Effects of intercropping with reciprocal hybridization F1
generation of Solanum photeinocarpum on cadmium
accumulation of Cyphomandra betacea seedlings

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Abstract. In this experiment, C. betacea seedlings were used as research materials, intercropping S. photeinocarpum of parents and positive and negative hybridization F1 generation with C. betacea seedlings, study the cadmium accumulation characteristics of S. photeinocarpum positive and negative hybridization F1 generation and effects of intercropping them with C. betacea seedlings on cadmium accumulation characteristics of C. betacea seedlings. Intercropping with S. photeinocarpum of farmland ecotype and mine ecotype had no significant on the cadmium contents in roots and shoots of C. betacea seedlings, and intercropping S. photeinocarpum of positive and negative hybridization F1 generation decreased the cadmium contents in roots and shoots of C. betacea seedlings. Hybridization decreased the cadmium contents in roots and shoots of S. photeinocarpum, but increased the cadmium accumulation amounts in roots and shoots of S. photeinocarpum. Therefore, intercropping with hybridization F1 generation of S. photeinocarpum could reduce the cadmium uptake of C. betacea.

1. Introduction
Cyphomandra betacea belongs to the family Solanaceae, a perennial evergreen semi-wood shrub or small tree [1]. Its fruit can be used as fresh fruit, juicy and sweet, rich in minerals and vitamin C and other trace elements beneficial to the human body [2]. However, in recent years, the problem of heavy metal pollution in orchard soils has become increasingly serious [3]. Frequent heavy metal pollution problems have endangered the growth of C. betacea and other fruit trees, and fruit quality and safety. And there are many reports that the depth and breadth of cadmium pollution are the most serious among all types of heavy metal pollution in orchard soils in China [4-5]. Therefore, it is of great theoretical and practical significance to carry out the remediation of heavy metal pollution orchards, especially the remediation of soil cadmium pollution.

Phytoremediation technology is one of the many methods used in the treatment of heavy metal pollution. For heavy metal pollution in orchard soil, intercropping technology can be used to apply phytoremediation technology to the orchard soil heavy metal pollution repair. Under heavy metal pollution, the effect of intercropping plant species on the absorption of heavy metal elements is mainly concentrated in the rhizosphere environment, and the plants can secrete organic acids through signal feedback to complex heavy metals [6], reducing the effectiveness of heavy metals, and reducing common plants absorption of heavy metals. Reasonably matched intercropping species can not only increase the accumulation of heavy metals by hyperaccumulator plants, but also reduce the absorption
of heavy metals by ordinary plants, and harvest agricultural products that meet health standards. It is an ideal way to repair heavy metal pollution [7]. At present, by planting grass in fruit gardens and intercropping hyperaccumulator plants with fruit trees, a composite model of fruit trees and crop content in the stems, leaves and shoots of  C. betacea seedlings were sampled. Two months after the growth of the plants, the whole plant and seedlings were sampled respectively, and the soil was stored separately. Plant samples were rinsed with tap water, and then rinsed repeatedly with deionized water. They were killed at 105 °C for 15 min, then dried to constant weight at 70 °C and weighed. Nitration of the sample by nitric acid perchloric acid (4:1 by volume), ICAP6300 ICP spectrometer determines of cadmium content [9]. Weighted 10 g of air dried soil sample through 1 mm sieve and put it into a 25 ml beaker, add 10 ml of 0.01 mol/L CaCl₂, mix well, and let it stand for 30 minutes, with checked PHS-25 pH value tester determinates of pH value of suspension [11].

3. Results and discussion

3.1 Cadmium content in C. betacea seedlings
Among the treatments, only intercropping with farmland ecotype increased the cadmium content in roots of C. betacea seedlings by 2.96% (p > 0.05) compared with the monoculture of C. betacea seedlings (Table 1). Intercropping with mine ecotype, positive and negative decreased the cadmium content in roots of C. betacea seedlings by 2.76% (p > 0.05), 13.49% (p <0.05), and 12.14% (p <0.05), respectively, compared with the monoculture of C. betacea seedlings. The order of the cadmium content in the stems, leaves and shoots of C. betacea seedlings was: monoculture > intercropping with farmland ecotype > intercropping with mine ecotype > intercropping with negative > intercropping with positive. The cadmium content in the stems, leaves and shoots of C. betacea seedlings after

2. Materials and methods

2.1 Materials
The mine ecotype S. photeinocarpum collected from Tangjiashan lead zinc mine, Hanyuan Country, Sichuan Province, China, and the farmland ecotype S. photeinocarpum collected from the farm of Ya'an campus of Sichuan Agricultural University, Yucheng Country, Sichuan Province, China. Positive hybridization: Farmland ecotype is the female parent, and mine ecotype is the male parent. Negative hybridization: Mine ecotype is the female parent, and farmland ecotype is the male parent [9].

2.2 Experimental design
The soil from the farm of Sichuan Agricultural University was dried by air. The 21 cm × 20 cm (diameter × height) plastic basin was used to load 3.0 kg of air dried soil screened by 6.72 mm (3 mesh) and 10 mg/kg cadmium (added into the soil in the form of CdCl₂·2.5H₂O analysis pure form) was added [10]. The soil was kept moist and placed for 30 days. The soil was turned over and mixed irregularly to make the soil fully and evenly mixed. In the same month, C. betacea seeds were placed in the climate box for breeding. In August 2015, the seeds of parents and F1 hybrids of S. photeinocarpum were placed in the climate box for breeding.

The parents of S. photeinocarpum and F1 generation seedlings (about 3 cm high, 2 true leaves spread) were mixed with C. betacea seedlings (about 10 cm high, 3 true leaves spread) in a pot. There were 1 plant in each pot for mixed planting of S. photeinocarpum and 3 plants in each pot for single planting of C. betacea, 2 plants in each pot for mixed planting, each treatment repeated 6 times. In the process of cultivation, water in time to keep the field water capacity of the soil at about 80%, and pay attention to weed removal and pest control.

Two months after the growth of the plants, the whole plant and C. betacea were sampled respectively, and the soil was stored separately. Plant samples were rinsed with tap water, and then rinsed repeatedly with deionized water. They were killed at 105 °C for 15 min, then dried to constant weight at 70 °C and weighed. Nitration of the sample by nitric acid perchloric acid (4:1 by volume), ICAP6300 ICP spectrometer determines of cadmium content [9]. Weighted 10 g of air dried soil sample through 1 mm sieve and put it into a 25 ml beaker, add 10 ml of 0.01 mol/L CaCl₂, mix well, and let it stand for 30 minutes, with checked PHS-25 pH value tester determinates of pH value of suspension [11].
intercropping with positive decreased by 7.35% ($p > 0.05$), 4.14% ($p > 0.05$), and 4.08% ($p > 0.05$), respectively, compared with the monoculture of C. betacea seedlings.

| C. betacea | Roots (mg/kg) | Stems (mg/kg) | Leaves (mg/kg) | Shoots (mg/kg) |
|------------|---------------|---------------|----------------|----------------|
| Monoculture | 150.48±5.29a | 26.39±0.88a   | 28.04±0.79a    | 27.48±0.82a    |
| Inter. farmland | 154.93±4.99a | 25.32±1.00a   | 27.55±0.95a    | 26.67±1.08a    |
| Inter. mine | 146.32±4.25ab| 24.94±0.66a   | 27.28±0.68a    | 26.36±0.68a    |
| Inter. positive | 130.18±2.98b | 24.45±0.54a   | 26.88±0.61a    | 25.97±0.60a    |
| Inter. negative | 132.21±3.49b | 24.55±0.78a   | 26.99±0.72a    | 26.08±0.74a    |

Values are mean ± SD (n = 6). Different lowercase letters indicated significant differences among treatments at 0.05 levels.

3.2 Cadmium accumulation of S. photoinocarpum
The differences in cadmium content of different S. photoinocarpum treatments were significant ($p < 0.05$), and the differences in cadmium accumulation were not significant ($p > 0.05$, Table 2). The cadmium content in mine ecotype was the highest. The cadmium content in positive was the lowest, and its cadmium accumulation was the lowest.

| S. photoinocarpum | Cadmium content (mg/kg) | Cadmium accumulation (μg/plant) |
|-------------------|-------------------------|---------------------------------|
| Farmland          | 154.98±4.12ab           | 76.18±2.16b                     |
| Mine              | 160.13±4.41a            | 84.02±2.38a                     |
| Positive          | 142.01±3.48c            | 70.02±1.94c                     |
| Negative          | 146.65±3.89bc           | 72.66±2.04bc                    |

Values are mean ± SD (n = 6). Different lowercase letters indicated significant differences among treatments at 0.05 levels.

3.3 Soil pH and soil available cadmium concentration
Compared with the monoculture of C. betacea seedlings, intercropping with S. photoinocarpum increased the soil pH, and the differences were significant ($p < 0.05$, Figure 1). There were no significant differences of soil pH among four intercropping treatments.

Intercropping with positive significant ($p < 0.05$) decreased the soil available cadmium concentration compared with the monoculture of C. betacea seedlings (Figure 2). The other treatments of soil available cadmium concentration had no significant ($p > 0.05$) differences compared with the monoculture of C. betacea seedlings.

4. Conclusions
Intercropping with S. photoinocarpum of farmland ecotype and mine ecotype had no significant on the cadmium contents in roots and shoots of C. betacea seedlings, and intercropping S. photoinocarpum of positive and negative hybridization F1 generation decreased the cadmium contents in roots and shoots of C. betacea seedlings. Intercropping with S. photoinocarpum of positive hybridization F1 generation the cadmium contents in roots and shoots of C. betacea seedlings were lowest, compared with monoculture has respectively reduced by 13.49% and 4.08%. Respectively intercropping S. photoinocarpum of parents and positive and negative hybridization F1 generation with C. betacea seedlings, the cadmium accumulation amount of S. photoinocarpum of positive and negative hybridization F1 generation was all higher than parents, with higher ability of remediation of soil cadmium pollution, and especially the S. photoinocarpum of positive hybridization F1 generation was most advantageous. Hybridization decreased the cadmium contents in roots and shoots of S. photoinocarpum, but increased the cadmium accumulation amounts in roots and shoots of S.
Therefore, intercropping with hybridization F1 generation of *S. photeinocarpum* could reduce the cadmium uptake of *C. betacea*.

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