

## Effects of Exogenous Trehalose on Photosynthetic Characteristics of Waxy Maize Seedlings under Salt Stress

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**Abstract:** Salt stress is one of the important factors influence the quality and yield of waxy maize. Taking Jinnuo 72 as the research material, the seeds were treated with trehalose solution of 1 mmol·L<sup>-1</sup>. The pot experiment was carried out to set five levels of soil with salt content of 0.04%, 0.20%, 0.35%, 0.52% and 0.68%. The photosynthetic characteristics were determined of waxy maize seedlings. The results showed that the net photosynthetic rate could be significantly increase by exogenous trehalose treatment of waxy maize seedlings. The transpiration rate, stomatal conductance and chlorophyll content could be significantly increase by exogenous trehalose of waxy maize seedlings at 0.20% and 0.35% salt concentration. The intercellular CO<sub>2</sub> concentration could be significantly reduced by exogenous trehalose treatment of waxy maize seedlings at 0.35% salt concentration. The nitrogen content could be significantly increase by exogenous trehalose treatment of waxy maize seedlings at 0.04%, 0.20% and 0.35% salt concentration. The photosynthetic characteristics could be change by exogenous trehalose of waxy maize seedlings under salt stress, thus, the photosynthetic, nitrogen and chlorophyll contents were improved of seedlings under salt stress. Salt tolerance could be improved by using trehalose treated waxy maize seeds and seedlings.

### 1. Introduction

Soil salinization is an abiotic stress that seriously affects crop growth [1]. The salinized soil is mainly distributed in the coastal areas in China, and due to the unscientific application of chemical fertilizers by agricultural producers and the lack of good agricultural irrigation measures, soil salinization has a tendency to expand [2-4]. Many studies have shown that salt stress can lead to a decrease in crop photosynthetic rate, ion toxicity, stomatal closure and insufficient CO<sub>2</sub> supply, etc., which may lead to crop yield reduction or death [5-7]. Waxy maize, which originated in China, is a variant of ordinary maize. Waxy maize contains more vitamins than ordinary maize, so it is called the "gold crop". At present, waxy maize is widely planted in China. It is an important raw material for the wine and also be used to livestock feed. Waxy maize is a kind of crop that is not salt-tolerant. When the soil salt content reaches a certain value, it will affect the development or death of waxy maize.

Trehalose is a kind of natural oligosaccharide with nonreducing properties and stable structure, which is widely used. Trehalose has many excellent characteristics, such as drought resistance, cold resistance, heat resistance, water retention, moisture retention, stability, etc., which can help plants protect cell membrane and protein structure under high temperature, cold, drought, high osmotic pressure and other adversities, avoid membrane structure denaturation, and enable organisms to maintain normal life activities [8-9]. At present, a large number of studies had shown that crop resistance could be improved to low temperature, drought and salt injury by exogenous trehalose

[10-12]. Zhang Nishang et al. found that the photosynthesis could proceed normally of apple leaves use trehalose under drought stress and protected photosynthesis mechanism [13]. Some foreign scholars had found that introducing trehalose synthase gene can improve drought, cold and salt tolerance of plants [14]. Wang Di et al. showed that the activity of antioxidant enzymes can be improved by spraying trehalose on wheat seedlings under high temperature stress, removed the oxidative stress caused by high temperature, and enable the normal growth of wheat seedlings [15]. Tian Lixin et al. showed that trehalose could alleviate the damage of salt stress to maize seedlings and enhance the resistance of maize seedlings to salt stress [6]. However, the effect of trehalose treatment on the photosynthetic characteristics of waxy maize seedlings under salt stress is seldom reported. Therefore, the purpose of this study is to explore the effects of exogenous trehalose on the changes of photosynthetic characteristics of waxy maize seedlings under salt stress, and to provide theoretical and technical basis for the rational cultivation of waxy maize under salt stress.

## **2. Materials and Methods**

### **2.1 Experimental Materials**

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

### **2.2 Experimental Method**

Adopt pot experiment: set control group and treatment group. The evenly selected waxy maize seeds were soaked in 1% sodium hypochlorite solution for disinfection for 10 min, then rinsed with clean water. The seeds were soaked in water (control group) and trehalose solution (treatment group) for 12 h, respectively. Soil substrates were treated with 0, 40, 80, 120 and 160 mmol·L<sup>-1</sup> NaCl solutions, so that the soil salt content was 0.04%, 0.20%, 0.35%, 0.52% and 0.68%, respectively. Two kinds of seeds were planted in five different salt concentrations pots, respectively. Repeat 3 times. Sowing 10 seeds evenly in each pot. When the seedlings height was about 5 cm, each pot of the treatment group was sprayed with another 5 ml trehalose. The net photosynthetic rate, transpiration rate, stomatal conductance, intercellular CO<sub>2</sub> concentration, chlorophyll content and nitrogen content in the seedlings were measured about 10 cm of seedlings height.

### **2.3 Determine items and Measurement Methods**

The net photosynthetic rate, transpiration rate, stomatal conductance and intercellular CO<sub>2</sub> concentration were measured using CI-340 ultralight portable photosynthesis detector (CI-340 Hd-held system (USA CID, Inc.)). Which were measured in the middle of leaf from 11:00 to 12:00 a.m. Chlorophyll content (SPAD) and nitrogen content (mg/g) were determined by plant nutrition analyser (TYS-3N).

### **2.4 Data Processing**

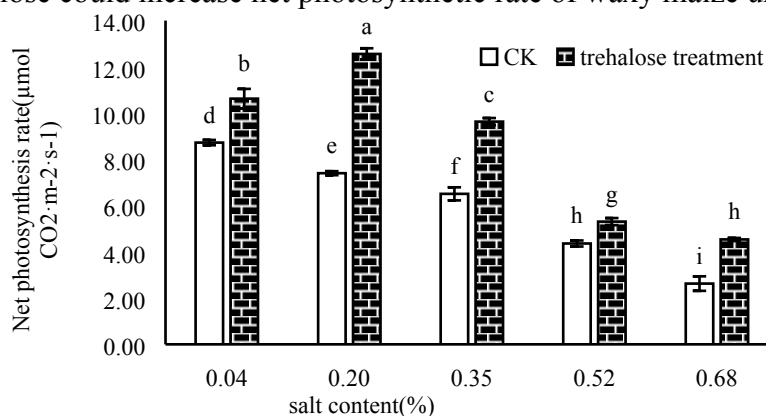
Excel 2012 was used to analyse the data and make diagrams, and DPS was used for statistical analysis of the data.

## **3. Results and Analysis**

### **3.1 Effect of Trehalose on Net Photosynthetic Rate of Waxy Maize Seedlings under Salt Stress**

Net photosynthetic rate is the amount of carbon dioxide actually absorbed by a plant in the process of photosynthesis minus the amount of carbon dioxide produced by its own respiration, which could directly represent the strength of photosynthesis of a plant. As can be seen from figure 1, with the increase of soil salt content, the net photosynthetic rate of waxy maize seedlings in the control group showed a downward trend, and the net photosynthetic rate in the treatment group increased significantly compared with the control group. Especially, net photosynthetic rate increased by 69.73% and 72.62% at 0.20% and 0.68% salt concentration, respectively. The results

indicated that trehalose could increase net photosynthetic rate of waxy maize under salt stress.

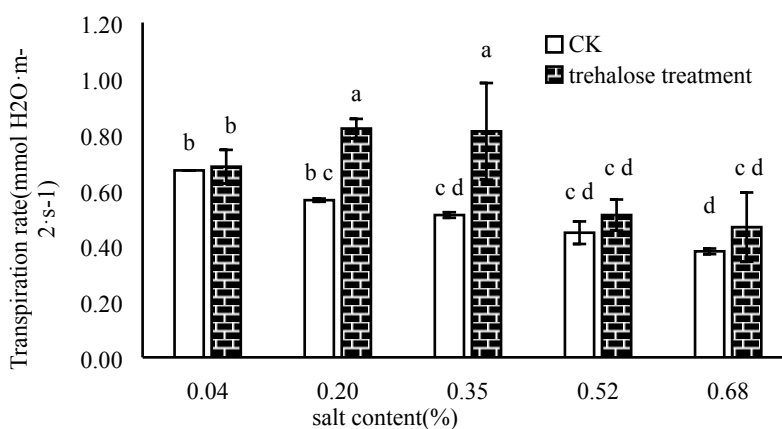


**Fig 1.** Changes of net photosynthetic rate of waxy maize seedlings treated with trehalose under different soil salt

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

### 3.2 Effect of Trehalose on Transpiration Rate of Waxy Maize Seedlings under Salt Stress

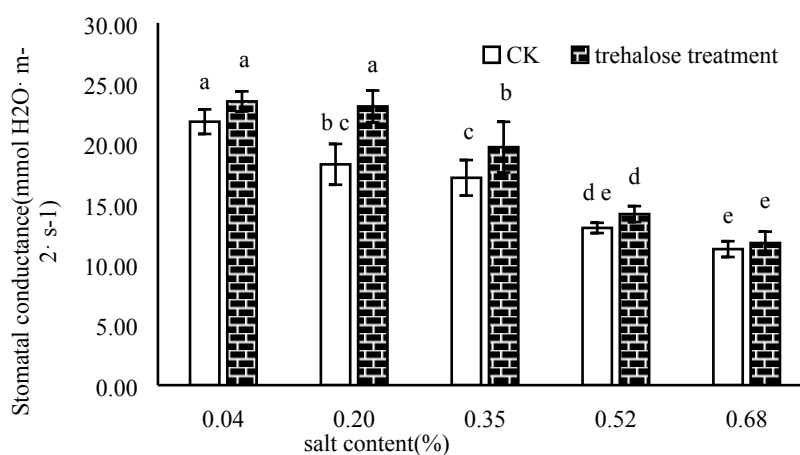
Transpiration rate is the amount of water lost per unit area of plant leaves in a period of time. As can be seen from figure 2, compared with the control group, transpiration rate of waxy maize seedlings in treatment group was significantly increased by 46.43% and 58.82% at 0.20% and 0.35% salt concentration, respectively. However, treatment group had no significant effect under 0.52% and 0.68% salt concentration. The results showed that transpiration rate could be significantly increased by exogenous trehalose treatment of waxy maize seedlings under the condition of low salt concentration.



**Fig 2.** Changes of transpiration rate of waxy maize seedlings treated with trehalose under different soil salt

### 3.3 Effect of Trehalose on Stomatal Conductance of Waxy Maize Seedlings under Salt Stress

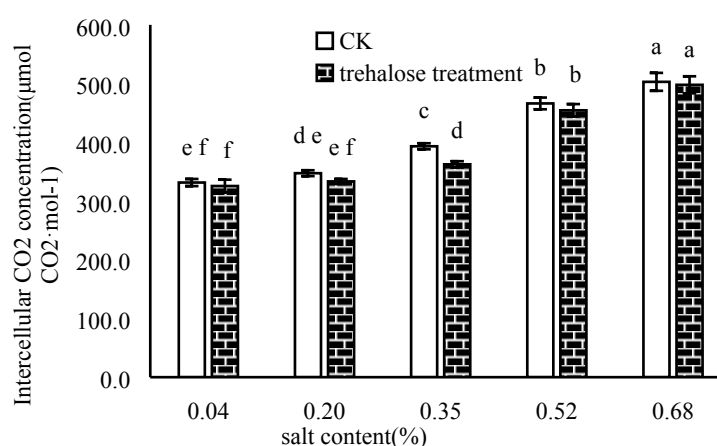
Stomatal conductance is the opening degree of stomata in plant leaves, which affects the photosynthesis, respiration and transpiration rate of plants. As can be seen from figure 3, with the increase of soil salt content, the stomatal conductance of waxy maize seedlings in the control group and the treatment group decreased gradually. The stomatal conductance in the treatment group was significantly higher than the control group, increasing by 26.26% and 14.79% at 0.20% and 0.35% salt concentration, respectively. The results showed that promote stomatal could open by exogenous trehalose treatment of waxy maize seedlings under salt stress.



**Fig 3.** Changes of stomatal conductance of waxy maize seedlings treated with trehalose under different soil salt

### 3.4 Effects of Trehalose on Intercellular CO<sub>2</sub> Concentration of Waxy Maize Seedlings under Salt Stress

As shown in figure 4, the intercellular CO<sub>2</sub> of waxy maize seedlings increased with the increase of salt concentration. The intercellular CO<sub>2</sub> concentration in the treatment group was significantly lower than the control group, which decreased by 7.81% at 0.35% salt concentration. Under other soil salt contents, the treatment group could reduce intercellular CO<sub>2</sub> concentration, but not significantly.

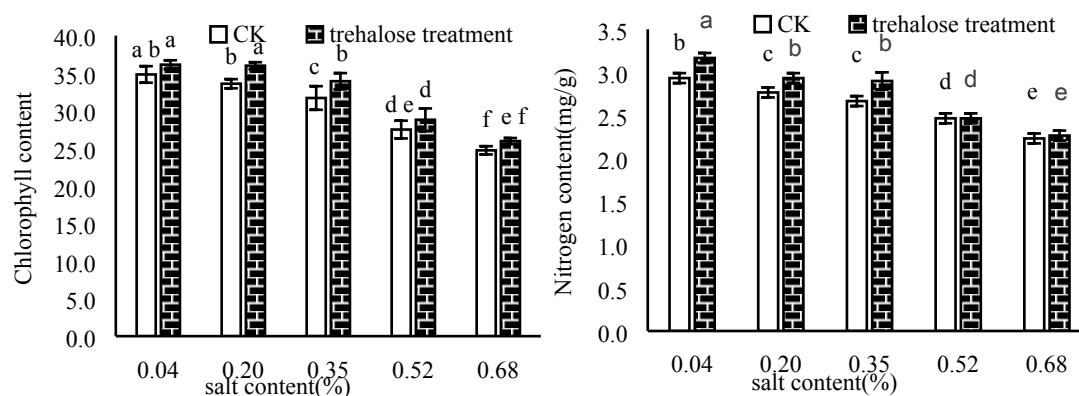


**Fig 4.** Changes of intercellular CO<sub>2</sub> concentration of waxy maize seedlings treated with trehalose under different soil salt

### 3.5 Effects of Trehalose on Chlorophyll Content and Nitrogen Content of Waxy Maize Seedlings under Salt Stress

Chlorophyll is the main pigment in photosynthesis of plants, and it is a kind of substance that affects photosynthesis. The nitrogen content in seedlings affects the photosynthetic capacity of seedlings, and the higher the nitrogen content, the stronger the photosynthesis of the seedlings. As can be seen from figure 5, the chlorophyll content in both control group and treatment group significantly decreased with the increase of waxy maize seedlings of soil salt content under between 0.20% and 0.68% salt concentration. The treatment group compared with the control group, the chlorophyll content significantly increased 7.14% and 6.94% at 0.20% and 0.35% salt concentration of waxy maize seedlings, respectively. As can be seen from figure 5, the nitrogen content of waxy maize seedlings in both the control group and the treatment group showed decreasing trend with the increase of salt content. The nitrogen content could significantly increase by exogenous trehalose of waxy maize seedlings, increased by 10.34%, 3.57% and 7.41% at 0.04%, 0.20% and 0.35% salt concentration, respectively. But the nitrogen content could increase to some extent of waxy maize

seedlings under other soil salt content, but not significantly.



**Fig 5.** Changes of chlorophyll content and nitrogen content of waxy maize seedlings treated with trehalose under different soil salt content

#### 4. Discussion

A lot of research indicates that net photosynthetic rate, transpiration rate, stomatal conductance and intercellular CO<sub>2</sub> concentration are inhibited under salt stress [16-17], thus reduced the photosynthesis of plants and caused inhibition of plant growth and death [18]. Under normal environmental conditions, plants can adjust the internal water potential balance by themselves under various stresses, once this balance is broken, the internal gas exchange system will be damaged and affect the growth and development of plants [19-20]. Studies have shown that leaf stomatal conductance could be decrease under various stresses, thus resulted the net photosynthetic rate, transpiration rate and intercellular CO<sub>2</sub> concentration decreased [21-22]. Similar conclusions were drawn in this experiment, stomatal conductance, net photosynthetic rate and transpiration rate of waxy maize seedlings decreased under salt stress. Trehalose could effectively improve photosynthesis of rice under salt stress [23]. In this experiment, the stomatal conductance, net photosynthetic rate and transpiration rate of waxy maize seedlings under salt stress were improved by soaking seeds and spraying 1 mmol·L<sup>-1</sup> trehalose. Iordachescu M et al. found that *arabidopsis thaliana* maintains its growth by exogenous trehalose under salt stress [24]. Garcia A B et al. found that trehalose could alleviate the damage of salt stress to rice, and reduce the accumulation of sodium and the loss of chlorophyll in leaves [25]. In this experiment, it was found that exogenous trehalose could promote chlorophyll synthesis and increase nitrogen content of seedlings under the condition of low soil salt content. Zeng Hongxue found that salt damage affects chloroplast thylakoid composition, the absorption and transformation of light energy, electron transfer and carbon assimilation in leaves. Excessive salt content could reduce the activity of enzymes involved in photosynthesis, chlorophyll content, stomatal conductance and CO<sub>2</sub> concentration, thus reducing the photosynthetic rate [26]. Garg A K et al. found that the resistance of rice to abiotic stress could be improved by introducing trehalose gene into rice [27]. Li Gailing found that salt stress significantly reduced the net photosynthetic rate, transpiration rate and stomatal conductance of black wheat leaves, and with the extension of stress time, the intercellular CO<sub>2</sub> concentration increased first and then decreased [28]. The application of exogenous trehalose alleviated the declining trend of net photosynthetic rate, transpiration rate and stomatal conductance, and improved photosynthesis. This experiment also reached a similar conclusion. Trehalose improved net photosynthetic rate, stomatal conductance and transpiration rate, and accelerated the photosynthetic rate.

#### 5. Conclusion

Net photosynthetic rate could significantly increase by trehalose treatment of waxy maize seedlings under salt stress. Under low salt stress, the transpiration rate and stomatal conductance

could significantly increase by trehalose treatment of waxy maize seedlings. The intercellular CO<sub>2</sub> concentration could significantly reduce by trehalose treatment of waxy maize seedlings under a certain value of soil salt content. Under low salt stress, the nitrogen content and chlorophyll content could significantly increase by trehalose treatment of waxy maize seedlings. The photosynthetic characteristics could be change by trehalose treatment, reduce the damage to photosynthesis under salt stress, and thus improve the photosynthesis of waxy maize seedlings under salt stress. Salt tolerance could be improved by trehalose treated waxy maize seeds and seedlings.

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