

Comparison of Zinc Accumulating Efficiency between *Brassica juncea* Coss. and *Brassica chinensis* Linn.

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The zinc accumulating efficiencies of *Brassica juncea* Coss. and *Brassica chinensis* Linn. which may be used as a guideline for treatment of zinc-contaminated soil, were compared. The plants were grown in soil with 0, 10, 100, and 1,000 mg zinc added. Detection of the zinc accumulation in both plant species at 46 days showed that *B. chinensis* was more effective in zinc accumulation than *B. juncea* when 0, 10, and 100 mg zinc were added. However, for *B. juncea*, zinc was accumulated more when 1,000 mg zinc was added. The zinc accumulating efficiencies of the shoots and roots of each species were also compared. For *B. juncea*, zinc accumulating efficiency in the roots and the shoots were not significantly different. However, for *B. chinensis*, the zinc accumulating efficiency of shoots tended to be higher than that of the roots. It can be concluded that the species of plants can accumulate zinc differently, depending on the concentration of zinc in the contaminated soil. *B. juncea* accumulated more zinc (4,825 mg/kg dry wt. plant) when 1,000 mg of zinc was added, while *B. chinensis* accumulated more zinc (4,766 mg/kg dry wt. plant) when 100 mg of zinc was added.

Keywords: phytoremediation, Zn-contaminated soil, *Brassica* spp.

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การศึกษาเปรียบเทียบประสิทธิภาพการสะสมสังกะสีใน ผักกาดเขียวปลี (*Brassica juncea* Coss.) และผักกาด เขียววางตุ้ง (*Brassica chinensis* Linn.)

นัยนันท์ อริยκανนัท กุลภา โสรรัตน์ และ ธิดาวิสุทธิ จันทน์เครือ (2546)

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การศึกษาเปรียบเทียบประสิทธิภาพการสะสมสังกะสีในผักกาดเขียวปลี (*Brassica juncea* Coss.) และผักกาดเขียววางตุ้ง (*Brassica chinensis* Linn.) มีขึ้นเพื่อเป็นแนวทางในการนำพืชไปใช้บำบัดดินที่ปนเปื้อนสังกะสี โดยทำการปลูกพืชทั้งสองชนิดในดินที่เติมสังกะสี 0, 10, 100 และ 1,000 มก. จากการวิเคราะห์หาปริมาณสังกะสีในพืชทั้งสองชนิดที่มีอายุ 46 วันเท่ากันพบว่า ผักกาดเขียววางตุ้งสามารถสะสมสังกะสีได้มากกว่าผักกาดเขียวปลีอย่างมีนัยสำคัญ เมื่อมีการเติมสังกะสีในดิน 0, 10 และ 100 มก. แต่ผักกาดเขียวปลีมีประสิทธิภาพในการสะสมสังกะสีมากกว่าอย่างมีนัยสำคัญ เมื่อมีการเติมสังกะสีลงไปในดิน 1,000 มก. จากการศึกษาการสะสมสังกะสีในแต่ละส่วนของพืชเพื่อเปรียบเทียบประสิทธิภาพในการสะสมสังกะสีระหว่างส่วนที่อยู่เหนือดิน (ใบและลำต้น) และส่วนที่อยู่ใต้ดิน (ราก) ของพืชแต่ละชนิดพบว่า การสะสมสังกะสีในส่วนที่อยู่เหนือดินและใต้ดินของผักกาดเขียวปลีไม่มีความแตกต่างกันอย่างมีนัยสำคัญ แต่สำหรับผักกาดเขียววางตุ้งพบว่าการสะสมสังกะสีของส่วนที่อยู่เหนือดินจะมากกว่าส่วนที่อยู่ใต้ดิน จากผลการศึกษาครั้งนี้สามารถสรุปได้ว่า พืชทั้งสองชนิดมีความสามารถในการสะสมสังกะสีได้แตกต่างกันขึ้นอยู่กับปริมาณสังกะสีในดิน โดยผักกาดเขียวปลีจะสะสมสังกะสีได้มากที่สุด (4,825 มก./กก.น้ำหนักแห้งพืช) เมื่อมีการเติมสังกะสีในดิน 1,000 มก. ในขณะที่ผักกาดเขียววางตุ้งจะสะสมสังกะสีได้มากที่สุด (4,766 มก./กก.น้ำหนักแห้งพืช) เมื่อมีการเติมสังกะสีในดิน 100 มก.

คำสำคัญ การฟื้นฟูสิ่งแวดล้อมโดยใช้พืช, ดินที่ปนเปื้อนสังกะสี, ผักกาดเขียว

INTRODUCTION

The use of land as a dumping site for sewage, industrial waste, metal, paint, cosmetics, fertilizer and pesticides can cause zinc-contamination of the soil. Although zinc is an important supplement for living organisms, it can also do harm if it taken at high doses.

Phytoremediation is a cleanup method for contaminated soil, groundwater, surface water, wastewater, and polluted air by using plants and/or plants that have a special mechanism to work with some microorganisms in the ordinary media. Phytoremediation is the most suitable remediation for a wide area since it offers a lower cost method for heavy metal-contaminated soil remediation compared to other remediations, such as soil removal, soil washing and solidification.⁽¹⁾ This method saves energy because the main energy used is solar energy, there is no soil removal which causes environmental impact and it is also safe to be used. Furthermore, the contaminated metal can be recovered.

Four categories of phytoremediation include *Phytovolatilization* which is the use of plants to make volatile chemical species of soil elements, *Rhizofiltration* which is the use of plant roots to remove contaminants from flowing water, *Phytostabilization* which is the use of plants to transform soil metals to less toxic forms, but not remove metals from the soil, and *Phytoextraction*, sometimes called phytoaccumulation, which is the use of plants to uptake contaminants from soils.⁽²⁾ Phytoextraction uses plants that have the ability to uptake heavy metals or contaminants through its roots, and

accumulate them in shoot tissues. Important factors to accomplish phytoextraction are: forms and quantities of contaminants in the environment, the accumulations of plant root, the transposition of contaminants in plant tissue, the tolerance of plant to contaminant, plant productivity, and accumulation factor. There are many plants used in phytoextraction on a research scale these days, such as *Thlaspi carerulescens*, which can accumulate 30 g of Zn/kg dry wt. in shoot, *Ipomoea alpina*, which can accumulate 12.3 g of Cu/kg dry wt. in shoots, and *Psychotria douarrei*, which can accumulate 47.5 g of Ni/kg dry wt. in shoots.⁽³⁾

However, this method has some limitations. Contaminants at level deeper than the plant root level cannot be treated. Phytoremediation is a time consuming method. The contaminants must be in the form that the plant can uptake. Also, there are only a few species of plants which are considered hyperaccumulators.

In this experiment, the removal of zinc from soil was studied because the water dissolvable and rapid movable characteristics of zinc and zinc compounds can cause pollution problems in a short period of time. The zinc accumulating efficiency between *Brassica juncea* Coss. and *Brassica chinensis* Linn. was compared. It was expected that these two plants could be used as effective hyperaccumulators. The results from this study will be applied to reduce zinc-contaminated soil pollution using the typical species in Thailand and to add to the database for zinc-contaminated soil removal research.

MATERIALS AND METHODS

Materials

56 sets of clay pots with 7 inch diameters were prepared for planting. *B. juncea* Coss. and *B. chinensis* Linn. seeds from Chia Tai Co., Ltd. and commercial mixed soil from “Mai Long Mai Ru” were used.

Methods

Soil properties

The properties of soil that affect plant growth and the absorption of Zn were measured. The physical properties such as soil texture and moisture content were analyzed using a hydrometer method⁽⁴⁾ and a gravimetric method.⁽⁵⁾ The chemical properties, including soil pH, cation exchange capacity, organic matter and total nitrogen, were measured using a potentiometric method (soil:distilled water = 1:1), NH₄OAc method,⁽⁶⁾ Walkley Black method⁽⁷⁾ and Kjeldahl method.⁽⁸⁾

Zn-contaminated soil and planting preparation

Soil preparation

Four compositions of soil were prepared with ZnSO₄·7H₂O (0, 10, 100 and 1,000 mg) added and 7 replicates were done for each composition and each species. In brief, there were 28 units for each plant species, and 56 units overall.

Plant preparation

20 seeds of each plant were planted into each soil pot and then watered once a day in the morning (before 10 am) with the amount of water that did not exceed the water holding capacity of the soil.

Zn in soil and plants analysis

Both crops were harvested 46 days after planting by separating soil from the plants.

Soil analysis

Soil was spreaded out and air-dried, then sieved through a 2 mm sieve. Zn was extracted from the soil with 0.1 N HCl. An atomic absorption spectrophotometer (AAS) was used to analyze the amount of Zn.

Plant analysis

Each plant was cut into shoots and roots, weighed and rinsed with tap water followed by distilled water, then put in a 65-80°C oven for 24-72 hours. Shoot and root were weighed, separately. Zn was extracted from the plant with 0.1 N HCl and an atomic absorption spectrophotometer (AAS) was used to analyze the amount of Zn.

Statistical analysis

One-way ANOVA (SPSS programme for Windows) at 95% significance was used to analyze Zn in soil, root and shoot of both crops.

RESULTS AND DISCUSSION

Soil Properties

The physical and chemical properties of soil influence the behavior of metals in soil and plant growth. The soil properties are shown in Table 1.

Texture is an important characteristic of soil, affecting drainage conditions, water holding capacity, amount and size of pores and plant root development. Consequently, the rate of water intake, aeration and soil fertility

are all influenced by soil texture. The soil type used in this study was clay.

pH has a great effect on dissolution and precipitation of zinc. At low pH values, the solubility of zinc is high and as the pH is raised (pH > 7), zinc availability to plants decreases. As the pH is increased, the ionic form of zinc is changed to the insoluble hydroxide.⁽⁹⁾ As pH 6.21 zinc (II) ion added to the soil does not precipitate with other compounds such as hydroxide and carbonate, which plant cannot use.

Cation Exchange Capacity (CEC) shows the ability in absorbing cations (heavy metals) and also the ability to exchange these cations with soil solution, which are the form that plants can utilize. The CEC of soil used in this experiment was quite high (40 cmol_c/kg). Therefore, it can be inferred

that zinc ions added to the soil would not be easily leached but will be adsorbed by soil colloids.

Organic matter is the source of plant nutrients, especially of N and S, in soils. The organic matter content of soil used in this study was quite high, showing the fertility of the soil.

Nitrogen is an essential nutrient for plant growth. The total N content is found to be a function of soil organic matter content. Plants satisfy their nitrogen requirement from the inorganic fraction. The organic fraction serves as a reserve of nitrogen in plant nutrition and will be released after decomposition and mineralization of organic matter. The N content of the Ap horizons of most cultivated soils is estimated to be in the range between 0.05-0.4% N.⁽⁵⁾ The total N content of soil used in this experiment was slightly low.

Table 1. Some physical and chemical properties of soil.

Parameters	
Soil texture	Clay (clay 62.12 %, silt 15.57 %, sand 22.31 %)
Moisture content	18.02 %
Soil pH	6.21
Cation Exchange Capacity	40 cmol _c /kg
Organic matter	6.20 %
Total nitrogen	0.114 %

Plant Growth in soil with different Zn concentrations

The growth of *B. juncea* and *B. chinensis* were different in each composition. Both species grew very well in soil with 10 mg of Zn added. Chlorosis, small and dry plants were

sometimes found in the fourth composition of the experiment, which was the soil with 1,000 mg of Zn added. Plants were harvested and observed morphologically after 46 days.

Each *B. juncea* had 4-5 leaves. They had some wavy edge leaves, small purple shoots and many branch roots as a distinguished form. For *B. chinensis*, they had smaller leaves than *B. juncea*, when comparing size of leaf within the same Zn concentration. The size of both species in every composition were smaller than those in the market. Planting without adding fertilizer and the unsuitable environmental condition might be the cause of this.

Zinc Accumulated in Plants

For *B. juncea* and *B. chinensis*, there was no significant difference in zinc accumulation between 0 and 10 mg Zn added soil (Figure 1). The amount of zinc in the soil used in this study is approximately 10 mg/kg. Generally, the level of zinc in soil that is not toxic to plants and the critical level of zinc in soil is in the range of 3-50 and 300-500 ppm.⁽¹⁰⁾

It can be concluded from the result of this experiment that 1 kg of *B. chinensis* can accumulate zinc significantly better than *B. juncea* in the same weight when these plants are grown in the 0, 10, 100 mg Zn added soil, but *B. juncea* can accumulate more zinc when planted in the 1,000 mg Zn added soil.

The results showed that the species of plants can accumulate zinc differently, depending on the concentration of zinc in the contaminated soil.

B. juncea accumulated most zinc (4,825 mg/kg dry wt. plant) when 1,000 mg of zinc was added, while *B. chinensis* accumulated most zinc (4,766 mg/kg dry wt. plant) when 100 mg of zinc was added. Therefore, it can be assumed that *B. juncea* can grow and accumulate this heavy metal in zinc-rich soil (1,000 mg zinc-added soil) better than *B. chinensis*.

Since Reeves, and Baker,⁽¹¹⁾ stated that, for phytoremediation purposes, there should be interest in any species that consistently shows zinc levels above 3,000 mg/kg, *B. chinensis* and *B. juncea* can be considered as zinc-accumulation plants.

Ebbs, and Kochian,⁽¹²⁾ suggested that Indian mustard (*Brassica juncea*) has been shown to be effective in phytoextracting Zn, particularly after the synthetic chelate EDTA has been applied to the soil. A hydroponic experiment comparing two kinds of grasses (*Avena sativa* and *Hordeum vulgare*) to Indian mustard indicated that, although shoot Zn concentrations were greater for Indian mustard, the grasses were considerably more tolerant.⁽¹³⁾ reported that *Brassica juncea* (L.) Czern., *B. napus* L., and *B. rapa* L. were the most effective in removing Zn from the contaminated soil. When the soil was supplemented with Gro-Power, a commercial soil Supplement, removal of Zn by plant shoots doubled to more than 30,000 mg Zn/pot (4.5 kg).

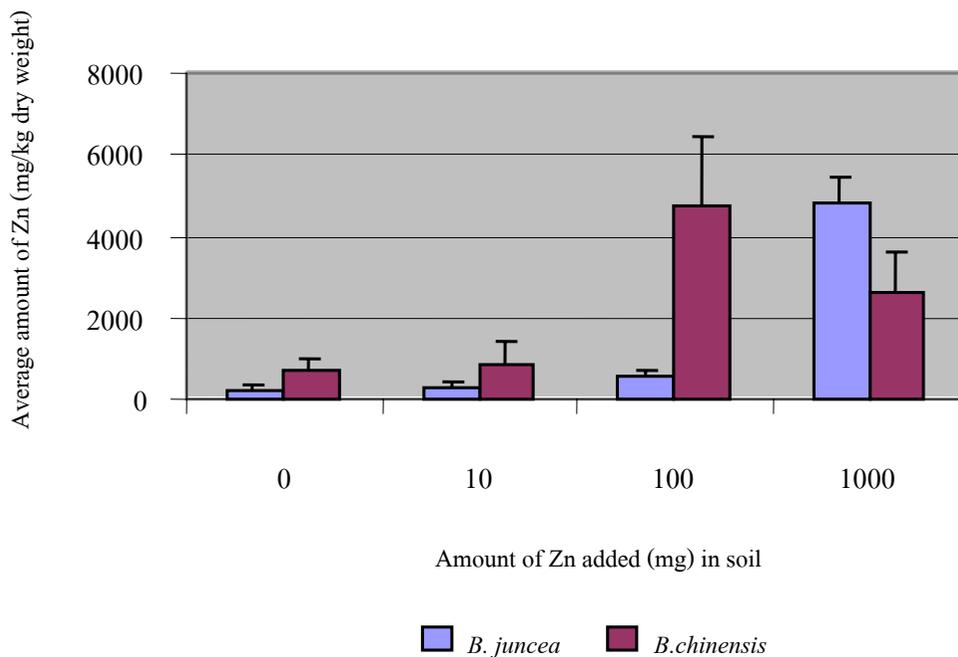


Figure 1. Average amount of Zn in *B. juncea* and *B. chinensis* (mg/kg dry weight).

Zinc Accumulated in Shoots and Roots

B. juncea

From the experiment, 1 kg of shoots of *B. juncea* can accumulate less Zn than the root of the same weight when grown in soil with 0, 10 and 100 mg of Zn added (Figure 2). On the other hand, shoots of *B. juncea* can accumulate more Zn than the root

when grown in soil with 1,000 mg of Zn added.

The statistical analysis showed that the difference of Zn accumulation efficiency between shoots and roots of *B. juncea* were not significant in soil with 0, 10 and 100 mg of Zn added.

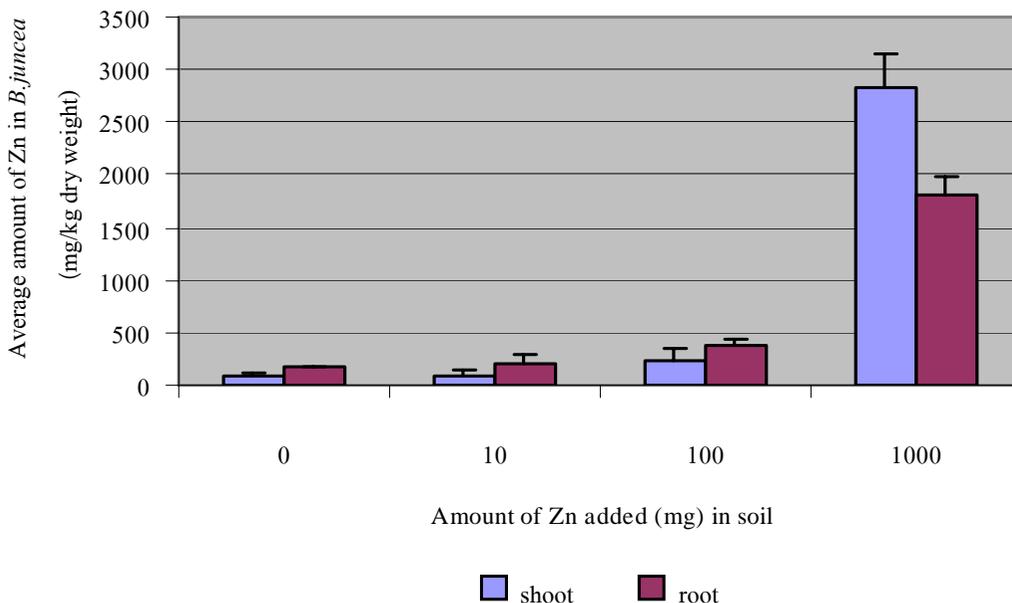


Figure 2. Average amount of Zn in shoots and roots of *B. juncea* (mg/kg dry weight).

B. chinensis

The experiment revealed that for every composition, 1 kg of shoots of *B. chinensis* can accumulate more Zn than the roots (Figure 3). The statistical analysis indicated that in soil with 10, 100 and 1,000 mg of Zn added, shoots of *B. chinensis* tended to accumulate more Zn than the roots.

The highest levels of zinc accumulated in *B. juncea* and *B. chinensis* were 2,815 and 4,178 mg/kg (or µg/g) in their shoots.

Hyperaccumulators have the ability to accumulate large amounts of contaminants by translocating metals from roots to shoots in the plant. A nonaccumulator plant species

normally absorbs 20-100 µg/g zinc while *Thlaspi caerulescens* has been shown to accumulate up to 40,000 µg/g zinc in their shoots.⁽¹⁴⁾

Salt, and Kramer,⁽¹⁵⁾ suggested that the translocation of metal cations from roots to shoots is governed by two main processes: movement into the xylem and volumn flux through the xylem; the latter is mediated by root pressure and transpiration. The change of the amount of zinc up taken by *B. juncea* may affect these processes while the change may affect *B. chinensis* differently due to the dissimilarity of their physiology.

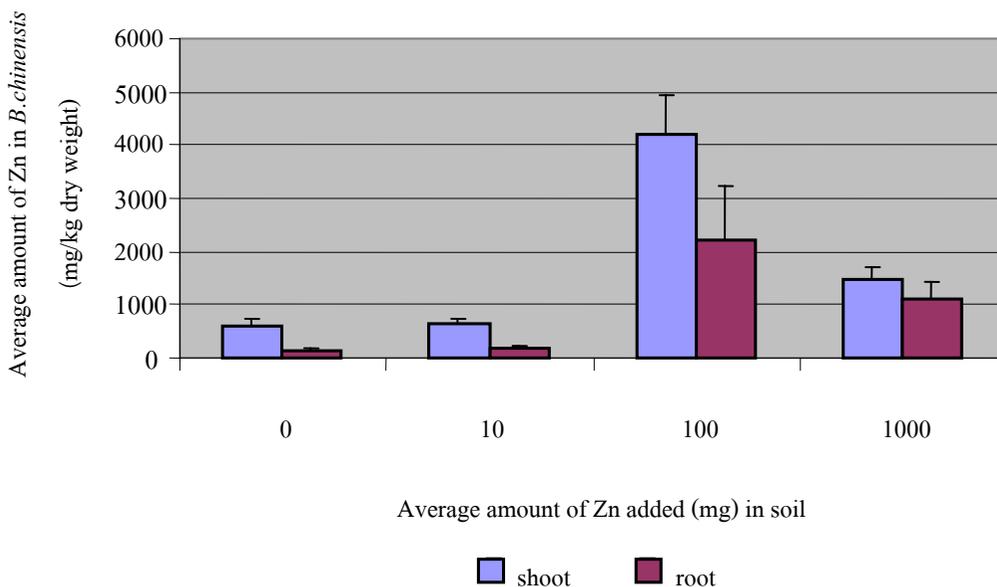


Figure 3. Average amount of Zn in shoots and roots of *B. chinensis* (mg/kg dry weight).

CONCLUSION

B. chinensis has higher efficiency in accumulating zinc than *B. juncea* in the low zinc-concentration conditions but *B. juncea* has higher efficiency in the high zinc-concentration condition. *B. juncea* accumulated most zinc (4,825 mg/kg dry wt. plant) when 1,000 mg of zinc was added, while *B. chinensis* accumulated most zinc (4,766 mg/kg dry wt. plant) when 100 mg of zinc was added.

The plant selection that will be appropriate for phytoremediation should depend not only on the nature

of the soil and the concentration of zinc in the soil, but also on the metal-accumulating efficiency of each type of plant.

It can be concluded that *B. chinensis* and *B. juncea* are interesting species for phytoremediation. Because both species have a short crop period and are easily found in Thailand, they could be used to reduce zinc contamination. However, *B. chinensis* will be suitable for zinc removal in lower level contamination sites, while *B. juncea* will tolerate high level contamination areas.

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