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Review article

Effective Transformation and Utilization of Selenate/Selenite in Soil by Tea Plants Infected with Endophytic *Herbaspirillum sp*. Strain WT00F

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Abstract

A novel technique was used to transform and utilize selenate/selenite in soil. Tea seedlings ontaining *Herbaspirillum sp*. strain WT00F were cultivated and then transplanted in leniferous soils. The grown tea plants not only effectively reduced the level of selenate/selenite in ferous soils but also enhanced selenium enrichment in tea leaves.

Introduction: As a nonmetallic element, selenium (Se) is a member of the chalcogens. In the nature, Se usually occurs in organic and inorganic forms. Its organic form (e.g. selenocysteine and selenoproteins, etc) mainly presents in living organisms where the organic selenocompounds can be metabolized [1,2], whereas its inorganic form (e.g. selenate and selenite oxoanions) primarily exists in natural environments. Selenate (SeO_4^{2-}) and selenite (SeO_3^{2-}) are water soluble so that they have potential mobility and bioavailability in the environment [3]. At low dosage, Se stimulates the growth of the plant whereas at high dosages it causes plant damage [4-6]. The deficiency of Se has been thought to be associated with over 40 human diseases [7,8] but the excessive intake of Se seriously damages human health [9]. Although the distribution area of Se is limited, Se-pollution in the surrounding land of selenium mining area is quite serious. Soluble Se^{6+} and Se^{4+} can be reduced to insoluble non-toxic elemental selenium (Seo) by microbes. The reduction of selenate/selenite to elemental selenium by microbes is an effective way to remove them from contaminated soil, water and drainage [10]. *Herbaspirillum sp*. strain WT00F is an endophytic bacterium isolated from tea plant (*Camellia sinensis L*) [11]. It enters plants via wound infection and only colonizes in Camellia plants as a specialist. Like *Herbaspirillum sp*. strain WT00C [12], this bacterium has a strong capability of reducing selenate and selenite to form red elemental selenium (Seo) or selenoproteins. Moreover, this bacterium also stimulates lateral root formation and bud growth of tea cuttings once it enters tea pole via the incision at the end of tea cuttings [13]. Thus, we attempted to use this bacterium to promote tea plants to transform and utilize selenate/selenite in soil and found that the tea plants colonized by *Herbaspirillum sp*. strain WT00F effectively utilized selenate/selenite in soil and enriched selenium in tea leaves.

Technological Scheme

The technical route was simply described as follows: *Herbaspirillum sp*. strain WT00F was cultured in the modified LB medium (yeast extract 5g, peptone 10g, NaCl 1.5g in 1L H_2O, pH7.0) at 37 °C until OD600 of 1.0. Tea cuttings (5-7 cm length) were soaked in the bacterial culture supplemented 1% glycerol and diluted with H_2O (1:1) for 1h. Then, tea cuttings infected by *Herbaspirillum sp.* strain WT00F were planted in the nursery to grow into tea seedlings. Finally, tea seedlings were transplanted in seleniferous soils. The field management was performed according to the conventional method for tea cultivation.

Experimental Effects and Discussion

Transformation and utilization of selenate/selenite in soil were very effective via planting tea plants colonized by the strain WT00F. Figure.1 showed selenium enrichment in tea leaves. In the tea plants colonized by the strain WT00F, selenium amount of tea leaves was increased 2-3 fold compared to the tea plant without colonization of the strain WT00F. Effective transformation of selenate by the bacterium inside tea plant accelerated selenate absorption of tea-plant roots and thus reduced the level of selenate in soil. The

possible mechanism of transforming and utilizing selenate/selenite in soil by tea plants infected with the strain WT00F is proposed as follows: when tea plants grow in seleniferous soils, selenate/selenite enters tea plants via root absorption and reaches to tea-plant stem through vessel transportation. As we known, selenate/selenite are soluble but quite toxic [14], whereas elemental selenium (Se0) is insoluble and atoxic [15,16]. *Herbaspirillum sp*. WT00F predominantly colonizes inside the stem of tea plants, and effectively reduces selenate/selenite to produce elemental selenium, selenodiglutathione, glutathioselenol, selenocysteine, selenomethionine and selenoproteins when inorganic selenocompounds are absorbed via roots and transported into the stems of tea plants through the conducting system of tea-plant xylem. Thus, the strain WT00F not only decreases the toxicity of inorganic selenocompounds absorbed by roots but also forms organic selenium and Se-nanoparticles. Organic selenium and Se-nanoparticles can be further metabolized by tea-plant cells and finally accumulated in the tissues of tea leaves. In conclusion, applying this technique not only effectively reduces the level of selenate/selenite in seleniferous soils but also enhances Se-enrichment in tea leaves.

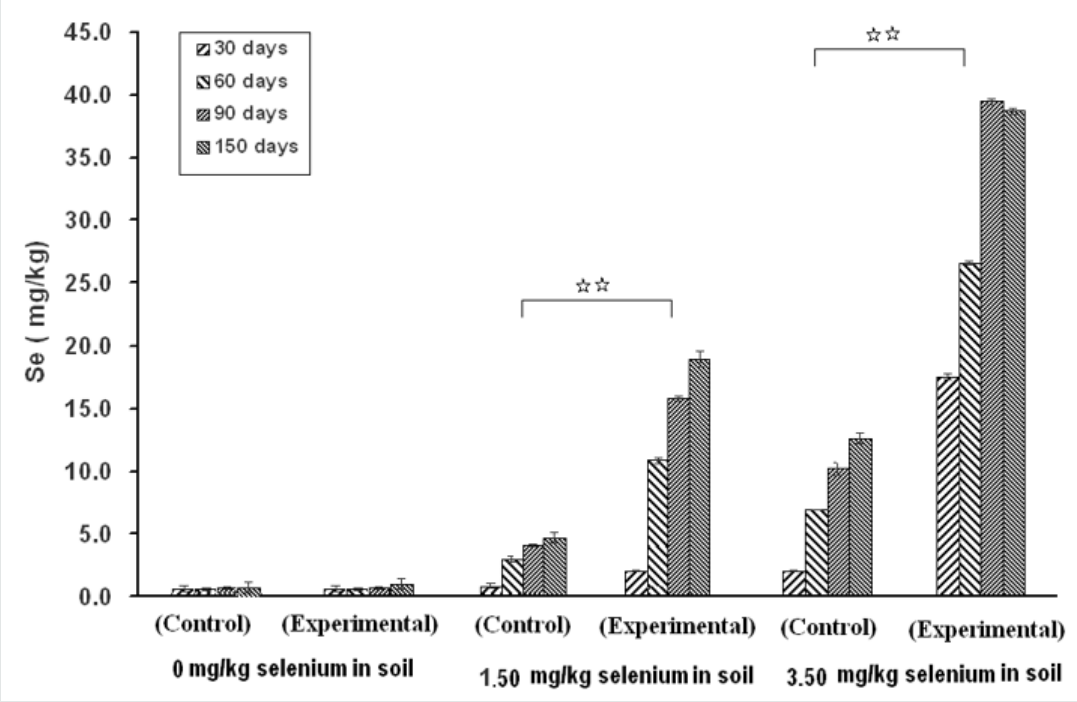

Figure 1: Selenium contents of tea leaves. The data were collected at 30, 60, 90 and 150 days after the survival and growth of tea seedlings transplanted in the soils containing 0, 1.5 and 3.5 mg/kg selenate. Control: tea seedlings without bacterial treatment; Experimental: tea seedlings infected with the strain WT00F; ☆☆: P<0.01.

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