Effects of Intercropping with Post-Grafting Generation of *Cosmos sulphureus* on Total Potassium Content in Grape Seedling under Cadmium Stress

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**Abstract.** The effects of intercropping with post-grafting generation of *Cosmos sulphureus* on total potassium content of grape seedling were investigated through pot experiment under cadmium stress. The results showed that the intercropping treatments had significant effect on soil available potassium content and total potassium content in root, stem and leaf of grape seedling, while intercropping with ungrafted and post-grafting generation of *C. sulphureus* all decreased the potassium content of grape seedling and soil compared with grape monoculture. The potassium content of grape seedling and soil were all ranked as: grape monoculture (MG) > intercropping with the ungrafted generation of *C. sulphureus* (UG) > intercropping with the post-grafting generation of *C. sulphureus* of self-rooted grafting by the same one seedling (PSG) > intercropping with the post-grafting generation of *C. sulphureus* of self-rooted grafting by two different sizes seedlings (PSDG) > intercropping with the post-grafting generation of *C. sulphureus* of self-rooted grafting by two same sizes seedlings (PSSG). Therefore, intercropping with post-grafting generation of *C. sulphureus* could not increase but reduce the potassium content, which is one of essential minerals that contributes to plant growth and development.

1. Introduction

*Cosmos sulphureus* is also known as sulfur cosmos and yellow cosmos, which is considered a half-hardy annual, in shades of yellow, orange, and red, and its native habitat is Central America [1]. Plant grafting is widely applied in breeding of the new variety, improved varieties, rejuvenation of mature trees and so on [2], and grafting significantly affects plant growth and there is a difference among grafted plants [3]. The intercropping is a technology that could provide an increase in productivity per unit area and a diversified production of food in the same area, generating better temporal distribution of income, an increase of vegetative protection of soil against erosion, and better control of weeds as compared with the sole cropping system [4]. The different intercropping systems showed different amounts of dry matter accumulation, potassium content and potassium concentration [5]. There is a research showing that intercropping with *C. sulphureus* and *Impatiens balsamina* reduced the nutrient absorption of grape seedlings under Cadmium stress [6]. Therefore, we used...
grape seedlings and *C. sulphureus* to intercrop to study the effects on potassium content in grape seedling.

2. Materials and methods

2.1 Materials

The seeds of the *C. sulphureus* used in this experiment were collected from the same yellow-flowered and double-pealed *C. sulphureus* in the farmland surrounding Chengdu Campus of Sichuan Agricultural University in October 2014. The species of the experimental cultivar of grape is Kyoho and the cutting seedlings were purchased from seedling base in Longquanyi area of Chengdu in May 2015. The soil for the experiment was collected from the surrounding farmland of Chengdu campus of Sichuan Agricultural University in October 2014, pH 7.09, total potassium 18.02 g/kg, and available potassium 149.59 mg/kg. The basic physical and chemical properties of the soil are based on references [7]. Cd was used as a heavy metal for testing and it was added to the soil samples in the form of CdCl₂·2.5H₂O according to design concentration.

2.2 Experimental design

In October 2014, the seeds were collected from the same flower of *C. sulphureus*. Then, they were put in the climate chamber to germination, and the interval between two growing seedlings was 2 weeks. Grafting treatment was carried out when the first batch 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 *C. sulphureus* transplanted directly, and the seeds were collected for preservation as the ungrafted generation of *C. sulphureus* (UG). (2) Self-rooted grafting by the same one seedling: the seedlings of *C. sulphureus* were cut off from 6 cm above the ground. The upper parts were scion and the lower parts were rootstock, and then retained rootstock leaves. Scions and rootstocks were physiologically consistent and collected seeds for preservation as the post-grafting generation of *C. sulphureus* of self-rooted grafting by the same one seedling (PSG). (3) Self-rooted grafting by two different sizes seedlings: *C. sulphureus* 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 *C. sulphureus* which were about 5 cm high, and the leaves of rootstock were retained after grafting. There was a big difference between the scion and rootstock in physiology, and the seeds were collected for preservation as the post-grafting generation of *C. sulphureus* of self-rooted grafting by two different sizes seedlings (PSDG). (4) Self-rooted grafting by two same sizes seedlings: when the *C. sulphureus* seedlings were about 10 cm high, one was cut off from 6 cm above the ground, kept the lower parts as rootstocks; one was cut off from 6 cm above the ground, kept the upper parts as scion (4 cm). The leaves of rootstock were retained after grafting. Scion and rootstock were different in physiology, and seeds should be collected for preservation as the post-grafting generation of *C. sulphureus* of self-rooted grafting by two same sizes seedlings (PSSG).

In May 2015, the seeds in offspring of *C. sulphureus* that treated with different grafting technologies were put in the climate chamber to germination and further cultivation. Then, the seedlings of *C. sulphureus* 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 seedling intercropping with UG, grape seedling intercropping with PSG, grape seedling intercropping with PSDG and grape seedling intercropping with PSSG. One *C. sulphureus* 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 dug up and divided into three parts of root, stem and leaf, then washed with tap water firstly, followed by deionized water for three times. Finally, weighed the
fresh weight 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 potassium content of different parts of the grape seedlings and available potassium content in soil.

2.3 Statistical analyses.
Statistical analysis was carried out by with 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 K content in root of grape seedling
Under cadmium stress, the total potassium content in root of grape seedling intercropping with UG, PSG, PSDG and PSSG were significantly lower than monoculture (Figure 1, \( P < 0.05 \)). In terms of the total K content of roots, intercropping with UG, PSG, PSDG and PSSG were decreased by 28.12%, 31.58%, 50.88% and 57.24%, respectively.

![Figure 1. Total K content in root of grape seedling](image)

Different lowercase letters indicated significant differences among treatments at 0.05 levels. MG = grape monoculture, I. UG = grape seedling intercropping with the ungrafted generation of \textit{C. sulphureus}, I. PSG = grape seedling intercropping with the post-grafting generation of \textit{C. sulphureus} of self-rooted grafting by the same one seedling, I. PSDG = grape seedling intercropping with the post-grafting generation of \textit{C. sulphureus} of self-rooted grafting by two different sizes seedlings, I. PSSG = grape seedling intercropping with the post-grafting generation of \textit{C. sulphureus} of self-rooted grafting by two same sizes seedlings.

3.2 Total K content in stem of grape seedling
Under cadmium stress, the total potassium content in stem of grape seedling intercropping with UG, PSG, PSDG and PSSG were all lower than monoculture (Figure 2, \( P < 0.05 \)). In terms of the total K content of stems, intercropping with UG, PSG, PSDG and PSSG were reduced by 3.12%, 17.23%, 21.35% and 30.07%, respectively.
Figure 2. Total K content in stem of grape seedling

Different lowercase letters indicated significant differences among treatments at 0.05 levels. MG = grape monoculture, I. UG = grape seedling intercropping with the ungrafted generation of C. sulphureus, I. PSG = grape seedling intercropping with the post-grafting generation of C. sulphureus of self-rooted grafting by the same one seedling, I. PSDG = grape seedling intercropping with the post-grafting generation of C. sulphureus of self-rooted grafting by two different sizes seedlings, I. PSSG = grape seedling intercropping with the post-grafting generation of C. sulphureus of self-rooted grafting by two same sizes seedlings.

Figure 3. Total K content in leaves

Different lowercase letters indicated significant differences among treatments at 0.05 levels. MG = grape monoculture, I. UG = grape seedling intercropping with the ungrafted generation of C. sulphureus, I. PSG = grape seedling intercropping with the post-grafting generation of C. sulphureus of self-rooted grafting by the same one seedling, I. PSDG = grape seedling intercropping with the post-grafting generation of C. sulphureus of self-rooted grafting by two different sizes seedlings, I. PSSG = grape seedling intercropping with the post-grafting generation of C. sulphureus of self-rooted grafting by two same sizes seedlings.

3.3 Total K content in leaf of grape seedling

Under cadmium stress, the total potassium content in leaf of grape seedling intercropping with UG, PSG, PSDG and PSSG were significantly lower than monoculture (Figure 3, $P < 0.05$). In terms of the total K content of leaves, intercropping with UG, PSG, PSDG and PSSG were decreased by 29.42%, 32.25%, 34.85% and 54.22%, respectively.
3.4 Available potassium content in soil
Intercropping with UG, PSG, PSDG and PSSG significantly decreased the available potassium content in soil under cadmium stress (Figure 4, \(P < 0.05\)), which reduced by 35.24%, 37.21%, 39.19% and 47.00%. During the five treatments, grape seedling intercropping with UG, PSG and PSDG had no significant difference, but compared with grape monoculture, they significantly reduced. Also, the soil available potassium content was the lowest by intercropping with PSSG compared with grape monoculture.

![Figure 4. Available K content in soil](image)

Different lowercase letters indicated significant differences among treatments at 0.05 levels. MG = grape monoculture, I. UG = grape seedling intercropping with the ungrafted generation of \(C. \) sulphureus, I. PSG = grape seedling intercropping with the post-grafting generation of \(C. \) sulphureus of self-rooted grafting by the same one seedling, I. PSDG = grape seedling intercropping with the post-grafting generation of \(C. \) sulphureus of self-rooted grafting by two different sizes seedlings, I. PSSG = grape seedling intercropping with the post-grafting generation of \(C. \) sulphureus of self-rooted grafting by two same sizes seedlings.

4. Conclusions
Under cadmium stress, after four intercropping treatments, the results showed that intercropping with ungrafted and post-grafting generation of \(C. \) sulphureus could affect the total potassium content in root, stem and leaf of grape seedling. Specifically, intercropping with ungrafted generation or the post-grafting generation by four different grafting technologies of \(C. \) sulphureus, the total potassium content in root, stem and leaf of grape seedling all decreased compared with the grape monoculture. Furthermore, the soil potassium content also reduced by intercropping compared with the grape monoculture. The content of total potassium and soil potassium was all the lowest by intercropping with the post-grafting generation of \(C. \) sulphureus of self-rooted grafting by two same sizes seedlings, compared with grape monoculture.

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