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The effect of Cu concentration in soil and phosphorous fertilizer on plant growth and Cu uptake by *Brassia juncea L.* grown on contaminated soils

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Abstract. An experiment was carried out in the greenhouse conditions with *Brassica juncea L.* grown on alluvial soils that had previously been contaminated at different concentrations of Cu. The main purposes of the research were to determine the effects of Cu and phosphorus applications on plant growth and Cu uptake by *Brassica juncea L.* Mature plants were harvested for the Cu accumulation analysis. The soil samples from each growing pot were extracted by HNO_3 0.43N in order to determine the content of Cu^{2+} mobilization in soil, while the plant samples were acid digested for determining the total Cu concentration. Atomic Absorption Spectroscopy (AAS) was employed to determine Cu concentrations in soils and plant samples. The results showed that adding Cu to soils has strong effects on *Brassica juncea L.* growth and the uptake rate of Cu by the plants. The height and the biomass of plants were reduced dramatically by 36% and 53% respectively at the rate of 200 ppm Cu. In addition, phosphorous fertilizer also effectively improved plant growth and reduced Cu concentrations in plant of *Brassica juncea*. At the application rate of 100 kg P_2O_5/ha, the height and biomass of plant were increased to 30% and 31% respectively, and the Cu content in plants of *Brassica juncea* was reduced by 14% comparing with the control samples.

Keywords: Cu in soil; Phosphorous fertilizer; Cu uptake.

1. Introduction

Soil pollution by heavy metals is a serious problem that can have affect on plant growth and human health. The contaminants of majors concern (Cu^{2+}, Zn^{2+}, As^{2+}, Cd^{2+} and Pb^{2+}) arise from number of industrial, mining and agricultural activities. The high concentration of heavy metals in soil is reflected by higher concentration of heavy metals in plants and, consequently, in animal and human bodies [2]. Therefore, plants usually have been used as indicators of metal pollution or accumulation in soil. Besides, plants are also used as accumulators for soil remediation, which is called phyto-remediation [5, 7]. The base of phyto-remediation is pollutant uptake or bounding by plants [7]. Other possibility to decrease available concentration of pollutants is stabilization. Phytostabilization can gain results from either physical or chemical effects of plants, and of chemicals, such as phosphate, lime or clay minerals [3].

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The examination of the heavy metal, i.e. Cd and Zn on plant growth and uptake investigated by Zhang et al., also assist to elaborate a possible combination of phosphate application on heavy metal uptake [1, 4]. The main purpose of this research is to investigate the relation between contents of Cu in soil and Cu accumulation in plant, and also the role of phosphate in reducing the uptake of Cu by *Brassica juncea L.*

2. Materials and methods

2.1. Pot experiments

Alluvial soils collected for pot studies originated (0-20 cm depth) from Quynh Do Village (Thanh Tri District, Hanoi City) where the soil is affected by waste water from Hanoi City during agricultural production. The chemical properties of soil for pot studies are: CEC: 23 Cmol/kg, pH (KCl): 6.15, OM: 2.44%, total N: 0.32%, total P_2O_5: 0.19%, total K_2O: 1.02%, total Cu: 21.29 ppm, mobilization Cu^{2+} (HNO_3 0.43N): 13.38 ppm.

The greenhouse experiment was carried out in the 2007 year. The soil used for experiment was artificially polluted by $CuSO_4$ with significantly different rates of Cu application (Table 1). Each pot with 5 kg of soil was sown with seeds of *Brassica juncea L.* and water to the moisture level of about 70-80% of the field capacity.

Table 1. The treatments layout

Treatments	Fertilizers ($N+K_2O$) kg/ha	Cu and phosphate added Cu (ppm)	P_2O_5 kg/ha
Trt.1	(75+30)	0	0
Trt.2	(75+30)	50	0
Trt.3	{75+30)	100	0
Trt.4	(75+30)	200	0
Trt.5	(75+30)	100	40
Trt.6	(75+30)	100	60
Trt.7	(75+30)	100	80

2.2. Sampling and chemical analysis

Plant and soil samples were taken and analysed at the harvest time (45 days after sowing). Plant samples (leaves and shoots) are collected and washed with pure water and then dried at 70°C until stabilisation of weight. The monitoring indicators for plants growth include plant height and biomass. Total Cu in soil and plant tissues, Cu^{2+} (HNO_3 0.43N) in soil was determined by Atomic Absorption Spectroscopy (AAS).

3. Results and discussion

3.1. Effects of Cu application on plan growth and Cu accumulation in plant

The results of the effects of added Cu on plant growth and heavy metal accumulation in *Brassica juncea L.* plants are presented in Table 2. Some heavy metals, e.g. Cu, at a low dose are essential microelement for plants, but in higher doses they may cause metabolic disorders and growth inhibition for most of plant species.

Table 2. Effects of added Cu on plant growth and Cu content in plants (fresh weight)

Treatments	Plant height		Plant weight		Cu accumulation in plant	
	cm	%	g/pot	%	ppm	%
Trt.1	19.5	100	70.3	100	2.8	100
Trt.2	15.0	77	55.9	80	3.2	115
Trt.3	14.7	75	50.0	71	5.2	190
Trt.4	12.5	64	33.1	47	8.5	308

The data in Table 2 show the effects of Cu concentration in soil on the growth rate of *Brassica juncea L.* The height of plants reaches the highest value of 19.5 cm at Trt.1, and drops dramatically to 12.5 cm at Trt.4 (drop by 36%) following the rate of added Cu increasing to 200 ppm. The effects of added Cu on biomass are the same way of the effects on the height of plants. The biomass decrease by 53% at the application rate of 200 ppm Cu (Trt.4) compared to the control sample (Trt.1). It is clear that there is a reduction of plant growth (plant height and biomass of *Brassica juncea L.*) with increasing concentration of Cu in soil. The accumulation of heavy metal in plant tissues of *Brassica juncea L.* is reflected the concentration in soil. The contents of Cu in plant increase upon the rates of Cu application, especially at the high rate of Cu above 100 ppm. It can be said that the increase of rate of Cu uptake and accumulation in plants is much faster than the decrease of rate of plant growth. For example, the contents of Cu in plant increase by 208% when the rate of Cu application increases to 200 ppm. Meanwhile, the rate of biomass decreases only by 53%. The results showed that the uptake of Cu by *Brassica juncea L.* plants corresponded to the increasing level of Cu contamination of soil, while the biomass was reduced at the high level of Cu concentration.

3.2. Relationship between content of Cu^{2+} in soil with plant growth and Cu accumulation in plant

The relationship between Cu concentration in soil and biomass of *Brassica juncea* is presented in Table 3. The highest plant height and biomass is recognized at Trt.1 where the mobilization of Cu^{2+} (HNO_3 0.43 N) is about 13 ppm. But with a higher level of Cu^{2+} in soil, the growth rate of *Brassica juncea* decreases significantly. This trend may be explained by the toxicity of Cu to plant. These results might be also used to evaluate the level of Cu pollution in soil. Based on this experiment, one can suggest that the phyto-toxic threshold of Cu^{2+} mobilization to *Brassica juncea* might be listed at around 30 ppm, and serious effects can be seen with the content higher than 50 ppm. The relationship between Cu^{2+} concentration in soil and Cu content in plant shows a significant positive effect (Fig. 1).

Table 3. The contents of Cu^{2+} in soil, Cu content in plant and plant height (fresh weight)

Trt	Cu^{2+} in soil (ppm)	Plant height (cm)	Biomass (g/pot)	Cu content in plant (ppm)
Trt.1	13.4	19.5	70.3	2.8
Trt.2	15.5	15.0	55.9	3.2
Trt.3	27.2	14.7	50.0	5.2
Trt.4	51.1	12.5	33.1	8.5

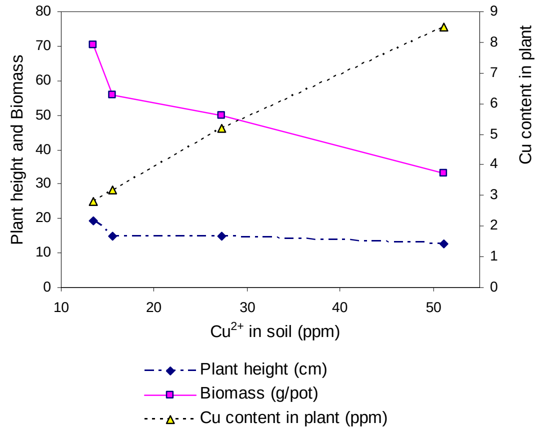

Fig. 1. The relationship between Cu^{2+} in soil with plant height (Y_1), biomass (Y_2) and Cu content in plant (Y_3).

3.3. Effect of phosphate fertilizer on plant growth and Cu accumulation in plant

In contrast with the results of Cu application, phosphate fertilizers have a positive effect on growth of *Brassica juncea* at all application rates. The plant height and biomass of *Brassica juncea* increase up to 130% and 131% respectively comparing to the control sample (Trt.3) without phosphate fertilizer (Table 4).

Table 4. The effects of added phosphorus on plant growth and Cu accumulation in plants of *Brassica juncea* (fresh weight)

Treatment	Plant height		Plant weight		Cu content in plant	
	cm	%	g/pot	%	ppm	%
Trt.3	14.7	100	50.0	100	5.2	100
Trt.5	16.3	114	53.1	106	5.1	98
Trt.6	17.4	118	57.3	115	4.8	92
Trt.7	19.1	130	65.5	131	4.5	86

The data in Table 4 show that phosphate fertiliser not only improves the plant growth, but also is the factor influencing on heavy metal uptake by plant. The positive effects of phosphate fertilizer on reducing Cu accumulation in plant were recognized at all rates of phosphate application. However, this effect is significant only at rates greater than 60 kg P_2O_5/ha. At 100 kg P_2O_5/ha rate, the content of Cu in plants of *Brassica juncea* is reduced by 14% compared to the control sample.

In general, there is a reduction in growth rate of *Brassica juncea* when the content of Cu in soil increases whereas the content of Cu in plant decreases (Fig. 2). The data found in this study indicated that Cu^{2+} concentration in soil has strong effects on the growth of *Brassica juncea* even at moderate concentration of about 30 ppm.

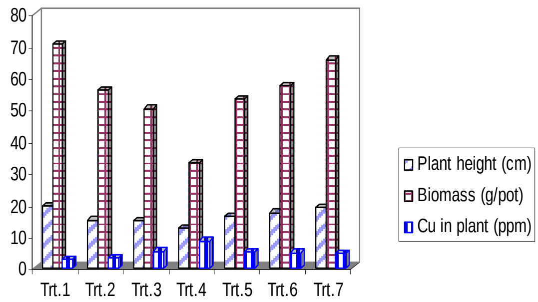

Fig. 2. Effects of Cu and phosphate fertilizer on plant growth and Cu accumulation in plant of Brassica.

4. Conclusions

The growth rate of *Brassica juncea* is significantly affected by the Cu^{2+} concentration in soil at 30 ppm, and strongly affected by the concentration at about 50 ppm which is equivalent to the application rate of 200 ppm Cu. The height and the biomass of plants are reduced dramatically by 36% and 53% respectively when the rate of added Cu increases to 200 ppm. However, a further study is needed in order to establish the maximum amount of Cu that the plants to grow in these soils.

In addition, a significant reduction of Cu content in *Brassica juncea* is found when phosphorous fertilizer application at the rate higher than 80kg P_2O_5/ha. At the application rate of 80kg P_2O_5/ha, the height and biomass of plant are increased by 30% and 31% respectively, and the content of Cu in plant is reduced by 14% compared to the control sample.

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