The influence of application hyperaccumulator plant straw on photosynthetic pigment content and photosynthetic parameter of lettuce under cadmium stress

Le Liang¹, Qiaoman Ao², Ying Zhu², Yan Zhao², Ran Zhang² and Yi Tang^{1*}

¹Institute of Pomology and Olericulture, Sichuan Agricultural University, Chengdu, Sichuan, 611130, China

²College of Horticulture, Sichuan Agricultural University, Chengdu, Sichuan, 611130, China

*Corresponding author's e-mail: 95459425@qq.com

Abstract. Pot experiments were conducted to study the effects of straw application of three hyperaccumulator plants (*Solanum nigrum*, *Bidens pilosa* and *Galinsoga parviflora*) influence photosynthetic pigments content and photosynthetic parameters of lettuce (*Lactuca sativa*) under Cd stress. The results showed that: compared with no straw application, the photosynthetic pigment content of lettuce was increased by applying three kinds of hyperaccumulator plants straw (*S. nigrum*, *B. pilosa* and *G. parviflora*), but there was no significant difference among the three kinds of hyperaccumulator plant straws; also improved the net photosynthetic rate (Pn), Stomatal conductance (Gs), transpiration rate (Tr), intercellular CO₂ concentration (Ci) of lettuce, among them, the effect of *G. parviflora* straw was the most obvious. Application of three kinds of hyperaccumulator plant straw promoted the growth of lettuce by increasing the photosynthetic pigment content and photosynthetic parameter of lettuce under Cd stress.

1 Introduction

Straw returning is an agricultural measure being vigorously promoted, which can avoid air pollution caused by straw burning, it can also promote crop growth by improving soil fertility and soil properties, and reduce the bioavailability of heavy metals such as copper, cadmium (Cd) and zinc [1]. One of many heavy metals, Cd pollution in soil mainly comes from transportation, agricultural inputs, sewage irrigation and sludge fertilization [2]. Cd can seriously harm plants and affect human health, there are many ways to alleviate Cd toxicity: intercropping [3], grafting [4], application of biomass charcoal [5] and so on. Applying straw of *Tagetes erecta* and *Solanum photeinocarpum* to Cd-contaminated soil at 10 mg/kg significantly increase tomato biomass and photosynthetic pigments content, and significantly decrease Cd content [6]. Hyperaccumulator plant can effectively repair heavy metal pollution. Soil application of hyperaccumulator plant straw has little research on Cd accumulation in leafy vegetables. Therefore, this experiment explored the effects of hyperaccumulator plant straw on photosynthetic pigments and photosynthetic parameters of lettuce under Cd pollution. It provides a new method for leafy vegetables to alleviate heavy metal pollution.

2 Materials and Methods

2.1. Materials

In June 2015, three kinds of Cd hyperaccumulator plants were collected from the surrounding farmland (not contaminated by heavy metals) in the Sichuan Agriculture University: *Solanum nigrum*, *Bidens pilosa* and *Galinsoga parviflora*, after removed the seeds, they were washed with distilled water. After 15 minutes of killing fresh at 105 °C, they are dried to constant weight at 75 °C. Cutting the dried straw into about 1 cm segments and set aside.

Lettuce is a common glass lettuce in Sichuan, it has high purity, good quality, good tolerance and resistance, and can be planted all year round and sustainable harvest for more than 50 days.

The tested soil was paddy soil, which was taken from farmland not polluted by heavy metals around Chengdu campus of Sichuan Agricultural University, its basic physical and chemical properties are as follows: pH 6.36, organic matter 18.10 g/kg, total nitrogen 1.04 g/kg, total phosphorus 1.36 g/kg, total potassium 20.78 g/kg, hydrolyzable nitrogen 43.90 mg/kg, available phosphorus 92.71 mg/kg, available potassium 134.25 mg/kg, available Cd not detected. CdCl₂·2.5H₂O analytical pure solution was added to the tested soil for heavy metal Cd.

2.2. Experimental design

In June 2015, the air-dried and crushed soil was sifted through 300 meshes and loaded into barrels, each containing 20 kg. The Cd concentration was 10 mg/kg by adding analytical pure CdCl₂·2.5 H₂O solution [7], after

30 days of moist storage, all contaminated soils were fully blended and packed in a pot of 3 kg per pot.

Full lettuce seeds were select for disinfection, evenly spread them on wet gauze, put them into 20 °C light incubator for germination, and then sow them in a pot dish with nutritive soil to raise seedlings. After one week of lettuce seedling raising, three kinds of hyperaccumulator plant straw were applied to the prepared Cd contaminated soil, 6 g per pot, and mixed evenly, with no straw as the control. The experiment was divided into four treatments: no straw, *S. nigrum* straw, *B. pilosa* straw and *G. parviflora* straw. After a week of balancing and watering, selected healthy lettuce seedlings and transplant them into Cd-contaminated soil prepared with straw. Four plants were transplanted in each pot and nine pots were treated in each pot. After transplanting, the seedlings were placed in plastic greenhouse, and the soil moisture was maintained at about 80%. After the lettuce grew 50d, chlorophyll content and photosynthetic parameters of lettuce were determined.

2.3. Index determination

Determination of chlorophyll content by acetone-ethanol mixture method [8]. Measurement of net photosynthetic rate (Pn) by LI-6400 XT portable photosynthetic meter, stomatal conductance (Gs), transpiration rate (Tr), intercellular CO₂ concentration(Ci). The intrinsic light intensity was set at 1000μmol/ (m²·s), the concentration of CO₂ was 400 μl/L, and the temperature was 25 °C [9].

2.4. Statistical analyses

All data are collated with Excel 2010 software; SPSS 20.0 was used for variance analysis and Duncan's new complex range method was used for multiple comparisons.

3 Results

3.1. Effects of soil application of hyperaccumulator plant straw on photosynthetic pigment content in lettuce leaves

Application of hyperaccumulator plant straw increased the chlorophyll content of lettuce (Table 1). The application of *B. pilosa* straw and *G. parviflora* straw significantly increased chlorophyll a content in lettuce leaves. The content of chlorophyll a in lettuce leaves treated with *B. pilosa* straw was the highest, 25.27% higher than that of the control ($P < 0.05$). When application *B. pilosa* straw, lettuce had the highest chlorophyll b content, but there was no significant difference among the treatments. The different trend of total chlorophyll content of lettuce was the same as that of chlorophyll a content. The order from high to low was *B. pilosa* straw > *G. parviflora* straw > *S. nigrum* straw > no straw. The different trend of carotenoids in lettuce was the same as that of chlorophyll a and total chlorophyll. The carotenoid content of lettuce treated with *B. pilosa* straw was the highest, 42.86% higher than that of the control ($P < 0.05$). The ratio of chlorophyll a to chlorophyll b in lettuce increased after the application of hyperaccumulator plant straw, and the treatment with *G. parviflora* straw was the highest.

Table 1. Effects of hyperaccumulate plant straw on the photosynthetic pigment of lettuce

Plant straw	Chlorophyll a (mg/g)	Chlorophyll b (mg/g)	Total chlorophyll (mg/g)	Carotenoids (mg/g)	Chlorophylla/b
No straw	0.91±0.01c	0.29±0.01a	1.20±0.01c	0.28±0.01b	3.14
<i>S. nigrum</i> straw	0.96±0.02bc	0.29±0.01a	1.25±0.03c	0.35±0.01ab	3.31
<i>B. pilosa</i> straw	1.14±0.01a	0.33±0.01a	1.47±0.01a	0.40±0.01a	3.45
<i>G. parviflora</i> straw	1.04±0.01ab	0.30±0.01ba	1.34±0.02b	0.37±0.01ab	3.47

Different lowercase letters in the same column indicate significant difference ($P < 0.05$).

3.2. Effects of soil application of hyperaccumulator plant straw on photosynthetic parameters of lettuce leaves

All treatments increased Pn in lettuce leaves, there was no significant difference between *B. pilosa* straw and *G. parviflora* straw treatments, but they were significantly higher than no straw treatment, which increased by 14.06% ($P < 0.05$) and 14.06% ($P < 0.05$), respectively (Table 2). Leaf Gs of lettuce increased after the

application of hyperaccumulator plant straw, and the order of its size was: *G. parviflora* straw > *S. nigrum* straw > *B. pilosa* straw > no straw, there was no difference among the treatments, but it was significantly higher than the control. For lettuce leaf Ci, the application of *G. parviflora* straw increased by 6.13% compared with the control ($P < 0.05$). Hyperaccumulator plant straw significantly increased leaf Tr of lettuce under Cd stress. The order of Tr in each treatment was as follows: *G. parviflora* straw > *B. pilosa* straw > *S. nigrum* straw > no straw. The Tr of lettuce leaves was the highest after applying *G. parviflora* straw, 46.01% significantly higher than that of the control ($P < 0.05$).

Table 2. Effects of hyperaccumulate plant straw on photosynthetic parameter of lettuce

Plant straw	Pn ($\mu\text{mol}/(\text{m}^2 \text{ s})$)	Gs ($\text{mol}/(\text{m}^2 \text{ s})$)	Ci ($\mu\text{mol}/\text{mol}$)	Tr ($\text{mmol}/(\text{m}^2 \text{ s})$)
No straw	13.58 $\pm$ 1.06c	0.26 $\pm$ 0.02b	285.37 $\pm$ 10.36d	3.26 $\pm$ 0.18d
<i>S. nigrum</i> straw	15.76 $\pm$ 1.29a	0.39 $\pm$ 0.02a	290.72 $\pm$ 13.13c	4.54 $\pm$ 0.11b
<i>B. pilosa</i> straw	15.49 $\pm$ 0.60b	0.36 $\pm$ 0.03a	297.36 $\pm$ 4.19b	4.28 $\pm$ 0.23c
<i>G. parviflora</i> straw	15.49 $\pm$ 1.12b	0.40 $\pm$ 0.02a	302.87 $\pm$ 3.57a	4.76 $\pm$ 0.13a

Different lowercase letters in the same column indicate significant difference ($P < 0.05$).

4 Conclusion

Under Cd stress, the chlorophyll content of lettuce leaves was significantly increased by applying straw from hyperaccumulator plants (*S. nigrum* straw, *B. pilosa* straw and *G. parviflora* straw) in soil, but there was no significant difference among treatments; the photosynthetic parameters of *G. parviflora* straw increased most obviously when applied. Therefore, under the condition of Cd pollution, the application of three hyperaccumulator plants straws (*S. nigrum* straw, *B. pilosa* straw and *G. parviflora* straw) were conducive to the normal growth of lettuce.

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