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# Effects of intercropping with post-grafting generation of *impatiens balsamina* on phosphorus uptake in grape seedlings under cadmium stress

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**Abstract.** A pot experiment was conducted to study the effects of intercropping with post-grafting generation of *Impatiens balsamina* on phosphorus (P) uptake in grape roots, stems, leaves and available P content in soil under cadmium (Cd) stress. In the experiment, four grafting treatment [ungrafted (UG), self-rooted grafting by the same one seedling (PSG), self-rooted grafting by two different sizes seedlings (PSDG) and self-rooted grafting by two same sizes seedlings (PSSG)] were used to intercrop with grape seedlings. The result showed that intercropping with PSG significantly increased the content of total P in grape roots and stems, but other treatments all reduced it. The content of total P in grape leaves for the treatment of intercropping with PSDG was 24.36% higher than the monoculture and other treatments reduced it. The treatment of intercropping with UG significantly increased the content of soil available P. Therefore, under Cd stress, intercropping with post-grafting generation of *I. balsamina* has an impact on grape content.

## 1. Introduction

Cadmium (Cd) is one of the five toxic elements of heavy metals. It has the characteristics of long decomposition period, high mobility, high toxicity and difficult degradation. It is easy to be absorbed and enriched by crops in production activities, which seriously affects the yield and quality of crops [1-2]. In agricultural production, phytoremediation is mainly adopted for heavy metal pollution control and intercropping is the main agronomic measure [3]. Intercropping can not only improve the effective use of soil nutrients, water and light by crops, thereby increasing crop yields, but also improve the soil environment and increase crop uptake of nutrients [4-5]. *Impatiens balsamina* is a Cd accumulator flower plant [6] and research shows that grafting with self-rooted seedlings can promote the growth of the progeny of *I. balsamina* under Cd pollution, increase its absorption and accumulation of Cd, and thus improve its ability to repair Cd-contaminated soil [7]. Therefore, this study investigated the effects of intercropping with post-grafting generation of *I. balsamina* on grape P then to screen post-grafting generation of *I. balsamina* that can improve the grape P absorption.



## 2. Materials and methods

### 2.1. Materials

The seeds of the *I. balsamina* used in this experiment were collected from the same yellow-flowered and double-petaled *I. balsamina* in the farmland surrounding Chengdu Campus of Sichuan Agricultural University (30° 42' N, 103° 50' E) in October 2014. The cultivar of grape is Kyoho with cutting seedlings, and it was purchased from the nursery stock base in Longquanyi District, Chengdu, Sichuan, China, in May 2015. The soil samples used in this experiment were collected from the farmland surrounding Chengdu Campus of Sichuan Agricultural University, pH 7.09, total N 1.50 g/kg, total P 0.76 g/kg, total K 18.02 g/kg, total Cd 1.96 mg/kg, alkali N 94.82 mg/kg, available P 6.30 mg/kg, available K 149.59 mg/kg, Cd not detected. The basic physical and chemical properties of the soil and the determination of heavy metal Cd content are based on references [8].

### 2.2. Experimental design

October 2014, the seeds were collected from the same flower of *I. balsamina* which were put in the climate chamber to germination, the interval between two grow seedlings was 2 weeks. Grafting treatment was carried out when the first of seedlings were about 10 cm higher (the second batch of seedlings were about 5 cm higher), the grafting treatment as follows: (1) Ungrafted: the seedlings of *I. balsamina* transplanted directly, and the seeds were collected for preservation as the ungrafted generation of *I. balsamina* (UG). (2) Self-rooted grafting by the same one seedling: the seedlings of *I. balsamina* were cut off from 6 cm above the ground. The upper parts were scion and the lower parts were rootstock. Keep rootstock leaves. Scions and rootstocks were physiologically consistent and collected seeds for preservation as the post-grafting generation of self-rooted grafting by the same one seedling of *I. balsamina* (PSG). (3) Self-rooted grafting by two different sizes seedlings: *I. balsamina* seedlings were about 10 cm high, cut off from 6 cm above the ground, the lower parts were rootstock. The scions were cut the upper seedling (4 cm) from seedlings of *I. balsamina* were about 5 cm high, and the leaves of rootstock were kept after grafting. There is a big difference between the scion and rootstock in physiology, and the seeds were collected for preservation as the post-grafting generation of self-rooted grafting by two different sizes seedlings of *I. balsamina* (PSDG). (4) Self-rooted grafting by two same sizes seedlings: the *I. balsamina* seedlings were about 10 cm high, and cut off from 6 cm above the ground and divided into two parts. One part was rootstock, other part was scion (4 cm), and the leaves of rootstock were retained after grafting. Scion and rootstock are different in physiology, and collected seeds for preservation as the post-grafting generation of self-rooted grafting by two same sizes seedlings of *I. balsamina* (PSSG).

In May 2015, the seeds in offspring of *I. balsamina* that treated with different grafting technology were put in the climate chamber to germination and further cultivation. Then, the seedlings of *I. balsamina* transplanted together with grape seedlings into pot which prepared with soil by 5 mg/kg Cd when the two true leaves expanded. Five treatments were applied in this experiment: grape monoculture, grape intercropping with UG, grape intercropping with PSG, grape intercropping with PSSG, grape intercropping with PSDG. One *I. balsamina* seedling of different treatments and one grape seedling were transplanted into each pot. For each treatment with six replicates and the pots placed completely random. The distance between pots was 15 cm, and the pot position exchanged periodically to weaken the impact of the marginal effects. The soil moisture content was maintained at 80% of field capacity until the plants were harvested.

After 2 months, grape seedlings were harvested and divided into three parts of root, stem and leaf, then washed with tap water firstly, followed by deionized water for three times. Finally, the fresh weight of plants were weighed and then simmered for 15 min at 110 °C. After that, the tissues of all plants were dried at 80 °C until constant weight, weighed, passed through a 100-mesh sieve to determine the content of P different parts of the grape seedlings by molybdenum antimony colorimetric method [8] and available P content in soil by Molybdenum antimony colorimetric method [9].

### 2.3. Statistical analyses

Statistical analysis was carried out by using SPSS 18.0 statistical software. The data were analyzed by one-way ANOVA, with the least significant difference at the 5% confidence level.

## 3. Results and discussion

### 3.1. Total P content in grape roots

The intercropping patterns decreased the content of total P in grape roots except the treatment of intercropping with PSG (Figure 1) which was 8.16% higher than the monoculture. Intercropping with UG, PSDG and PSSG decreased the content of total P compared to monoculture of grape, and they were 27.89% ( $p < 0.05$ ), 25.17% ( $p < 0.05$ ) and 21.77% ( $p < 0.05$ ) lower than the monoculture, respectively. Sorting the content of total P in grape roots of each treatment from highest to lowest: I.PSG > MG > I.PSSG > I.PSDG > I.UG. Among all treatments, the content of total P in grape roots was highest by intercropping with PSG.

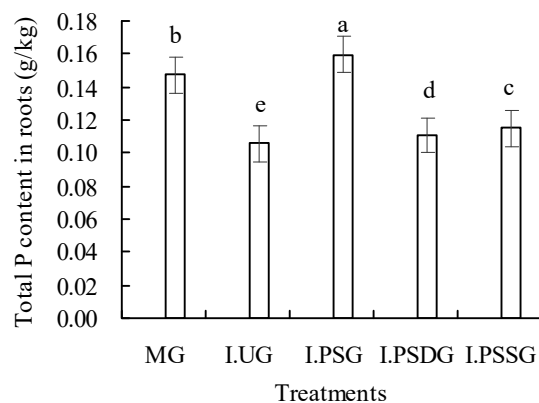


Figure 1. Total P content in grape roots. Different lowercase letters indicate significant differences based on one-way analysis of variance in SPSS 17.0 followed by the least significant difference test ( $p < 0.05$ ). Grape monoculture = MG, grape intercropping with the generation of ungrafted *I. balsamina* = I.UG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same one seedling = I.PSG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by two different sizes seedlings = I.PSDG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same sizes seedlings = I.PSSG.

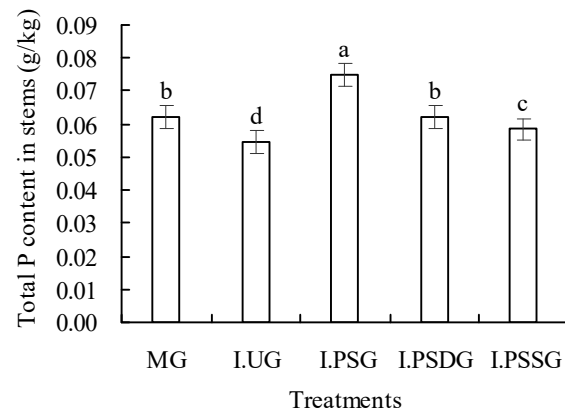


Figure 2. Total P content in grape stems. Different lowercase letters indicate significant differences based on one-way analysis of variance in SPSS 17.0 followed by the least significant difference test ( $p < 0.05$ ). Grape monoculture = MG, grape intercropping with the generation of ungrafted *I. balsamina* = I.UG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same one seedling = I.PSG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by two different sizes seedlings = I.PSDG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same sizes seedlings = I.PSSG.

### 3.2. Total P content in grape stems

Compared to monoculture, only intercropping with PSG significantly increased the content of total P in grape stems and it was 20.97% ( $p < 0.05$ ) higher than the monoculture. Intercropping with UG and PSSG reduced the content of total P compared to monoculture of grape, and they were 11.29% ( $p < 0.05$ ) and 6.45% ( $p < 0.05$ ) lower than the monoculture, respectively. For the total P content in grape stems, the order from large to small was ranked: I.PSG, MG = I.PSDG, I.PSSG, I.UG. Among all treatments, the content of total P in grape stems was highest by intercropping with PSG.

### 3.3. Total P content in grape leaves

Under Cd stress, for the total P content in grape leaves, the order from large to small was ranked: I.PSDG, MG, I.PSG, I.PSSG, I.UG (Figure 3). Compared to monoculture, in addition to intercropping with PSDG significantly increased the content of total P in grape leaves other treatments all significantly reduced it. The total P content in grape leaves for the treatment of intercropping with PSDG was 24.36% ( $p < 0.05$ ) higher than the monoculture. The total P content in grape leaves in the three intercropping patterns of intercropping with UG, PSG and PSSG were 26.92% ( $p < 0.05$ ), 5.13% ( $p < 0.05$ ) and 14.10% ( $p < 0.05$ ) lower than the monoculture, respectively.

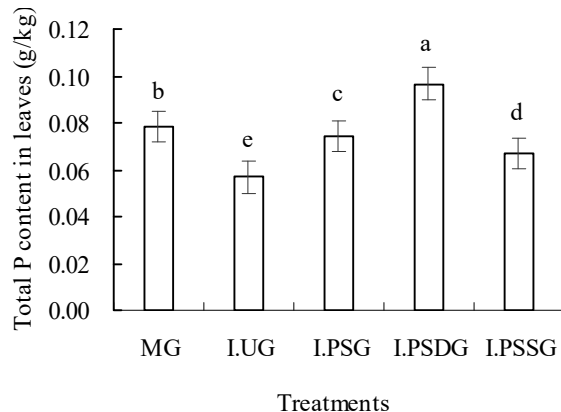


Figure 3. Total P content in grape leaves.

Different lowercase letters indicate significant differences based on one-way analysis of variance in SPSS 17.0 followed by the least significant difference test ( $p < 0.05$ ). Grape monoculture = MG. grape intercropping with the generation of ungrafted *I. balsamina* = I.UG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same one seedling = I.PSG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by two different sizes seedlings = I.PSDG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same sizes seedlings = I.PSSG.

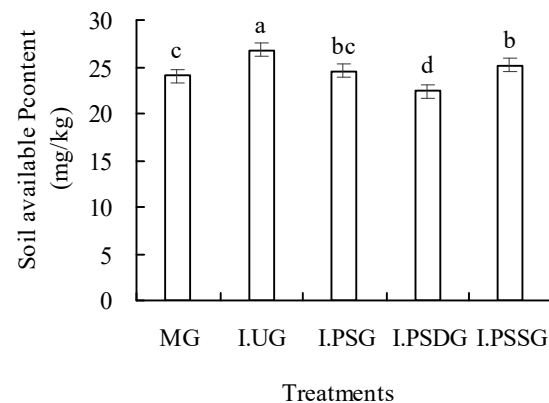


Figure 4. Soil available P content after intercropping. Different lowercase letters indicate significant differences based on one-way analysis of variance in SPSS 17.0 followed by the least significant difference test ( $p < 0.05$ ). Grape monoculture = MG. grape intercropping with the generation of ungrafted *I. balsamina* = I.UG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same one seedling = I.PSG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by two different sizes seedlings = I.PSDG, grape intercropping with *I. balsamina* of the post-grafting generation of self-rooted grafting by the same sizes seedlings = I.PSSG.

### 3.4. Soil available P content

Intercropping with UG, PSG, PSDG and PSSG had different effects on the content of soil available P (Figure 4). Intercropping with UG, PSG and PSSG increased the content of soil available P compared to monoculture of grape, and they were 11.57% ( $p < 0.05$ ), 2.04% ( $p > 0.05$ ) and 4.58% ( $p < 0.05$ ) higher than the monoculture, respectively. On the contrary, intercropping with PSDG can decrease the content of soil available P. The content of soil available P for the treatment of intercropping with PSDG was 6.67% ( $p < 0.05$ ) lower than the monoculture.

## 4. Conclusions

According to the experiment, we got the following conclusion: intercropping with PSG significantly increased the content of total P in grape roots (8.16% higher than the monoculture) and stems (20.97% higher than the monoculture), other treatments all reduced it. The content of total P in grape leaves for the treatment of intercropping with PSDG was 24.36% higher than the monoculture and other

treatments all significantly reduced it. The treatment of intercropping with UG significantly increased the content of soil available P (11.57% higher than the monoculture). Therefore, under Cd stress, intercropping with post-grafting generation of *I. balsamina* has an impact on grape content.

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