Effects of Trehalose on Physiological Characteristics of Waxy Maize under Salt Stress

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Abstract: Soil salinization is a global problem and an important factor affecting agricultural production and ecological environment. The salinized land area accounts for about 65.8% of the total land area in Tianjin. The maize variety "Jinnuo 72" was used as material, and pot culture method was adopted to explore the effects of trehalose on physiological characteristics of waxy maize under 0.04%, 0.20%, 0.35%, 0.53% and 0.68% soil salt content. The results showed that the treatment group compared with control group SOD activity significantly increased by 4.81%, 5.83%, 7.03% and 10.28% at 0.04%, 0.20%, 0.35% and 0.52% salt concentration, respectively. POD activity in treatment group compared with control group was significantly increased by 30.86% at 0.35% salt concentration. Under five different salt concentration, CAT activity significantly increased by 6.11%, 5.81%, 7.22%, 13.98% and 14.70%, respectively. MDA content in treatment group was significantly decreased compared with control group by 55.40%, 67.39%, 51.31% and 29.67% at 0.20%, 0.35%, 0.52% and 0.68% salt, respectively. The soluble sugar treatment group significantly increased compared with the control group at five salt concentration. The proline and soluble protein significantly increased compared with the control group at 0.20%, 0.35% and 0.52% salt concentration. Results showed that trehalose could increase the activity of antioxidant enzymes, effectively reduce the peroxidation of lipid membrane and regulate the content of osmotic substances in waxy maize seedlings, so as to improve the salt resistance of waxy maize seedlings.

1. Introduction

Soil salinization, as an abiotic stress, is one of the important factors affecting agricultural production and ecological environment and has become a global problem. According to the data, salinized land occupies more than 3 600×10^4 million hm^2, and its occupied area shows a trend of continuous expansion [1]. Salinization land area accounts for about 65.8% of the total land area in Tianjin [2]. Long-term accumulation of salt in soil will cause salt damage to crops, which could be divided into primary salt damage and secondary salt damage according to different ways of salt ion action [3]. Soil salinization is an important factor affecting crop production, which restricts the development of China agricultural level to a certain extent. It is of great historical significance to study and breed salt-tolerant crop varieties.

Waxy maize is a subspecies of Maize genus, also known as waxy maize because of its dull and waxy grain appearance [4]. It is sticky, waxy, fragrant, fine and soft, rich in starch, water-soluble polysaccharide and a variety of vitamins, rich in nutrients, and higher in protein, lysine and tryptophan compared with ordinary maize [5]. The salt tolerance of maize is at medium level and it is very sensitive to salt concentration. When salt concentration exceeds 0.0017mol/L NaCl, physiological metabolism of maize is inhibited, crop yield is affected, and plant death is seriously

caused [6-9]. Under the condition of high salt concentration, plants regulate high salt stress by means of salt expelling, dilute salt and salt rejection and maintain their own growth and development [10]. It has important nutritional and commercial value to study the salt-tolerant mechanism of waxy maize and to select high yield and good quality crop varieties.

Trehalose, as a non-reducing disaccharide molecule widely existing in plants, plays an important role in effectively maintaining osmotic pressure, protective membrane structure and cleaning reactive oxygen species in plants under abiotic stress such as drought, salt stress and temperature [11-13]. Trehalose accumulation could protect crops in response to salt stress. Exogenous trehalose could effectively improve the antioxidant enzyme activity of melon seedlings under salt stress [14], significantly improve the photosynthesis of tomato leaves [15], promote the accumulation of effective components in licorice seedlings and alleviate the damage of salt injury to licorice [16]. There have been previous studies on maize, but there are few reports on the mechanism of exogenous trehalose to alleviate salt stress on waxy maize. This paper studied the changes of physiological characteristics of waxy maize seedlings under salt stress by applying trehalose exogenously to the maize variety "Jinnuo 72", and provided theoretical and technical basis for the rational cultivation of waxy maize.

2. Materials and Methods

2.1 Experimental Materials

The variety of maize is Jinnuo 72, provided by Tianjin Zhongtian Runnong Technology Co., Ltd.

2.2 Experimental Method

The maize seeds with 1% sodium hypochlorite disinfection for 10min, distilled water rinse clean. The seeds of control group and treatment group were soaked with distilled water and 1mmol·L^{-1} trehalose for 12h, respectively. Seeds of the two treatments were sown in different pots with salt content in soil as substrate, and 10 holes were sown evenly in each pot. Soil concentrations with different salt contents were 0.04%, 0.20%, 0.35%, 0.53% and 0.68%, respectively, which were adjusted with 0, 40, 80, 120 and 160mmol·L^{-1} NaCl solutions, respectively. Each treatment was repeated three times. The activities of SOD, POD and CAT, the contents of MDA, proline, soluble sugar and soluble protein in the leaves were determined when the seedlings grew to 10cm.

2.3 Determine items and Measurement Methods

The activity of superoxide dismutase (SOD) was determined by hydroxylamine method. The activity of peroxidase (POD) was determined by guaiacol method. Catalase (CAT) activity was determined by visible light method. The content of malondialdehyde (MDA) was determined by TBA method. Soluble protein content was determined by Coomassie bright blue G-250 staining method. The content of soluble sugar was determined by anthrone colorimetry. The content of proline was determined by colorimetric method. (Kit: Nanjing Jiancheng Reagent Company).

2.4 Data Processing

Data processing and draw graphs were performed by using Excel 2013, and data statistical analysis was used by SPSS 18 software.

3. Results and Analysis

3.1 Effects of Trehalose on Activities of SOD, POD and CAT in Waxy Maize Seedlings under Different Salt Stress

SOD, POD and CAT could reflect the physiological and metabolic status in plants as well as the regulation and adaptability to the external environment. As can be seen from below figures, the activities of SOD, POD and CAT in the control group gradually decreased with the increase of salt content concentration. As can be seen from Fig 1, SOD activity in the treatment group was

significantly increased by 4.81%, 5.83%, 7.03% and 10.28% compared with the control group when salt content was 0.04%, 0.20%, 0.35% and 0.52%, respectively. When the salt content was 0.68%, compared with the control group, there was an increasing trend with an increase of 1.62%, but it was not significantly. As can be seen from Fig 2 and Fig 3, POD and CAT activities in the treatment group were increased compared with the control group under five different salt concentration conditions. POD activities increased by 9.04%, 14.27%, 30.86%, 16.46% and 18.98%, respectively. When salt content was 0.35%, Compared with the control group, the POD activity of treatment group showed the largest increasing trend. CAT activity significantly increased by 6.11%, 5.81%, 7.22%, 13.98% and 14.70%, respectively. It could be seen that under different salt concentration conditions, the activities of SOD, POD and CAT in the treatment group all increased, indicating that trehalose could improve the activity of antioxidant enzymes, which is beneficial to the physiological metabolic response of waxy maize and improve salt tolerance.

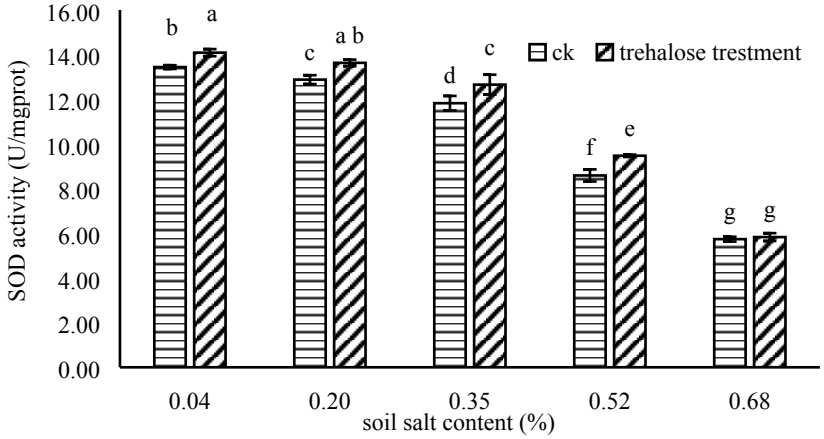

Fig 1. SOD activity in maize leaves under different NaCl concentrations

Different lower-case letters above the bar mean significant difference at the 0.05 probability leve1, the same below.

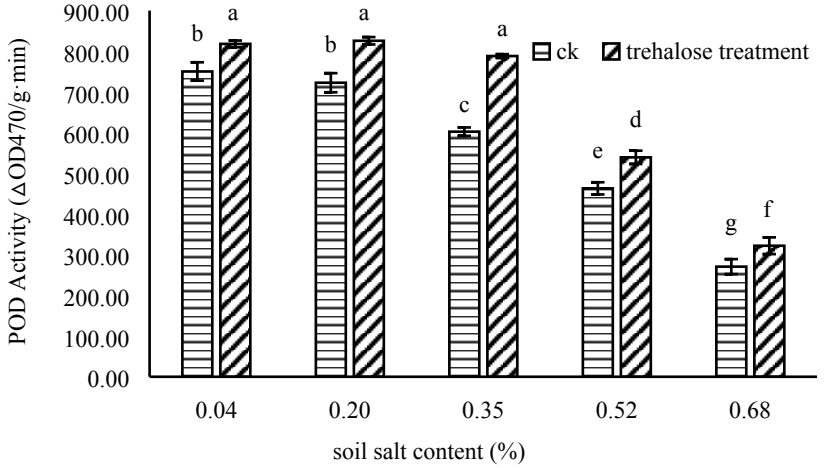

Fig 2. POD activity in maize leaves under different NaCl concentrations

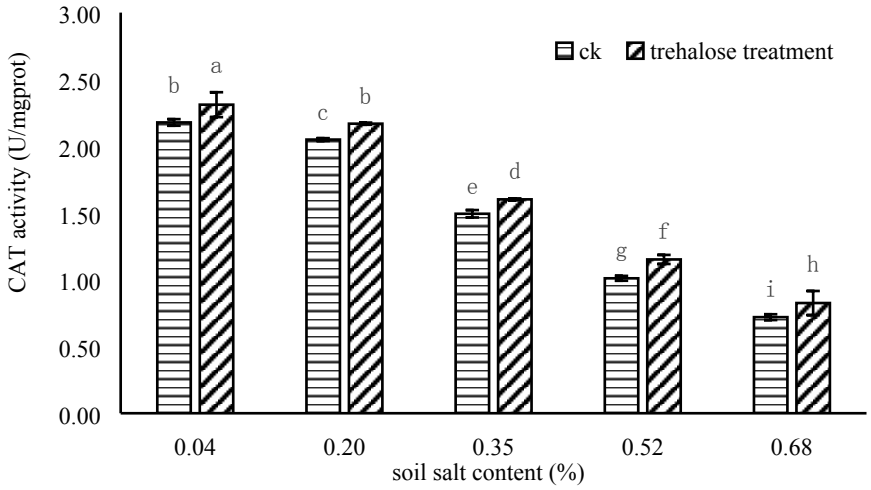

Fig 3. CAT activity in maize leaves under different NaCl concentrations

3.2 Effects of Trehalose on MDA Content in Waxy Maize Seedlings under Different Salt Stress

As can be seen from Fig. 4, the MDA content of waxy maize seedlings in the treatment group and the control group increased significantly with the increase of salt content. When the salt content was 0.20%, 0.35%, 0.52% and 0.68%, the MDA content in the treatment group was significantly lower than that in the control group. The content of MDA decreased by 55.40%, 67.39%, 51.31% and 29.67%, respectively. Under the concentration condition of salt content of 0.35%, MDA content decreased most significantly. Results showed that the content of MDA in waxy maize seedlings was reduced by exogenous trehalose, which prevent lipid membrane peroxidation and reduce salt stress damage.

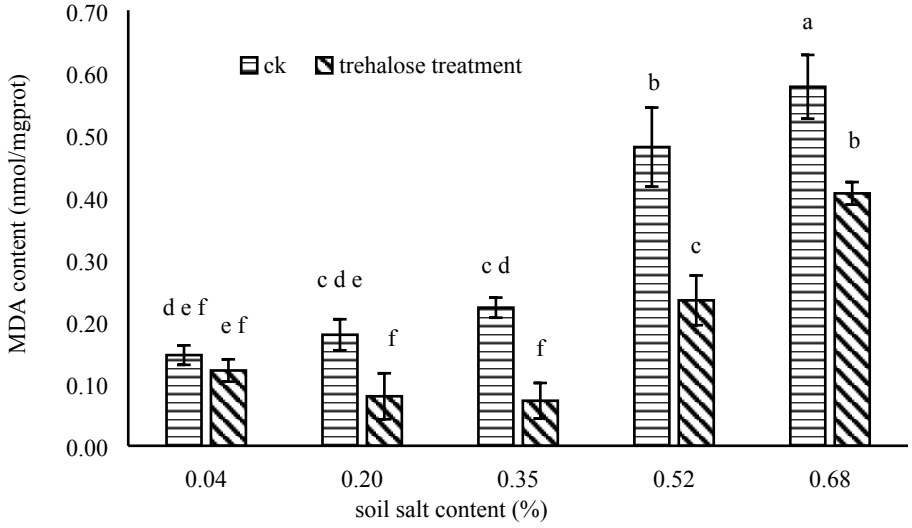

Fig 4. MDA content in maize leaves under different NaCl concentrations

3.3 Effects of Trehalose on Contents of Proline, Soluble Sugar and Soluble Protein in Waxy Maize Seedlings under Different Salt Stress

As shown in the below figures, compared with the control group, the contents of soluble sugar in the treatment group significantly increased under different salt stress conditions, the contents of proline and soluble protein in the treatment group significantly increased under 0.20%, 0.35%, 0.52% salt stress conditions. In the low salt concentration condition of 0.04%, the proline content was decreased by 5.12% compared with the control group, but not significantly (Fig.5). Compared with the control group, the content of soluble sugar in the treatment group was significantly increased by 10.12%, 9.16%, 16.58%, 6.88% and 11.65%, respectively, under different salt

concentrations (Fig.6). Compared with the control group, the soluble protein content in treatment group was significantly increased by 22.69%, 36.83% and 26.19% at salt concentrations of 0.20%, 0.35% and 0.52, respectively. The soluble protein in the treatment group was increased compared with the control group under 0.04% and 0.68% salt content, but it was not significantly (Fig.7). Results showed that exogenous trehalose could improve the salt resistance of waxy maize from plant osmotic regulation.

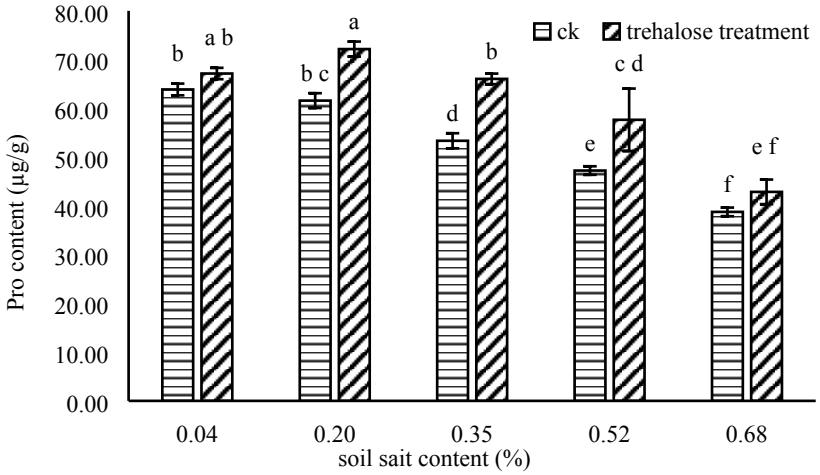

Fig 5. Proline content in maize leaves under different NaCl concentrations

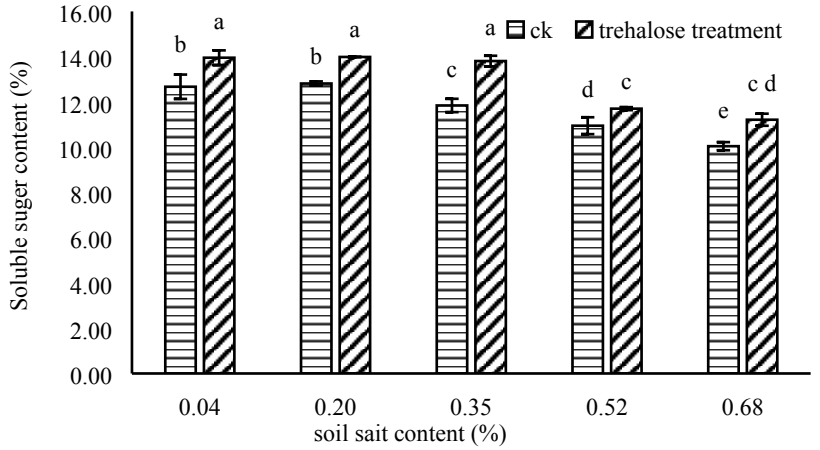

Fig 6. Soluble sugar content in maize leaves under different NaCl concentrations

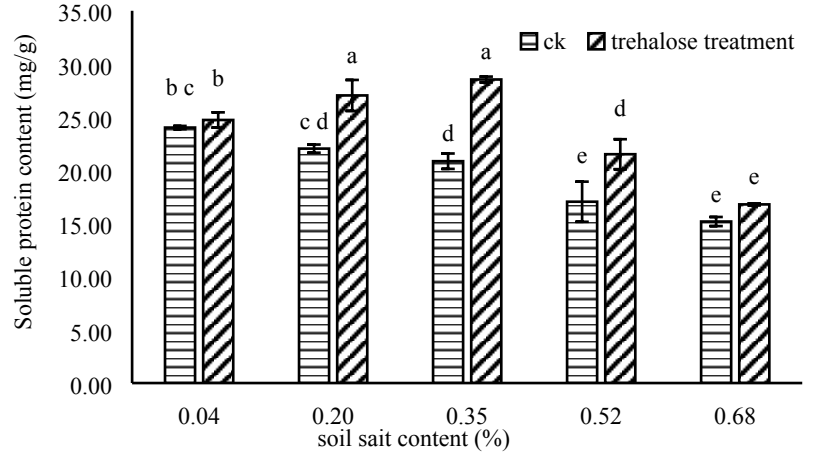

Fig 7. Soluble protein content in maize leaves under different NaCl concentrations

4. Discussion and Conclusions

Studies on the direct damage of plant cell membrane caused by salt stress have proved that the change of membrane physical state of lipid membrane is the reason for the initial response of plants to osmotic stress [17]. In order to prevent injury of reactive oxygen species (ROS) to cell membrane, SOD, POD and CAT are involved in the scavenging of ROS in plants to maintain a low level of ROS in plants and inhibit its accumulation in plants [18]. In this study, the activities of SOD, POD and CAT in the control group of waxy maize seedlings decreased with the increase of soil salt content. Exogenous trehalose was applied to effectively improve the scavenging capacity of reactive oxygen species in the plant, significantly alleviate the inhibition effect of salt stress on the activities of these three enzymes, and the activities of SOD, POD and CAT were maintained at a high level.

MDA content is an important index to measure the degree of membrane lipid damage, and the higher the content, the greater the damage to the membrane [19, 20]. Many studies have shown that under salt stress, exogenous application of appropriate concentration of trehalose can significantly reduce MDA content in rice [21] and wheat [22] seedling, which is consistent with the trend of this study. Salt stress results in the waxy maize seedling MDA content, membrane relative permeability increases and the biological membrane damage. Applying appropriate concentration of trehalose could effectively reduce the cytoplasmic membrane permeability and inhibit the accumulation of MDA. It may be due to the strong dewatering effect of trehalose [23], could protect the integrity of the biofilm structure under salt stress, so as to alleviate the damage of ROS to membrane lipids.

Proline, soluble sugar and soluble protein as osmotic regulation substances increased significantly under salt stress. Showed that waxy maize seedling damage to the cell membrane system under salt stress, cell by increasing the content of osmoregulation substance to maintain low osmotic potential, to prevent the loss of too much water in the cells, the results are consistent with the research results of Wang Peng et al. [24]. The accumulation of osmotic regulatory substances is conducive to improving the water-holding and water-absorbing capacity of cells [25]. Exogenous trehalose enhances the salt resistance of plants, alleviating the physiological development inhibition of plants subjected to salt injury [26].

In conclusion, under different salt concentration conditions, exogenous trehalose treatment of waxy maize seedling, SOD, POD and CAT as antioxidant enzymes, the activities of these enzymes significantly increased, and MDA content decreased. The contents of proline, soluble sugar and soluble protease as osmotic regulatory substances were increased. All of them effectively alleviated the damage caused by salt stress on waxy maize seedlings, improved the physiological metabolic response of plants, protected the stability and safety of biofilm system, maintained the osmotic potential of leaves, and provided favourable external environmental conditions for the growth of waxy maize.

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