Response of Lolium perenne Growth to Cadmium Stress

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Abstract. The chlorophyll, malondialdehyde and relative conductivity of Lolium perenne had different degrees of physiological response to Cd stress. Lolium perenne had certain physiological adaptability to Cd stress, which could reduce the damage of Cd to Lolium perenne cells. Low concentration of Cd stress could also promote the synthesis of chlorophyll and increase the photosynthetic products. Once the concentration of Cd was too high, the growth of Lolium perenne would be affected. The accumulation of Cd in plants exceeds a certain limit, which damages the physiological function of plants and affects the normal growth of plants. It is suggested that Lolium perenne has a certain remediation ability to cadmium contaminated soil. The experiment can provide a certain reference for the screening of cadmium tolerant plants and the remediation of cadmium contaminated soil.

Key words: Lolium perenne L., cadmium stress, biomass, physiological characteristics.

1.  Introduction
Lolium perenne L. is a perennial herb, which is widely used and has strong resistance [1]. With the continuous development of industry and urbanization, industrial and domestic sewage discharge and exhaust emissions are the main causes of soil heavy metal pollution [2]. Cadmium is one of the most toxic heavy metal elements to plants, which is easy to be absorbed and accumulated by plants [3]. However, the area of cadmium contaminated land in China has reached 13000 hm² [4].

Phytoremediation technology can reduce the enrichment of heavy metals in soil and transfer the heavy metals from soil to plants. However, when the concentration of heavy metals in plants exceeds a certain amount, it will inhibit the growth of plants and even lead to plant death. Lolium perenne has a strong ability to enrich cadmium. Sun et al. and Li et al. found that under low concentration of cadmium stress, seed germination and plant growth were promoted, and the growth was significantly inhibited under high concentration cadmium stress[5-6]. In this experiment, the effects of different concentrations of cadmium stress on the biomass, chlorophyll, and malondialdehyde contents of Lolium perenne were studied, and its physiological changes under stress were understood, which provided theoretical basis for phyto remediation in cadmium contaminated areas. To understand its physiological changes under stress, and provide a theoretical basis for phyto remediation in cadmium contaminated areas.
2. Study methods
Lolium perenne seeds of the same size were disinfected with 0.1% HgCl2 solution for 10 min, washed with distilled water for 3 times, and then soaked for 12 hours. Lolium perenne seeds were planted in plastic pots with 120 seeds per pot, and were managed routinely. After four leaves were grown in seedlings, cadmium stress treatment began. Seven cadmium stress treatments were set up in this experiment (0 mg/L, 0.5 mg/L, 1 mg/L, 5 mg/L, 10 mg/L, 20 mg/L, 50mg/L). Repeat 3 times. 30 days later, the aboveground and underground biomass, chlorophyll content, malondialdehyde (MAD) content and relative electrical conductivity were measured. The data were sorted out by Microsoft Excel 2010 and analyzed by SPSS 13.0.

3. Results and analysis

3.1. Effects of Cd stress on biomass
Under the stress of different concentrations of cadmium pollution, the biomass of Lolium perenne showed different changes. A certain concentration of cadmium stress promoted the biomass growth of Lolium perenne, while high concentration of cadmium stress (50mg/L) inhibited the growth of aboveground parts (Table 1). However, they were not statistically significant. The underground biomass of Lolium perenne decreased first and then increased. When the concentration of Cd was 1.0 mg/L, the underground biomass was the least, only 13.4% of the control. Under Cd stress of 50.0 mg/L, the underground biomass of Lolium perenne increased. The results showed that the underground biomass of Lolium perenne under different concentrations of cadmium stress was significantly lower than that of the control group.

| Cd stress treatments (mg/L) | aboveground biomass(g) | underground biomass(g) |
|---------------------------|------------------------|------------------------|
| 0.0                       | 12.921±1.386a          | 4.890±1.575a           |
| 0.5                       | 14.653±1.201a          | 2.061±0.714ab          |
| 1.0                       | 14.758±1.744a          | 0.654±0.215b           |
| 5.0                       | 14.388±1.413a          | 0.982±0.095b           |
| 10.0                      | 14.363±1.328a          | 0.875±0.159b           |
| 20.0                      | 16.705±0.704a          | 1.136±0.316b           |
| 50.0                      | 12.886±0.822a          | 2.872±1.725ab          |

Note: different letters in the same column indicate significant differences (p<0.05).

3.2. Effects of Cd stress on chlorophyll content
As shown in Fig.1, the chlorophyll content of Lolium perenne was not significantly affected by low concentration of cadmium stress, and the chlorophyll content increased by 16.32% under 50.0mg/l cadmium stress. However, the results of statistical analysis showed that the chlorophyll content of Lolium perenne under different concentrations of cadmium stress was not significantly different from that of the control group (P>0.05). The results showed that Lolium perenne had a certain physiological adaptability to cadmium stress. In this study, Cd stimulated the defense mechanism of Lolium perenne, resisted the toxicity of Cd and prevented the damage of Cd to plant cell structure and function.
3.3. Effects of Cd stress on MDA content

As shown in Fig. 2, the MDA content of Lolium perenne increased with the increase of stress concentration under 0-50.0 mg/l Cd stress, but the change was not obvious. When the concentration of Cd was 50.0 mg/L, the content of MDA in Lolium perenne reached the maximum value of 0.0172 umol/g, which was 6.8% higher than that of the control. There was no significant difference between the other treatments and the control. The results showed that Cd stress had little damage to Lolium perenne. Lolium perenne could resist the toxicity of Cd by its own mechanism. Under higher Cd stress, the content of MDA in Lolium perenne might increase, which affected the normal physiological metabolism and plant growth.

3.4. Effects of Cd stress on relative conductivity

As shown in Fig. 3, the conductivity increased gradually with the aggravation of Cd stress. Under 50.0 mg/l Cd stress, the relative conductivity was the highest, which was 2.6 times higher than that of the control (P < 0.05). The results showed that the cell membrane permeability of ryegrass was damaged by cadmium stress. With the increase of Cd stress concentration, the damage of plant cell membrane system was more serious.
4. Discussion

Cd is a non-nutritive element, which is toxic to plants and can affect the metabolism and growth of plant cells [7]. With the increase of Cd concentration, the aboveground biomass of Lolium perenne increased at first and then decreased. When the stress concentration reached 20.0mg/l, the aboveground biomass reached the maximum, then 20.0mg/l was the critical value of aboveground biomass decline, which indicated that the aboveground part of Lolium perenne had certain resistance to Cd, but it would be damaged beyond a certain limit. The underground biomass of Lolium perenne was lower than that of the control under different concentrations of Cd stress, which indicated that the resistance of Lolium perenne to Cd was low, and the more Cd accumulated in Lolium perenne roots, the more serious the toxicity.

Chlorophyll is the material basis of photosynthesis, and its content can directly reflect the growth status of plants. If the content of chlorophyll in plant leaves is high, the plant growth condition is good, otherwise, the plant growth condition is poor [8]. Under Cd stress, the chlorophyll content of Lolium perenne was significantly different from that of the control, which indicated that Lolium perenne had adaptability to Cd accumulation and could resist Cd toxicity. Previous experimental results showed that chlorophyll content increased first and then decreased with the increase of cadmium stress concentration, that is, it promoted chlorophyll synthesis at low concentration and inhibited chlorophyll synthesis at high concentration [5], but this experiment generally showed an upward trend. When cadmium concentration was 50.0 mg/L, chlorophyll content was the highest, and it could promote chlorophyll synthesis of Lolium perenne in this study.

Under stress, plant cells produce superoxide anion free radicals and hydroxyl free radicals, which induce membrane lipid peroxidation of unsaturated fatty acids in membrane to produce free radicals. Free radicals can induce membrane lipid peroxidation and damage cells [9]. MDA is the final product of membrane lipid peroxidation. It can destroy the structure of cell plasma membrane and affect the normal physiological metabolism of plants. MDA content in plants is an important indicator of whether the cell membrane is damaged or not [10]. The more MDA content, the more serious the damage to plants. MDA content in this experiment changed little. Only an increasing trend was showed under high concentration of Cd Stress. It can aggravate the peroxidation of cell membrane lipid of Lolium perenne, make a large amount of MDA accumulate in the plant, and cause damage to Lolium perenne. Relative conductivity is one of the important physiological indexes for measuring cell membrane damage [11]. The larger the relative conductivity index is, the more serious the damage of cell membrane structure is. The results of this experiment were consistent with the results of previous studies.
5. Conclusion
The results showed that the aboveground part of Lolium perenne was more resistant to Cd than the underground part, and the underground part was more susceptible to Cd. At the same time, the chlorophyll, malondialdehyde and relative conductivity of Lolium perenne had different degrees of physiological response to Cd stress. Lolium perenne had certain physiological adaptability to Cd stress, which could reduce the damage of Cd to Lolium perenne cells. Low concentration of Cd stress could also promote the synthesis of chlorophyll and increase the photosynthetic products. Once the concentration of Cd was too high, the growth of Lolium perenne would be affected. The accumulation of Cd in plants exceeds a certain limit, which damages the physiological function of plants and affects the normal growth of plants.

Acknowledgments
This work was supported by the project of the CAS Key Laboratory of Mountain Ecological Restoration and Bioresource Utilization & Ecological Restoration and Biodiversity Conservation Key Laboratory of Sichuan Province (KXYWS2004), the scientific research fund of Sichuan Provincial Education Department (16ZA0306, 18ZA0245), the scientific research fund of bamboo diseases and pests control and resources development key laboratory of Sichuan Province (17ZZ004, 17ZZ005), the project of LeShan Normal University (XJR17006, ZZ201821, DGZZ202019, LZDP008, DGZZ202010), the project of LeShan science and technology bureau (20ZDYJ0276).

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