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Effects of Zn^{2+} Stress on Seed Germination and growth of seedling

Wenbin Liu,Aizhen Shao,Chuanying Wang

Horticultural garden college, Yangzhou Agricultural University, Jiangsu, China

ABSTRACT

The effects of Zn2+ stress on seed germination, seedling growth and chlorophyll content were studied in order to better understand the effect of heavy metal Zn on the growth and development of green plants. The concentration gradient of Zn2+ was 20, 50,100,150,200,300,500,700mg / L, and deionized water was used as control. The results showed that under the Zn2+ stress condition, the germination index of the rhubarb seeds increased with the increase of Zn2+ concentration. Germination potential, germination rate and germination index were the highest when Zn2+concentration was 100mg / L, the conductivity was the lowest at zinc concentration of 100mg / L, the root length, stem length and chlorophyll content of Zn2+ gradually reduced. The results showed that the amount of Zn2+ could promote seed germination, but the root length, bud length and chlorophyll content of seedlings could be affected by different degrees. The zinc fertilizer should be used in the production.

KEYWORDS: Vegetable rhubarb, seed germination, seedling growth, conductivity, chlorophyll content

Introduction

Rheum rhaponticum L. is a perennial herb of Polygonaceae, and its petioles are edible. The root can be used as medicine, and there are more than 500 varieties [1] in the world. Vegetable rhubarb plant height 1 -1.5 m, rhizome stout. Stems erect, upper branches, sparsely pubescent, abaxially coat; flowering from 6-7 months, fruit from 7-8 months. Vegetable rhubarb as an achene, with 3 edges, along the edge of the three wings, and with a membrane-like species of wings, the seed 1000 grain weight of about 20 g, germination force can be maintained for more than 3 years [2-3]. Vegetable rhubarb with large juicey petiole, rich in vitamins A, C, B1, B2, Ca, P, K and other mineral elements, as well as the human body must be a variety of amino acids, succinic acid and other substances, these nutrients and most vegetables And fruit similar [4]. According to the data: In addition to containing a certain amount of sugar, protein, and a small amount of tannin, per 100 g fresh petiole carotenoids 4.05 mg, vitamin C150 mg [5], pectin content of 1.30 g, slightly higher than the apple And pears, organic acids are mainly malic acid. Vegetable rhubarb can also be used for the production of canned food such as canned, dessert, pie, jam, syrup and wine. It has become a traditional food in western people's daily life [6-8], organic anthracene, quinone derivatives and emodin, has a high medicinal value. At the same time, the vegetable rhubarb has the characteristics of large strain, strong adaptability, high yield, high yield, long harvest time and convenient management. A planting can be harvested continuously for 4-6 years, with stable economic yield traits and high economy Benefit [9].

It is necessary to study the soil conditions suitable for the growth of rhubarb in order to create a better condition for the growth of rhubarb and to meet the market demand. Plant growth and development in addition to the need for 'a lot of elements', but also a small amount of 'trace elements', these trace elements in the plant although the content is very small, but it plays a vital role in plant growth and development. Zinc is one of the indispensable trace elements in plant growth and development. It can act as a component of different auxiliary factors in six functional enzymes, and has the function of regulating enzyme activity. Zinc and carbohydrate conversion are closely related to the synthesis of chlorophyll to promote photosynthesis. Zinc is also essential for crop root cell membrane, cell structure stability and functional integrity. Zinc protects root and root cell membrane effects, which can improve the drought resistance of crops [10].

At present, China's research on vegetable rhubarb although the introduction of information, but not mature enough for the growth of vegetables, rhubarb essential nutrients on the introduction is very few. In this study, we studied and analyzed the germination characteristics and seedling growth status of the seeds of Rhubarb by heavy metal zinc, so as to better understand the stress effect of heavy metal zinc on the growth and development of rhubarb, and provide the technical basis for further soil remediation.

1. Materials and methods

1.1. Materials

The experiment was conducted by the introduction of rhubarb seeds from Henan Science and Technology College, and was collected from Xinguang Ancient Guzhai Experimental Base in June 2013. The experiment was conducted in Henan University of Science and Technology in November 2013. The seed weight of the seeds used was 22.5 g.

1.2. 1.2 Methods

1.2.1 Germination test

This experiment uses a method of germination on paper. Firstly, choose the harmless pests, the grain must be consistently full and no damage to the vegetables with a few pieces of rhubarb seeds, use distilled water to process immersion treatment for 24 hours, so that the seeds will be swelling. ZnSO4 solution was prepared at a concentration of 0, 20, 50, 100, 150, 200, 300, 500 and 700 mg / L, respectively, based on ZnSO4. Take 27 of clean culture dish , let the seeds soaked in the dry water, according to each dish 30 into a double-layer filter paper plate, respectively, by adding different concentrations of ZnSO4 solution, each concentration repeated 3 times, The side of the culture dish was labeled with a lid and placed in a constant temperature incubator at 21 °C. To ensure that the incubator is having 12 hours a day of light, with the corresponding concentration of zinc solution every day to add the evaporation of water, so that the seeds can be concentrated in the solution under the conditions of germination. The germination rate was calculated after 5 days, and the germination rate was calculated after 7 days. The root length, stem length and chlorophyll content of the seedlings were measured after 14 days.

Germination potential (%) = (the number of days required for the normal germination of seeds / total number of tested seeds) × 100%

Germination rate (%) = (total number of normal germinated seeds / total number of tested seeds) × 100%

Germination index (GI) = Σ (Gt / Dt) (Gt is the number of germination in the time t, Dt is the corresponding number of germination days) [11]

1.2.2 Determination of root length and stem length of seedlings

After 14 days of culture, 10 seedlings were randomly selected from each sample, and the root length and stem length were measured. Finally, the mean value was calculated.

1.2.3 Determination of electrolyte leakage rate

Test the Electrolyte exudation rate by using conductivity meter determination, with reference to Zhang Zhiliang's method [12].

Twenty different sorghum seeds were washed with different concentrations of ZnSO4 solution, and 20 seeds were soaked seeds. They were washed and dried with distilled water, placed in 50 mL Erlenmeyer flask, 20 mL of distilled water was added and soaked for 4. 5 h. The flask was shaken and the solution was homogenized and filtered into a 50 mL beaker. Each treatment was repeated three times to determine the conductivity of the filtered solution.

1.2.4 Determination of chlorophyll content

The leaves of Rhizoma nidulata were extracted with 80% acetone and centrifuged at 6000 rpm for 10 minutes. The supernatant was taken and the absorbance at 663 and 645 nm was measured with a spectrophotometer. According to Lambert-Beer the law calculates the chlorophyll content [13] in mg / g.

2. Results and analysis

2.1. Effects of Zn2+ Concentration on Seed Germination of

Germination rate and germination potential are often used to evaluate the commonly used indicators of seed germination, reflecting the seed germination rate and germination uniformity. It was found from Table 1 that the germination rate of rhubarb seeds was different with the concentration of Zn2+ solution in different concentrations of Zn2+ solution treated with different concentrations of Zn2+ solution in the presence of deionized water (0mg / L) And germination potential were higher than the control. Among them, when the Zn2+ solution was at 100mg / L, the germination rate and germination potential of the seeds were at the highest level, respectively, 86% and 82%, and the germination of the seeds of the rhubarb was promoted effect. However, under the high concentration of Zn2+ solution, the germination rate and germination potential of the rhubarb seeds were lower than those of the control. When the seed germination rate and germination potential of the seeds were 200mg / L, the seed germination rate Inhibition, and with the increase of Zn2+ concentration, inhibition gradually increased. The results showed that the effect of Zn2+ solution on the seed germination of Rhubarb was low.

The germination rate, germination rate and germination index of the seeds could reflect the resistance of the plant in the bud stage at a certain Zn2+ concentration. The greater the germination index, the faster the germination rate, the better the better. Germination index is an important parameter to reflect the quality of the seed.

As can be seen from Table 1, different concentrations of Zn2+ solution treatment of vegetable rhubarb germination index impact are relatively large. When the concentration of Zn2+ was 200mg / L, the germination index of the seeds was lower than that of the control, and the germination index was gradually decreased with the increase of the concentration of Zn2+ when the concentration of Zn2+ was 100mg / L. , The greater the inhibition of the seeds of rhubarb. The results showed that low concentration of Zn2+ was beneficial to seed germination. High concentration of Zn2+ could inhibit the seed germination.

（mg/L）	（%）	（%）		（cm）	（cm）
0（CK）	78	70	7.34	4.46	2.29
20	74	71	7.67	4.32	2.27
50	81	77	7.86	4.04	2.14
100	86	82	7.91	3.98	1.89
150	80	72	7.46	3.56	1.74
200	73	65	6.18	2.69	1.56
300	68	64	5.43	1.80	1.62
500	67	64	4.89	1.12	1.45
700	64	60	4.21	0.56	1.26

Table 1. The effects of zinc concentration on the germination of rhubarb and the root length and stem length

Effects of Zn2+ Concentration on Root Length and Stem Length of Vegetable Rhubarb Seedlings

Zn2+ as a plant essential life activities necessary nutrients, the amount of Zn2+ on the growth of plants is essential. As can be seen from Table 1, with the increase of Zn2+ concentration, the root length and stem length of each treated seed were gradually shortened. For example, in the treatment of 0mg / L Zn2+ solution, the root length of 4.46cm, stem up to 2.29cm, but in 700mg / L Zn2+ solution treatment, the root length is only 0.56cm, stem length is only 1.26cm, Under the conditions of almost do not see the normal growth of young roots and shoots. Although the root length of Zn2+ was less than 100mg / L, the root length was shortened, but the shortening was not so obvious. When Zn2+ concentration exceeded 100mg / L, the root length was shortened. The results showed that Zn2+ could inhibit the root length and stem length of Rhubarb, and the inhibitory effect on root length was more obvious with the increase of Zn2+ concentration. Heavy metal Zn2+ not only affected the germination rate and germination potential of seeds, but also affected the growth of rhubarb seedlings. The root length and stem length of rhubarb seedlings were decreased, which indicated that Zn2+ could inhibit the growth of rhubarb seedlings.

2.2. Effects of Zn2+ Concentration on Electrolyte Exudation Rate of Vegetable Rhubarb

Plant cell membrane is the interface and barrier of material exchange and information transfer between plant cells and the external environment. It is a selective membrane. The stability of the biofilm system is the basis for the normal physiological function of the cells and the key part of the plant's injury. So the permeability of the cell membrane can be used to evaluate the response of plants to heavy metal contaminants. The permeability of cell membrane is the main physiological index of plant metabolism and stress resistance. It can be detected by detecting the change of its conductivity [14] The From Table 2, it can be seen that the Zn2+ concentration is different from the conductivity measured for the locust of the genus rhubarb, which indicates that the Zn2+ concentration has an effect on the cell membrane permeability of the vegetable rhubarb seeds. When the Zn2+ concentration is lower than 100mg / L, the conductivity is lower than the control (0mg / L), and with the increase of Zn2+ concentration, the conductivity of the seed is gradually decreasing. At 100mg / L, the conductivity is the lowest. When the concentration of Zn2+ continues to increase, the conductivity gradually increased, more than 200mg / L, higher than the control (0mg / L).

Zn^{2+}浓度（mg/L）	电导率（Us/cm）
0（CK）	36.33 c BC
20	35.67 cd C
50	34.67 d CD
100	32.67 e D
150	34.33 d CD
200	36.33 c BC
300	38.00 b AB
500	39.67 a A
700	39.33 ab A

Note: The lowercase letters after the same number of digits indicate the difference between the 0.05 level, respectively, and the capital letters indicate the difference between the 0.01 level.

Table 2. Effect of Zinc concentration on conductivity of vegetable rhubarb

2.3. Effects of Zn2+ Concentration on Chlorophyll Concentration in Vegetable Rhubarb Seedlings

Chlorophyll is the main pigment of photosynthesis in plants, and its content directly affects plant photosynthesis. It can be seen from Table 3 that the contents of chlorophyll a, chlorophyll b and chlorophyll (a + b) decreased with the increase of Zn2+ concentration when the seedlings of rhubarb were grown on the 14th day.

（mg/L）	Chla (mg/g)	Chlb(mg/g)	Chla+b(mg/g)
0(CK)	0.7783	0.2504	1.0287
20	0.7586	0.2401	0.9987
50	0.7390	0.2298	0.9688
100	0.7292	0.2311	0.9603
150	0.6896	0.1911	0.8807
200	0.6692	0.1856	0.8548
300	0.6050	0.1627	0.7677
500	0.6043	0.1515	0.7558
700	0.6101	0.1420	0.7521

Table 3. Effect of Zinc concentration on chlorophyll content in vegetable rhubarb seedlings

3. Discussion

The effects of different concentrations of Zn2+ solution on the seed germination characteristics and seedling growth of Rhubarb were studied by using the method of germination on the paper. The results showed that Zn2+ solution had a low concentration promoting effect and high concentration inhibition on the germination of rhubarb seeds. In this experiment, when the concentration of Zn2+ was lower than 150mg / L, the germination rate, germination potential and germination index of the rhubarb seeds were promoted to different degrees. At 100mg / L, the germination rate, germination potential and germination index reached the highest level. When Zn2+ concentration is more than 150mg / L, the germination rate, germination potential and germination index of the topical rhubarb seeds are lower than that of the control, and the high concentration of zinc can destroy the internal structure of the rhubarb seeds Enzyme activity, thus inhibiting the seed germination. The conductivity of seeds was the lowest at the concentration of Zn2+ at 100mg / L, and higher than 150mg / L. The effect of heavy metal Zn2+ on the membrane system of the seeds was determined by measuring the electrolyte permeation rate of the seeds treated with different concentrations of Zn2+ solution. The lower the conductivity, the seed cell exudate will become less, the higher the integrity of the cell membrane. This experiment further shows that low concentration of Zn2+ can promote seed germination.

Root length, stem length and chlorophyll content can reflect the growth of seedlings of rhubarb with different concentrations of Zn2+ solution. In this experiment, the root length, stem length and chlorophyll content of the seedlings decreased with the increase of Zn2+ concentration, which indicated that Zn2+ could not only inhibit the growth of young roots and shoots of rhubarb, But also on the synthesis of leaf green also inhibit the role.

Zn2+ is a necessary nutrient for plant growth. Appropriate amount of Zn2+ plays an important role in the synthesis of many key enzymes and the stability of protein structure. Zn2+ also has the effects of oxidative stress on membrane lipid peroxidation, plasma membrane damage and membrane permeability change has a stable and protective effect [15]. Studies have shown that heavy metals on the seed germination and seedling effects of a lower concentration of the stimulating effect and the higher concentration of the inhibitory effect [16]. In this paper, the germination of rhubarb seeds under Zn2+ stress was further studied. From the experimental results, the results of this experiment were consistent with those of previous studies. In the process of germination, Zn2+ could repair the damaged membrane system, and could effectively reduce the oozing of the seeds in the germination seed, and the permeability of the seeds during germination The damage of the seeds after low concentration of Zn2+ treatment was lower than that of the control, and the integrity of the membrane was also repaired. When the concentration of Zn2+ is too high, the permeability of the seed membrane of the seed is destroyed, the conductivity of the seed increases with the increase of zinc concentration, and the electrolyte leakage rate increases with the increase of zinc concentration. The results showed that the proper amount of Zn2+ could promote the germination of the seeds of rhubarb seeds by the determination of the conductivity of the seeds of rhubarb seeds treated with different concentrations of Zn2+ solution. The results also showed that zinc could inhibit the root length, bud length and chlorophyll content of rhubarb seedlings. Stobart and others believe that the reason for the decrease in chlorophyll content is that heavy metals inhibit the activity of chlorophyllate reductase and affect the synthesis of amino-r-pentanic acid, both of which are necessary for the synthesis of chlorophyll, resulting in a decrease in chlorophyll content [17], as to which concentration of zinc can make the root length of the seedlings, stem length, chlorophyll content and seed germination and other physiological indicators can be better coordinated, remains to be further studied.

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