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THE GROWTH AND CHROMIUM CONTENT OF BANANA PLANT (*Musa acuminata*) CV. AMBON LUMUT IN CHROMIUM STRESS CONDITION

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ABSTRACT

The plants that live in polluted Chromium (Cr) areas can experience a variety of disorders in the body. The disruption caused by Cr can cause damage DNA, proteins, photosynthetic pigments, and chloroplast ultrastructure of cell membranes. It can inhibit germination, photosynthesis, respiration, cell division and growth, resulting in lower yields. To find out how Cr stress influences to banana plants {Musa acuminata) cv. Ambon Lumut, the present research was conducted on the growth and content of Cr in banana plants. In this research, bananas was grown in-vitro with Cr concentrations of 0, 50, 100, 200 and 400 ppm in the medium. The results showed that Cr influenced of the growth and content of Cr in banana plants {Musa acuminata) cv. Ambon Lumut. It is likely that the greater the concentration of Cr in the medium was, the smaller the increase in weight was. Root-shoot ratio and Cr content in root and shoot tends to be greater by increasing Cr concentration in the medium.

Keywords: Cr content, Cr stress, growth, Musa acuminata.

INTRODUCTION

The present climate change is shown by various indicators, among others, higher temperatures, caused by the increase of carbon dioxide concentrations. Actually, the increase in carbon dioxide could be useful if the number of plants that live around us is sufficient for use in the process of photosynthesis. However, the reality indicates the number of plants is increasingly reduced, due to uncontrolled logging. In addition, a lot of hazardous waste dumped into the environment causes many plant species intolerant to the death. This waste is one of the heavy metals chromium (Cr), which can take a part in reducing the number of plants.

Chromium in the environment of plants comes from a variety of sources, both from natural sources and industrial waste. Tanning industry is one industry that uses material tanner containing heavy metals Cr. Waste is directly dumped without prior treatment and will certainly lead to contamination of water and soil in the vicinity. The content of Cr in the contaminated soil reaches an average of 200 ppm, even more than 200 ppm (Ririn, 2009; Diwan et a1., 2010), while the soil quality standard for the content of Cr, amounting to 76 ppm (US- EPA, 2004, in Andarani & Roosmini, 2010).

Chromium entering the body of plants can cause a variety of disorders. Toxic Effect of Cr in plants includes interference of the process of germination, growth of roots, stems and leaves that can impact the total dry matter production and yield. It also directly influences the enzymes or other metabolites (Shanker et al.,2005).

The results of previos studies showed that Cr may cause a reduction in the rate of photosynthesis, due to a reduction in the chlorophyll content of Vigna mungo (Hussain et al., 2006), Triticum aesticum (Subrahmanyam, 2008), Pisum sativum (Pandey et al , 2009), Brassica juncea (Ghani, 2011; Diwan et al., 2012), Sorghum bicolor (Revathi et at., 2011) and Hordeum vulgare (Yildiz et a1., 2012).

Such defects can be caused by Cr which damage DNA, proteins, photosynthetic pigments, and chloroplast ultrastructure of cell membranes, which finally inhibit germination, the process of photosynthesis, respiration, cell division and eventually disturbed the growth (Panda and Choudhury,2005).

According to Hossain et a1. (2012), in general heavy metal ions that exist in the root zone will compete with essential metal ions, which can lead to deficiency of essential metal ions and heavy metal ion excess. This will lead to heavy metal ions bind to proteins, DNA or other targets such as -SH groups and $-COO^-$ and can cause malfunctions of proteins and DNA or changes in enzyme activity. They ultimately lead to metabolic disorders such as inhibition of photosynthesis and plant respiration will lead to an increase in Reactive Oxygen Species (ROS) and cause oxidative stress.

Oxidative stress is caused by an increase in Reactive Oxygen Species (ROS), such as H_2O_2, O_2^-, OH^- (as a side product in a number of metabolic reactions in the cell organelles) exceeds the amount of antioxidants produced. Antioxidants can be either enzymes or non-enzymes. Antioxidant enzymes are catalase, glutathione reductase, ascorbate peroxidase and superoxide dismutase. Antioxidant non-enzymes are proline, ascorbic acid, glutathione, carotenoids and flavonoids (Azmat et al., 2009).

In normal conditions, the production of ROS in cells is small quantity and controlled. Heavy metal toxicity can increase ROS production more than 30-fold (Mittler, 2002). ROS react with a variety of fats, proteins, pigments, nucleic acids and cause lipid peroxidation, membrane damage and inactivity of enzymes, thus affecting cell viability (Diwan et al., 2010). One of the important processes is disrupted due to an increase in ROS is the reduction in the rate of photosynthesis in various plant species. This is because the chloroplast is an important target in oxidative stress (Pandey et a1., 2009).

Therefore, since the response of plants to heavy metals varies, then this research investigated how growth and Cr content of the banana plant *{Musa* spp) cv. Ambon Lumut in Cr stress condition.

MATERIAL AND METHODS

Banana plant in-vitro culture. In this research, the materials used were the plantlets of banana plants *[Musa acuminata)* cv. Ambon Lumut in-vitro culture. Plantlets were obtained through the stages of initiation and multiplication of shoot apex culture banana plants derived from Biogen Bogor. Completely randomized design (CRD) was employed in this experiment. Cr concentration in the growing medium, i.e. 0 ppm, 50 ppm, 100 ppm, 200 ppm and 400 ppm. The selection of concentrations was based on the concentrations commonly found in soil contaminated by Cr (Diwan et al., 2010). Sources of Cr used are $K_2Cr_2O_7$ added to the

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basic MS medium (Murashige and Skoog). Each treatment was repeated 10 times, so that the number of research units was 5 x 10 = 50 banana plants in a culture bottles. All cultures were maintained in an incubation chamber in 25-28 °C and in a TL 1000 Lux light irradiation for 12 hours per day. Maintenance was performed during 6 weeks.

Growth parameters. The increase in weight was the difference planlets before and after being treated by Cr on the growth medium. Root-shoot ratio was the ratio of dry weight of the roots of the banana plant with dry weight of the shoot. Dry weight was the weight obtained after the sample was dried in an oven in 65 °C± 2 °C for 72 hours.

Cr content of the banana plant. Cr content of the various organs of the banana plant (root and shoot) was measured by the method of concentrating the sample with concentrated nitric acid (HNO_3) at 300 °C for 1 hour (Liu et al., 2009). The dried plant samples as much as 2 grams was inserted into the crucible and added aquaregia cup as much as 20 mL (a mixture of 10 M HCl and 65% HNO_3 with a ratio of 3:1). The sample was then heated to dry in the hood. After that, 40 mL of 0.1 M HCl mixture and distilled water (1: l ratio) was added until a homogeneous solution and filtered into a 50 mL volumetric flask. The filtrate was added to 50 mL of distilled water, and then it was analyzed by using Atomic Absorption Spectrometer (AAS).

Statistical Analysis. The increase in weight in each treatment were analyzed by considering homogeneity with Bartlett test. If the result is homogeneous, it is followed by analysis of variance (ANOVA) of the track on the real level (α) of 0.05. Statistical analysis was examined using SPSS 18 Software.

RESULTS AND DISCUSSION

The increase in weight of banana plants showed the higher tendency of Cr content on the planting medium was, the smaller increase in weight was. It can be seen from Table 1, Figures 1 and 2. Root-shoot ratio and Cr content in root and shoot tends to be greater the increase of Cr concentration in the medium. It can be seen from Table 1 and Figure 3.

Table 1. The increase in weight, root-shoot ratio and the Cr content of the banana plant *(Musa acuminata)* cv. Ambon Lumut.
(an average of 10 replicates).

Treatment	Increase in weight (gram)	Root-shoot ratio	Cr content (mg/kg)	
			Root	Shoot
0 ppm	1.47	1.19	0.10	0.10
50 ppm	1.06	1.38	112.01	26.78
100 ppm	1.61	1.12	550.75	10.54
200 ppm	0.45	1.08	3331.97	299.97
400 ppm	0.32	2.60	5193.33	337.29

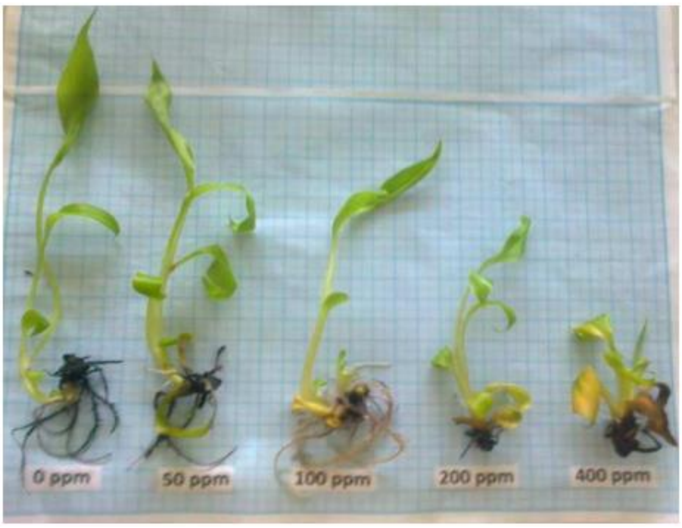

Figure 1. Plants after 6 weeks of treatment.

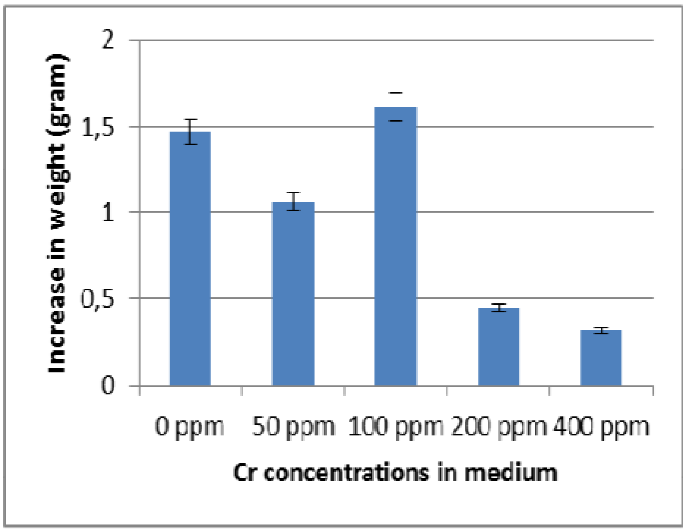

Figure 2. The increase in weight (gram) of the banana plant (*Musa acuminata*) cv. Ambon Lumut.

The increase in weight and root-shoot ratio were used as an indication of growth. The increase in weight tends to wane in increasing the concentrations of Cr in the planting medium. This was because Cr impact various processes in the plant body, which was in the process of photosynthesis occured among electron transfer inhibition, inactivation of the Calvin cycle enzymes, reducing CO_2 fixation and disorganization of the chloroplasts (Shanker *et al.,* 2005). The disruption of this process, of course, plant growth would also be influenced. Similarly, treatment of various studies with Cr could be seen that all the disturbances arising, caused by Cr influential in the production of Reactive Oxygen Species (ROS) that cause oxidative stress (Shanker *et al.,* 2005; Panda and Choudhury, 2005; Subrahmanyam, 2008; Pandey *et al.,* 2009; Yildiz *et al.,* 2012). ROS reacts with a variety of fats, proteins, pigments, nucleic acids and cause lipid peroxidation, membrane damage and inactivity of enzymes, thus it impact cell viability (Diwan *et al.,* 2010).

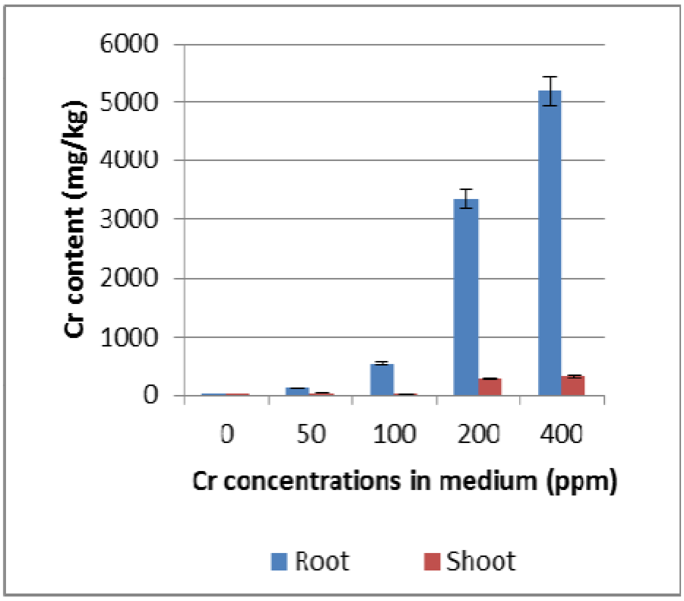

Figure 3. Cr content of the banana plant *(Musa acuminata)* cv. Ambon Lumut.

One of the important processes is disrupted due to an increase in ROS is the reduction in the rate of photosynthesis in various plant species. This is because the chloroplast is an important target in oxidative stress (Pandey et al., 2009).

While the root-shoot ratio shows that the greater the concentration of Cr in the medium was, the greater the root-shoot ratio was. This means that the growth forms more roots and it refers to the root growth.

It is considered as a very strong characteristic to assess the tolerance of plants to heavy metals (Diwan et al., 2010).

The content of Cr in the roots of banana Ambon Lumut is higher than those of in shoots. In a medium in a concentration of Cr 400 ppm, Cr content in the roots of 4-52 times is higher than at the shoot. In addition the results of previous research on various plants generally show the similarities. For example, at the Leersia hexandra Swartz Cr accumulated on the walls of root cells (Zhang et al., 2009; Liu et a1., 2009). Oliveira (2012) also concluded that the Cr mainly accumulated in the roots, little is translocated to the leaves. It is put forward based on the results of research Vernay et a1. (2007) on Lolium perenne that the roots accumulated Cr 10 times more than the leaves; research results Zayed et al. (1998) on some types of vegetables that can be known that Cr is running very slow translocation and accumulation in roots 100 times higher than the shoots; Huffman and Allaway research results (1973) in bean seed found that only 0.1% Cr accumulation, whereas at the root of 98% and the results of Liu et al. study. (2008) on Amaranthus viridis and Brassica oleracea is known that Cr accumulated mainly in the roots.

Plants have three basic strategies for protecting the body from the effects of toxic heavy metals, which become: (1) Excluder (heavy metals in the body is lower than its surroundings), (2) Indicator (heavy metals in the body the same as in the environment) and (3) Hyperaccumulator (heavy metals in the body more higher than its surroundings) (Mohanty & Patra, 2011). When considering these criteria, we can see that the banana plant cultivar Ambon Lumut includes heavy metals hyperaccumulator. Singh et al. (2013) divides hyperaccumulator plants into four groups, namely: low hyperaccumulator (1000 to 2000 mg/Kg), moderate hyperaccumulator (2000-3000 mg/Kg), hight hyperaccumulator (3000-5000 mg/kg) and very hight hyperaccumulator (more than 5000 mg/Kg). Although the banana plant cultivar Ambon Lumut includes hight hyperaccumulator plants, but only accumulated in the roots. So that is translocated to the shoot about 2-24%.

CONCLUSION

The results showed that the tendency of the greater Cr concentration in the medium was, the smaller the increase in weight was. Root-shoot ratio and Cr content in root and shoot tends to be greater in line with the increase of Cr concentration in the medium.

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