soil types [8], land use methods, fertilization measures, soil temperature. In terms of soil moisture, there are few reports on the dynamic changes of enzyme activity after sand covering and remediation of saline-alkali land [15-16].
In this study, we measured four enzyme activities related to soil nutrient cycling, namely urease, catalase, protease and alkaline phosphatase. After sand-covering and remediation, soil urease, catalase, protease and alkaline phosphatase were all improved to varying degrees than before remediation, which is similar to the results of Lv [17]. With the extension of cultivation years, the activities of the four soil enzymes have increased year by year, and there is a certain degree of positive correlation between the activities of the four soil enzymes, which was also confirmed by Lv [17]. This may be due to the low salt content and loose structure of sandy soil. After being mixed with saline-alkali soil, the capillary pores of the cultivated layer become larger and the siphon effect is weakened, which effectively inhibits the salt content of groundwater from reaching the cultivated layer [18]. Sand covering may also improve the physical properties of the soil, leading to changes in the number and structure of soil microorganisms on the surface of the soil, resulting in increased microbial activity and increased soil enzyme secretion by microorganisms [19].

To sum up, the activities of four soil enzymes (urease, catalase, protease, alkaline phosphatase) all increased to varying degrees after covering with sand. Among them, the impact of sand covering on soil urease and alkaline phosphatase reached In general, the activity of four soil enzymes is negatively correlated with soil physical properties (except porosity), and positively correlated with soil chemical properties. In the context of the long-term search for the utilization of saline-alkali land and the development of sustainable agriculture in the Loess Plateau-Mu Us Sandy Interlock, this study provides data support for revealing the long-term effects of sand-covering on soil enzyme activities and farmland soil fertility, and for studying sand-covering Long-term feasibility provides reasonable ideas.

References
[1] Wicke B. Bioenergy Production on Degraded and Marginal Land. Utrecht University, Nederland, 2011.
[2] Munns R, Tester M. Mechanisms of salinity tolerance. Annu. Rev. Plant Biol. 2008, 59: 651-681.
[3] Melero, S, Madejion, E, Ruiz, J C, Herencia, J F. Chemical and biochemical properties of a clay soil under dryland agriculture system as affected by organic fertilization. Eur. J. Agron. 2007, 26, 327-334.
[4] Yuan B C, Li Z Z, Liu H, Gao M, Zhang Y Y. Microbial biomass and activity in salt affected soils under arid conditions. Appl. Soil Ecol, 2007, 35: 319-328.
[5] Liang Y C, Yang Y F, Yang C G, Shen Q R, Zhou J M, Yang L Z. Soil enzymatic activity and growth of rice and barley as influenced by organic manure in an anthropogenic soil. Geoderma, 2003, 115: 149-160.
[6] Zhang T, Wan S, Kang Y, Feng H. Urease activity and its relationships to soil physiochemical properties in a highly saline-sodic soil. J. Soil Sci. Plant Nutr, 2014, 14 (2), 302-313.
[7] Wu, Y Y , Liu, R C , Zhao, Y G , Li, P P , Liu, C Q. Spatial and seasonal variation of salt ions under the influence of halophytes, in a coastal flat in eastern china. Environ. Geol. 2009, 57, 1501-1508.
[8] Krämer S, Green D M. Acid and alkaline phosphatase dynamics and their relationship to soil microclimate in a semiarid woodland. Soil Biol. Biochem, 2000, 32: 179-188.
[9] Yang, L., Bian, X.G., Yang, R.P., Zhou, C.l., Tang, B.P. Assessment of organic amendments for improving coastal saline soil. Land Degrad. Dev. 2018, 29, 3204-3211.
[10] Liu G M, Zhang X C, Wang X P, Shao H B, Yang J S, Wang X P. Soil enzymes as indicators of saline soil fertility under various soil amendments. Agriculture, Ecosystems & Environment, 2017, 237: 274-279.
[11] Demisie W, Liu Z Y, Zhang M K. Effect of biochar on carbon fractions and enzyme activity of red soil. Catena, 2014, 121: 214-221.
[12] Wang X Q, Xing X, Zhang F J, Xin K. Biological improvement of saline alkali soil reference
system: A review. Sciences in Cold and Arid Regions, 2018, 10 (6): 0516-0521.

[13] Wang, Y.G., Xiao, D.N., Li, Y., Li, X.Y. Soil salinity evolution and its relationship with dynamics of groundwater in the oasis of inland river basins: case study from the Fubei region of Xinjiang Province, China. Environ. Monit. Asses. 2008, 140, 291-302.

[14] Shi, Z., Li, Y., Wang, R., Makeschine, F. Assessment of temporal and spatial variability of soil salinity in a coastal saline field. Environ. Geol. 2005, 48, 171-178.

[15] Rady, M.M. Effect of 24-epibrassinolide on growth yield, antioxidant system and cadmium content of bean (Phaseolus vulgaris L.) plants under salinity and cadmium stress. Sci. Hort. 2011, 129, 232-237.

[16] Saha, S., Prakash, V., Kundu, S., Kumar, N., Mina, B.L. Soil enzymatic activity as affected by long term application of farm yard manure and mineral fertilizer under a ainfed soybean–wheat system in N-W Himalaya. Eur. J. Soil Biol. 2008, 44, 309-315.

[17] Lv, Z.Z., Liu, G.M., Yang, J.S., Zhang, M.M., He, L.D., Shao, H.B., et al. Spatial variability of soil salinity in Bohai Sea coastal wetlands, China: partition into four management zones. Plant Biosyst. 2013, 147, 1201-1210.

[18] Zhao, Y.G., Pang, H.C., Wang, J., Huo, L., Li, Y.Y. Effects of straw mulch and buried straw on soil moisture and salinity in relation to sunflower growth and yield. Field Crop. Res. 2014, 161, 16-25.

[19] Ren, J., Chen, J., Han, L., Wang, M., Yang, B., Du, P., Li, F. Spatial distribution of heavy metals, salinity and alkalinity in soils around bauxite residue disposal area. Sci. Total Environ. 2018, 628, 1200-1208.