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Effect of cadmium on growth of rice (*Oryza sativa* L) cultivar sokan originated from West Sumatra, Indonesia

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Abstract

Oriza sativa L sokan rice is the most famous of rice in West Sumatra, Indonesia. Cadmium accumulation and toxicity in Oriza sativa L sokan were characterized and identified. The low level of Cadmium concentration did not affect the growth of rice plant investigated, however the growth especially number of tiller increased at Cd level 15 μmol/L. Cd uptake by rice plant was found to be higher in root than in leaf and seed. This result indicated that root can be the first barrier of Cd uptake by rice plant.

Keywords: *Oriza sativa L sokan, cadmium, cadmium uptake, rice plant growth*

Introduction

Pollution of the natural environment by heavy metals has become a serious problem in some industrialised countries. The release of large quantities of heavy metals from industries into the environment has resulted in a number of environmental problems[1,2].

Heavy metal such as lead, cadmium and cobalt from the anthropogenic sources, metal plating, mining operations and other industries are among the most common pollutants found in industrial effluents, and become an environmental problem of worldwide concern including Indonesia[3,4]. For example the pollution of cadmium in Nigata prefecture, Japan, in the middle 1950s was caused by the long-term consumption of rice and soybean contaminated with cadmium, which originated from mineral refinery factories[5].

Currently, toxic effect of heavy metals on crop plants is receiving considerable attention[6-10]. Many studies have shown that plant crop species differ markedly in their ability to absorb heavy metals. Even within the same species, there are great genotypic differences in the effect of heavy metals on plant growth [11].

Hernandez et al.[6] reported that exposure to cadmium could cause perturbations in various plant processes such as plant growth and nitrogen assimilation.

The wide spread use of pesticides and chemical fertilizers, especially phosphate fertilizers, has led to the cadmium contamination in soil and crops. Cadmium is strongly phytotoxic and causes growth inhibition and even plant death due to its detrimental effect on many physiological processes. After absorption and accumulation by crops, cadmium can enter into food chains and pose risks to human beings. Toxicity of cadmium also causes oxidative stress, which can take place possibly by generating reactive-oxygen species (ROS) such as superoxide radicals, hydrogen peroxide (H_2O_2) and hydroxyl radicals. These oxygen species cause lipid peroxidation, which is reflected by increased malondialdehyde (MDA) concentration [12].

Rice is one of the most important crops in the world and the main staple food in Indonesia. Many studies have been conducted related to the effect of toxic heavy metals on photosynthesis, growth and yield of cultivated rice [13.17]. Unfortunately, there are very few reports on the effect on cadmium accumulation on growth of rice *Oryza sativa L* cultivar sokan originated from West Sumatra, Indonesia.

Materials and Methods

Reagents and equipment

All reagent used in the experiment i.e. $CdCl_2$, HNO_3, $HClO_4$, were of Analytical Grade and obtained from Wako, Japan. Deionized water was prepared in the laboratory. Metal concentration has been measured using Atomic Absorption Spectrophotometric (Rayleigh WFX 320 Beijing, China).

Soil and seed preparation

Soil samples were collected from surface layer (0-20 cm) of paddy field located in Sungai Sapih, Padang, West Sumatra, Indonesia. The soil samples were air-dried prior to use at the greenhouse pot experiment at Andalas University. The seed of rice (*Oriza Sativa L*) cultivar sokan was collected from West Sumatra, Indonesia. Seeds were surface washed with deionized water three times.

Plant growth

Sterilized seeds were shaken with deionized water for 48 hours, then transferred to filter paper and allowed to grow for 4 weeks until 3-4-tiller stage. The seedling growth environment was controlled at room temperature 28-29^{o}C. The tiller was then transferred into the pot experiment (20 x 30 x 22 cm) at the green house. Each pot was flooded with water up to 2 cm above surface of soil for 1 week to generate water logged conditions. Then cadmium solution of varied concentration and pH was added to the pot experiment up to 8 weeks.

Cadmium analysis

At harvest, the rice root, leaf and seed were collected. The collected samples were washed with deionized water several times. The sample was then dried at temperature 80^{o}C, until constant weight was obtained. The sample was then were ground into fine powder. 1 g of each sample was taken into a digestion tube, then digested with 5 mL nitric acid and perchloric acid (3:1 v/v). The sample was boiled at 90°C till all the sample dissolved. The solution was filtered and then ultrapure water was added to the filtrate to make the volume up to 50 mL.

Results and Discussion

Effect of solution of pH on Oriza sativa L cultivar sokan plant growth

The effect of pH on *Orizae sativa* L. cultivar sokan is shown in Figure 1. As shown in Figure 1 on increasing pH of 15 ìmol/L cadmium solution from 4.0 to 8.0, the plant died at pH higher than 7.5, as compared with blank and pH solution of 4.0. The yield response of plant growth to a variety of solutions of different pH was significantly different compared with control. Therefore, the plant growth and development appear to be pH solution dependent. At the constant solution of pH 4, the plant growth much depends on concentration of Cadmium. As show in Figure 2, the number of rice tillers from first week until seven weeks much depends on cadmium concentration. Table 1 shows that after 7 weeks of plant rice growth, the number of rice plant tillers reach maximum, ie. 22 at cadmium concentration 15 µmol/L and mostly died at cadmium concentration 20 µmol/L.

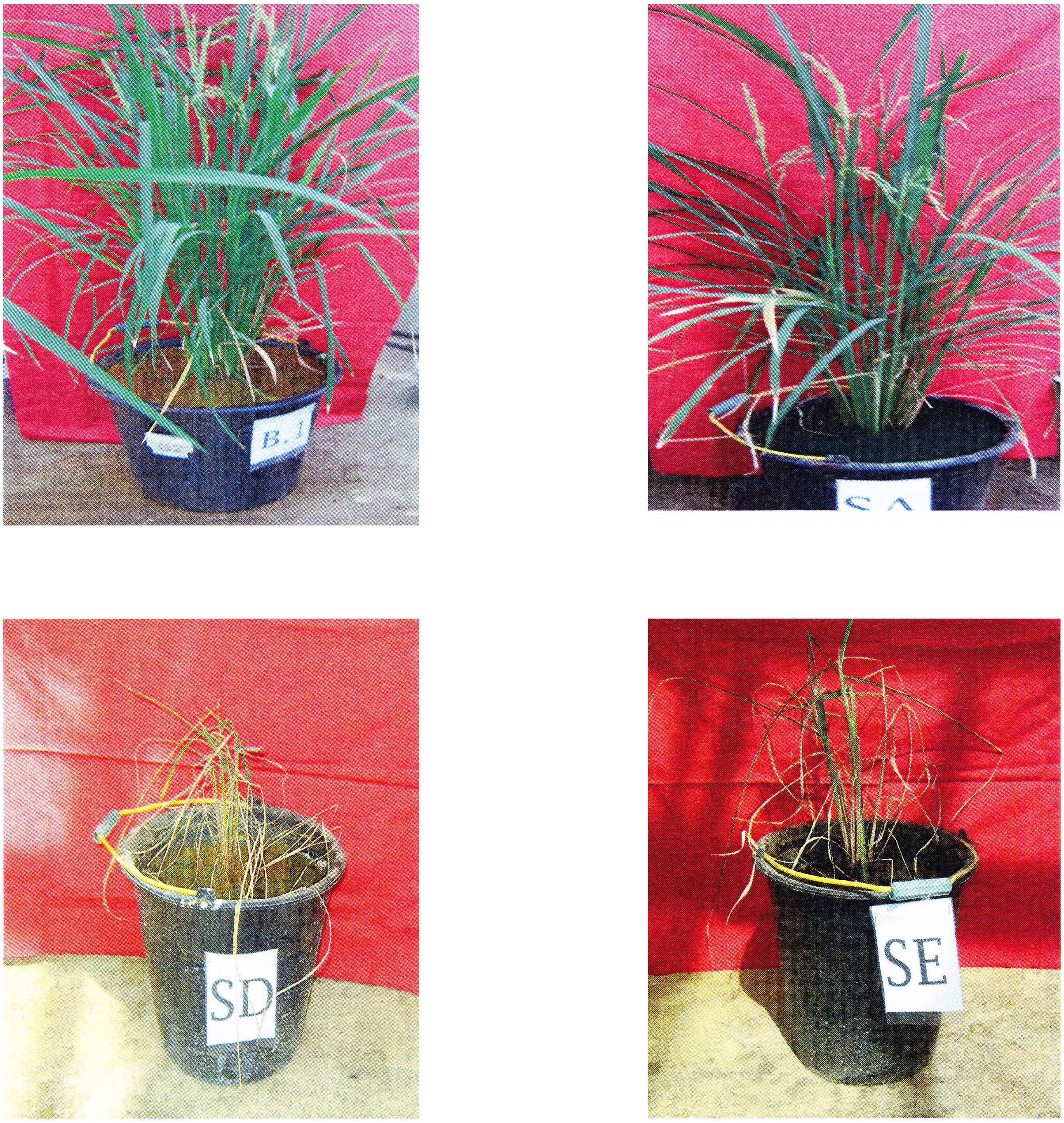

Fig. 1. Effect of solution of pH on Oriza sativa L cultivar sokan growth.
A = Control. Cadmium concentration added 15 μmol/L at solution of pH 4.0 (B), 7.5 (C) and 8.0 (D)

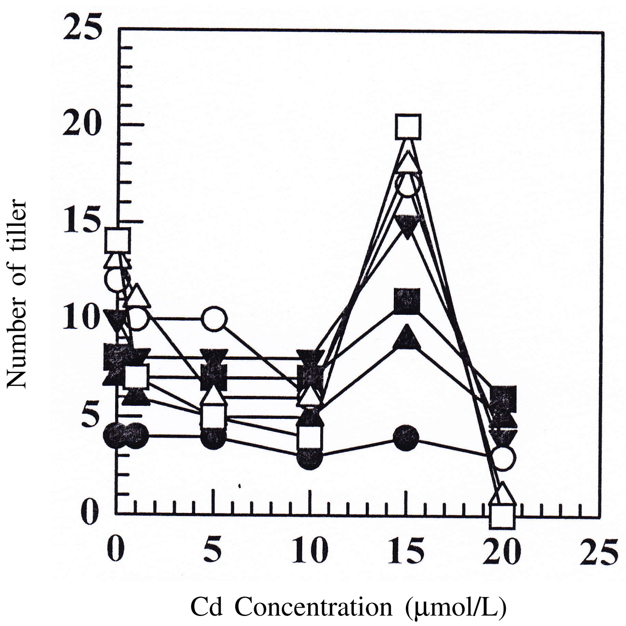

Fig. 2. Effect of Cadmium concentration on number of rice tillers of Oriza sativa L cultivar sokan. Solution of pH = 4.●= first week, ▲= 2nd week. ■ = 3rd week, ▼= 4th week, O: 5th week, △= 6th week and □= 7th week.

Table 1: Effect of cadmium concentration on number of rice plant tiller at pH 4.0

[Cd] μmol/L	No. of Rice Tiller						
	1st of Week	2nd of Week	3rd of Week	4th of Week	5th of Week	6th of Week	7th of Week
0	4	7	8	10	12	13	14
1	4	6	7	8	10	11	7
5	4	5	7	8	10	6	5
10	3	5	7	8	6	6	4
15	4	9	11	15	17	18	22
20	3	5	6	4	3	1	0

Cadmium can be directly taken from soil to the plant by efflux system. The mechanism of Cadmium detoxification in living cell plant exploit a variety processes, such as enzyme activity [18], compartmentalization by chelation to sulfhydryl group compounds such as cysteine, reduced glutathione and phytochelatin[19]. Phytochelatin, synthesized from glutathione is effective chelator for Cadmium ion in plant [20]. Phytochelatin is also known as sequester for cadmium by forming phytochelatin-cadmium complex.

Effect of Cadmium concentration on Cd uptake by Oriza Sativa L. cutivar sokan

The main source of cadmium intake is rice, especially for rice eating countries such as Indonesia. Approximately 50% of the daily intake of Indonesian comes from rice[21]. Therefore Cadmium contaminates and accumulates in agricultural products such as rice through water and soil pollution if waste discharge is not properly treated. Further greater use of fertilizer containing Cadmium increases the cadmium content in plants especially rice. The joint FAO/WHO expert committee on food additives (JCEFA) has proposed a maximum level of 0.2 mg/kg cadmium in rice. Accumulation of Cd in rice plant growth could be through numerous functional groups namely, amino, thiol, carboxyl, sulphydryl, carbonyl, hydroxide, imidazole and amide moieties[18], which have been suggested as the possible functionalities groups responsible for the binding of metal ions. The effect of Cadmium concentration on Cd uptake by *Oriza sativa* L cultivar sokan can be seen in Figure 3. As shown in Figure 3, Cadmium uptake by soil and rice root increases with increasing concentration of Cadmium, whereas the concentration of Cadmium on leaf and seed appears to be constant and slightly increases at concentration 15 μmol/L. Cadmium is highly soluble in the soil solution and can therefore easily transferred from soil to plant through root and accumulate therein. Figure 3 show that a first barrier against Cadmium uptake lies

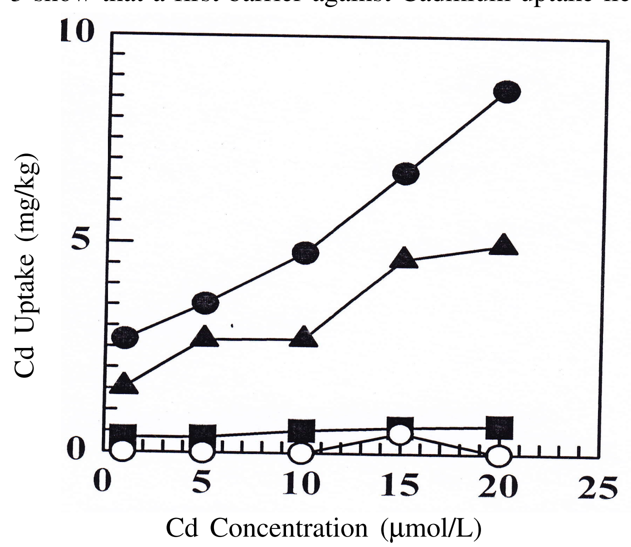

Fig. 3. Effect of Cadmium concentration on Cd uptake by Oriza sativa L cultivar sokan. Solution of pH=4, ●= soil, ▲=root, ■ = leaf and O=seed

mainly in root level. The root cell wall not only accumulates a high level of Cadmium but has to a certain extent an impact on Cadmium tolerance in other plants. This result is shown in Table 2.

Table 2: Effect of cadmium concentration added on the soil water on the level of cadmium present in soiland rice plant at pH 4.0

[Cd] added µmol/L	Level of Cadmium present, mg/kg			
	Soil	Root	Leaf	Seed
1	2.67	1.52	0.35	nd
5	3.53	2.67	0.37	nd
10	4.77	2.70	0.53	nd
15	6.70	4.64	0.63	0.49
20	8.72	4.99	0.67	nd

0.49 mg/kg cadmium was detected on the rice seed when 15 µmol/L was added into the soil. Moreover no cadmium has been detected at level 20 µmol/L, because the rice plant died when high level concentration of cadmium was present in the soil.

Effect of solution of pH on Cadmium uptake

The effect of solution of pH of 15 µmol/L Cadmium was investigated and the result can be seen in Figure 4. It is clear that pH of Cadmium solution could affect the cadmium uptake by soil, root, and leaf. Further more Cadmium content in the seed remains constant at pH 4.0 to 8.0. Munaf et al.[23] and Zein at al.[24] reported that the uptake capacities generally demonstrated a similar trend using no living cell of plant. The decreasing uptake of Cadmium ion by rice plant at pH higher than 4.0 may be attributed to the precipitation of insoluble cadmium species. The fact that the metals are present in their ionic state at a low pH (eg. 4.0) implies that the functional groups on the cell wall of the rice plant may also compete with other metals ion present in the soil. The data shows in Table 3 indicates that in solutions with pH from 4.0 to 8.0 cadmium concentration was decreased from soil, root, leaf and seed. Moreover at solution with pH 4.5, cadmium was found in seed at concentration 0.17 mg/kg. For most of the living cells, the sorption of heavy metals occurred due to enzyme activities. However the true mechanism of sorption process is still being investigated by many authors.

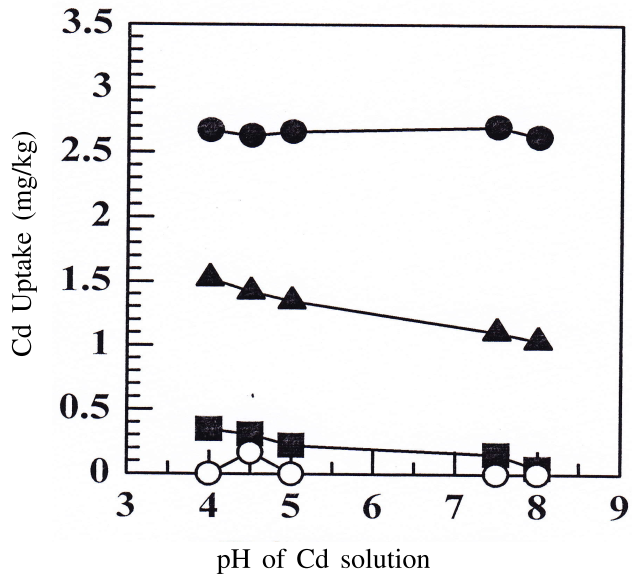

Fig. 4. Effect of pH of Cd solution on Cd uptake by Oriza sativa L cultivar sokan.
Cd concentration = 15 µmol/L, growth in 7 weeks,
●= soil, ▲= root, ■ = leaf and O = seed

Effect of Cadmium concentration of chlorophyll content of Oriza Sativa L cultivar sokan

The effect of Cadmium concentration on chlorophyll content of Oriza Sativa L cultivar sokan is shown in Figure 5. In rice plant, the most apparent symptom of Cadmium toxicity is chlorosis of the leaves. When rice plant was treated with Cadmium concentration from 1 to 20 µmol/L for 7 weeks, chlorophyll content observed in leaf increased and become maximum at Cadmium level 15 µmol/L. At 20 µmol/L of Cadmium there was decrease in

Table 3: Effect of solution of pH on cadmium uptake by *Oriza sativa,* L cv sokan. Cadmium concentration added 1 µmol/L

Solution of pH	Cadmium uptake, mg/kg			
	Soil	Root	Leaf	Seed
4.0	2.67	1.52	0.35	nd
4.5	2.63	1.42	0.31	0.17
5.0	2.66	1.35	0.22	nd
7.5	2.70	1.1	0.15	nd
8.0	2.62	1.06	0.48	nd

chlorophyll and proteins content in the leaf. Therefore it is clear that Cadmium could decrease the chlorophyll content in rice leaf.

Conclusions

The *Oriza sativa.* L cultivar sokan investigated shows similar effect of cadmium toxicity as compared with other varieties of rice.

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