

**Research Article**

**Mercury uptake by *Zea mays* L. grown on an inceptisol polluted by amalgamation and cyanidation tailings of small-scale gold mining**

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**Abstract:** Pollution of agricultural land by tailings from the Artisanal and Small-Scale Gold Mining (ASGM) can increase the concentration of mercury (Hg) in plant tissue thus; it will endanger the health of humans and animals that consume it. This study aimed to determine the amount of Hg taken up by maize plants on an inceptisol contaminated with tailings from amalgamation and cyanidation processes of small-scale gold mining. A field experiment was conducted at the experimental field of Faculty of Agriculture, University of Mataram by using a factorial randomized block design consisting of two factors, namely: (1) tailings (L), and (2) fertilization (N). The results indicated that adding amalgamation and cyanidation tailings increased the physical and chemical fertility of the soil studied but caused Hg pollution. Most of the Hg in plant tissue was concentrated in the roots of the plants. The content of Hg in the plant shoot was above the threshold value of mercury allowed in plants. The NK33 variety of maize can be categorized as a mercury tolerant plant as the plant grew well in a mercury-contaminated soil.

**Keywords:** *maize, mercury, nitrogen, small-scale gold mining, tailings*

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**Introduction**

At many ASGM spots, amalgamation and cyanidation gold processing plants are often seen side by side with agricultural lands thus the tailings produced is released into rice paddies field which has been casually used as tailings pond leading to agricultural land contamination with no strategy to contain or manage the contaminant burden of the waste (Krisnayanti et al., 2012; Krisnayanti, 2018). This situation has also occurred in Sekotong District as one of the ASGM spots in West Nusa Tenggara. In this location, the processing of gold ore is carried out by using amalgamation and cyanidation methods (Muddarisna et al., 2013). The amalgamation method is a process of binding gold from mineral rocks by using mercury (Hg) as an amalgam substance, whereas the cyanidation method is a further attempt to capture gold from the

amalgamation tailings using cyanide (CN) (Velásquez-López et al., 2011). Mercury concentrations found in tailings from the amalgamation system in the Sekotong area ranged from 741-7874 ppm (Anderson et al., 2010). Mercury concentrations of 78-7374 ppm were also recorded in ASGM spot in Lantung District, Sumbawa Regency (Krisnayanti, 2011). According to Indonesian's Government Regulation No.18 of 1999 for Management of Hazardous and Toxic Waste, the maximum threshold limit value for Hg in soil is 0.01 ppm. In China, the maximum limit of the total content of Hg in soil and plant tissue is between 1.5 and 0.02 ppm (Krisnayanti et al., 2012). The Hg concentration in agricultural land in Japan is > 5.4 ppm while for rice fields in the World it ranges from 0.004-1.5 ppm (Kabata-Pendias and Pendias, 2000) This is supported by another finding of high Hg concentrations (6600 ppm) in

tailings in Sekotong District (Krisnayanti et al., 2012). Tailing discharging from the two methods to the water system and lands is believed to cause mercury pollution on agricultural lands (Tomiyasu et al., 2013; Ismawati et al., 2015). When a high concentration of mercury in tailings that is above the threshold value discharged into agricultural land can certainly cause soil pollution. Because of its toxicity and persistent nature in the environment, it is necessary to understand the mercury concentration in the soil. It is important to know the level of threat to community health because mercury in the soil can be absorbed by plant tissue (roots, shoot, and seeds) which can eventually enter the human body through food consumption; thus it can give a fatal effect to human health. Therefore, the level and concentration of mercury in soil and plant tissue need to be analyzed.

This study aimed to obtain information that maize grown on an inceptisol contaminated with amalgamation and cyanidation tailings from ASGM area is safe or not to be consumed, and to determine the status of maize as a

hyperaccumulator plant that is tolerant, resistant and sensitive to mercury.

### Materials and Methods

The experiment was carried out in the Laboratory and Horticulture Plant Experimental field, Faculty of Agriculture, University of Mataram, from March to July 2012. The growing media consisted of three different growing media which were inceptisol (KT), inceptisol + amalgamation tailing + compost (TL1) and inceptisol + cyanidation tailing + compost (TL2). The soil used in this experiment was an inceptisol taken at a depth of topsoil (0-20 cm) from farmer' field in Sekotong which was not polluted by Hg that was proved by laboratory results. The amalgamation and cyanidation tailings were taken from the tailing ponds of the gold processing units in Sekotong, West Lombok. The soil and tailings obtained were then air-dried, sieved to pass through at 2 mm sieve and prepared for the growing media to be analyzed in the laboratory. The growing media characteristics are presented in Table 1.

Table 1. The growing media characteristics

Type of analysis	Growing Media		
	KT (inceptisol)	TL1 (inceptisol + amalgamation tailing+ compost)	TL2 (inceptisol + cyanidation tailing +compost)
Texture	loamy sand	sandy loam	silty loam
Organic-C (%)	0.82 (sr)	1.37 (r)	1.22 (r)
pH H <sub>2</sub> O	6.4 (am)	7.1 (n)	7.1 (n)
Total N (%)	0.04 (sr)	0.19 (r)	0.16 (r)
C/N	18.95 (t)	7.0 (r)	7.5 (r)
P-Olsen (ppm)	18.9 (t)	29.5 (st)	20.5 (st)
Exchangeable K (me/100 g)	0.69 (t)	0.40 (s)	0.37 (s)
Exchangeable Ca (me/100 g)	7.75 (s)	4.51 (r)	4.52 (r)
Exchangeable Mg (me/100 g)	2.31 (t)	2.28 (t)	4.02 (t)
Cation Exchange Capacity (me/100 g)	6.5 (r)	42.6 (st)	43.7(st)
Hg (ppm)	-	5.48	12.20

Note: Sr=very low; r=low; s=medium; t=high; st=very high; am= slightly acid; n=neutral. Source: Soil Research Center, Agricultural Research and Development Agency, Agriculture Department, 2005

The growing media contaminated by amalgamation and cyanidation tailing have a higher Hg concentration than the control treatment, and it was above the threshold limit value (> 1.5 ppm) for agricultural land. From the two tailings treatments, it appears that the cyanidation treatment has a higher concentration of Hg (12.20 ppm) than amalgamation tailings (5.48 ppm). The high Hg concentration in the cyanidation tailings

treatment showed that there had been a residue concentration of Hg from the amalgamation tailings in the cyanidation tailings. In this study, the variety of maize used was NK33 as a plant indicator to determine the amount of mercury uptake from soil contaminated by amalgamation and cyanidation tailings containing mercury. The NK33 maize variety is the most cultivated maize variety in Sekotong. Inorganic fertilizers (Urea, SP-

36, and KCl), compost, and maize seeds were obtained/purchased from the local agricultural shop. On a polybag (size 15 kg), a total of 10.5 kg of soil sample (70%) was added to 4.5 kg (30%) tailings (amalgamation and cyanidation) so that a total planting medium weight was 15 kg. A 75 g of compost was then added to the planting medium (soil and tailing mixture) and then incubated for 1 week.

During the incubation period, the water content was maintained to field capacity. For the control treatment, the growing media used was 15 kg of soil mixed with 75 g of compost without adding amalgamation and cyanidation tailings. This experiment was designed by a Block Factorial Design that was arranged randomly consisting of two factors, i.e. tailings (L), and nitrogen fertilizer (N). Each factor consisted of two levels. The first factor (tailings) consisted of (1) amalgamation tailings (L1), and (2) cyanidation tailings (L2). The second factor (N fertilizer) consisted (1) with N fertilizer (N1) and (2) without N fertilizer (N2). Two treatments were added in each treatment, i.e. (1) Inceptisol without N fertilizer and (2) Inceptisol with N fertilizer. Those four treatment combinations and two control treatments were arranged in a randomized block design with three replicates.

Phosphorus (P) fertilization was carried out on all treatments at the time of planting including control, as a basic fertilizer. P fertilizer given was SP-36 with a dose of 1.04 g SP-36/pot or equivalent to a dose of 150 kg P<sub>2</sub>O<sub>5</sub>/ha. N and K fertilizers were given regularly 3 times, i.e. at planting (0 days 0.54 g urea/pot or equivalent to 80 kg urea/ha), at 3 weeks after planting (1.09 g urea/pot or 160 kg urea/ha), and at 9 weeks after planting (1.09 g urea/pot or 160 kg urea/ha). K fertilizer used was KCl fertilizer, each given as much as 0.21 g KCl/pot (30 kg K<sub>2</sub>O/ha), 0.42 g KCl/pot (60 kg K<sub>2</sub>O/ha) and 0.42 g KCl/pot (60 kg K<sub>2</sub>O/ha) for all treatments. Two maize seeds were planted in each pot at ± 3 cm. After 1 week of planting, one of the healthiest maize plants was maintained.

Watering was done once a day, in the morning or evening. Watering was carried out until the soil reaches a field capacity (maximum soil capacity retains water). Weeding was done manually, by removing weeds and immersing them into the planting media. This was done to keep the concentration of Hg on the growing media not reduced. Harvesting was conducted when the maize plant was 105 days old by cutting the plants at the bottom of the stem. The obtained cob was separated and labelled for measurement and weighing. The same way was done for the top and bottom of maize plants. The dry crop was done

after harvest for ±24 hours to constant weight. Total mercury analysis of the growing media, maize shoot, maize roots, and maize seeds was performed in the laboratory of Soil Science, Agriculture Faculty, University of Mataram by using Cold Vapour Atomic Absorption Spectroscopy (CVAAS; Huaguang model F732-S, China).

The methods of Moreno et al. (2005) and Krisnayanti et al. (2012) were used for substrate analysis. One gram of sub-sample was pre-digested overnight, at room temperature, with aqua regia (15 mL; analytical grade nitric and hydrochloric acids) in borosilicate beakers. The following day the preparations were digested at 120°C for two hours. Deionised water was then added (20 mL), the digest solutions were filtered (Whatman no. 42), and then made to 100 mL. Ten millilitres of the aliquot was analysed with 1 mL of 5% nitric acid and 1 mL of freshly prepared SnCl<sub>2</sub> (10%) as the reducing agent. Sequential 1:0 dilutions were performed in nitric acid (5%), where necessary, to yield an absorbance on the standard curve. The limit of detection for mercury in solution following the described methodology was 0.1 ng/mL. The mercury absorbance from replicates analysis (n ¼ 10) of a 10 ng/mL mercury standard solution was reproducible with less than 5% variation. The data obtained were analyzed by analysis of variance (ANOVA). In order to find out the significant differences between the variables measured, further tests were conducted by using Duncan's Multiple Range Test (DMRT) at 5% level.

## **Results and Discussion**

### ***The height of maize plant***

The results statistical analysis presented in Table 2 show that the tailings factor (T) and fertilizer factor (N) was significantly different at each observation time. The similar trend was shown by the interaction between the tailings and N fertilization treatments where there was a significant effect on almost all observation times, except at the observation time of 2 weeks after planting. While the source of group variant (replication) at each time of observation did not show any significant differences. As shown in Figures 1A, B, and C, it was clear that the treatment of N fertilizer to the growing media has a significant effect on maize height. Introducing N fertilizer indeed increased the maize growth from ages 1 to 9 weeks after planting (WAP). This shows that N fertilizer is an important element needed in order to increase plant vegetative growth. Therefore, it is suggested that N fertilization needs to be applied to the growing media to obtain optimal NK33 maize growth. The treatment of N fertilizer in the growing media has

a significant effect on plant height (Figure 1). Application of N fertilizer improved the maize growth from the first week to 9 weeks after planting. This shows that N fertilizer is an

important element needed in increasing plant vegetative growth. Therefore, N fertilization needs to be added to the growing media to obtain optimal NK33 maize growth.

Table 2. Analysis variant of the maize height

Source	Observation time (week after planting)								
	1	2	3	4	5	6	7	8	9
Group	ns	ns	ns	ns	ns	ns	ns	ns	ns
Factor T	**	**	**	**	**	**	**	**	**
Factor N	**	**	**	**	**	**	**	**	**
Interaction TxN	**	ns	**	**	**	**	**	**	**
DMRT 5%	0.46	0.38	0.86	2.84	3.23	3.77	4.18	1.71	1.09

Note: ns = non-significant; \*\* = significant

**Maize dry weight**

The results of the statistical analysis presented in Table 3 indicated that the tailings factor (T) has a significant effect on the dry weight of maize plants on all parts of plant tissue (roots, shoot, and cob).

Table 3. Analysis of variance of maize dry weight

Source	Root	Shoot	Cob
Group	ns	ns	ns
Factor T	**	**	**
Factor N	**	**	**
Interaction TxN	**	ns	**
DMRT 5%	0.66	2.79	2.85

Note: ns = not significant; \*\* = significant

The same indication also showed in N fertilizer treatment, where fertilization had a significant effect on the dry weight of the plants obtained. Whereas the interaction between tailings treatment and N fertilization showed a very strong correlation with root and cob parameters, but it did not show in plant shoot parameters. While the source of group diversity (replication) did not show any significant differences. N fertilizer is the main element needed in large quantities to support plant growth. Thus, plant growth is highly dependent on N availability. Fertilizing N into the soil will increase N availability for plants which will ultimately increase crop production. The effect of N fertilizer on the dry weight of plant tissue (roots, shoot, and cob) is presented in Figure 2. As shown in Figure 2A it was clear that the treatment of N fertilizer in the growing media of amalgamation tailing has a significant effect on the crop dry weight. Fertilizer treatment increased the crop dry weight in root, shoot, and cob. This shows that the initial N content in amalgamation tailings was

insufficient to support optimally NK33 maize growth. Therefore, N fertilization needs to be added to the growing media of amalgamation tailing to support good growth of maize. The similar trend happened in the growing media of cyanidation tailing and control treatment (Figures 2 B and C). The N fertilization treatment increased the amount of crop dry weight in parts of the root, shoot, and cob. This shows that N is an essential nutrient to support plant growth and development.

**Total Hg in plant tissues**

Data presented in Table 4 show that most of the Hg absorbed by plants were concentrated in the root tissue of maize, which was equal to 95.7-98.6%; only about 1.4-4.3% Hg was concentrated in the shoot and no mercury was detected on maize seeds (below threshold <0.02 ppm). It was also seen that the N fertilizer treatment has a significant effect on the total Hg in the roots and plant shoot in both growing media (TL1 and TL2). The total Hg in plant tissue in the N fertilizer treatment was higher than the treatment without N fertilization. This is caused by an increase in plant growth followed by increasing of Hg absorption by the plant. In addition, the treatment of growing media affected the total Hg in plant tissue. The total Hg on cyanidation tailing was both higher than amalgamation tailing with or without N fertilization. This can be understood considering the total Hg concentration in cyanidation tailings was higher than amalgamation tailing. High concentration of Hg in the plant root tissue may be caused by low Hg mobility. It was stated that the mobility of Hg in plant tissues is very slow and most of it only concentrated in plant roots (Kabata-Pendias and Pendias, 2000). Another factor that affects the level of Hg accumulation in plants are the species and the variety (McGrath et al., 2001).

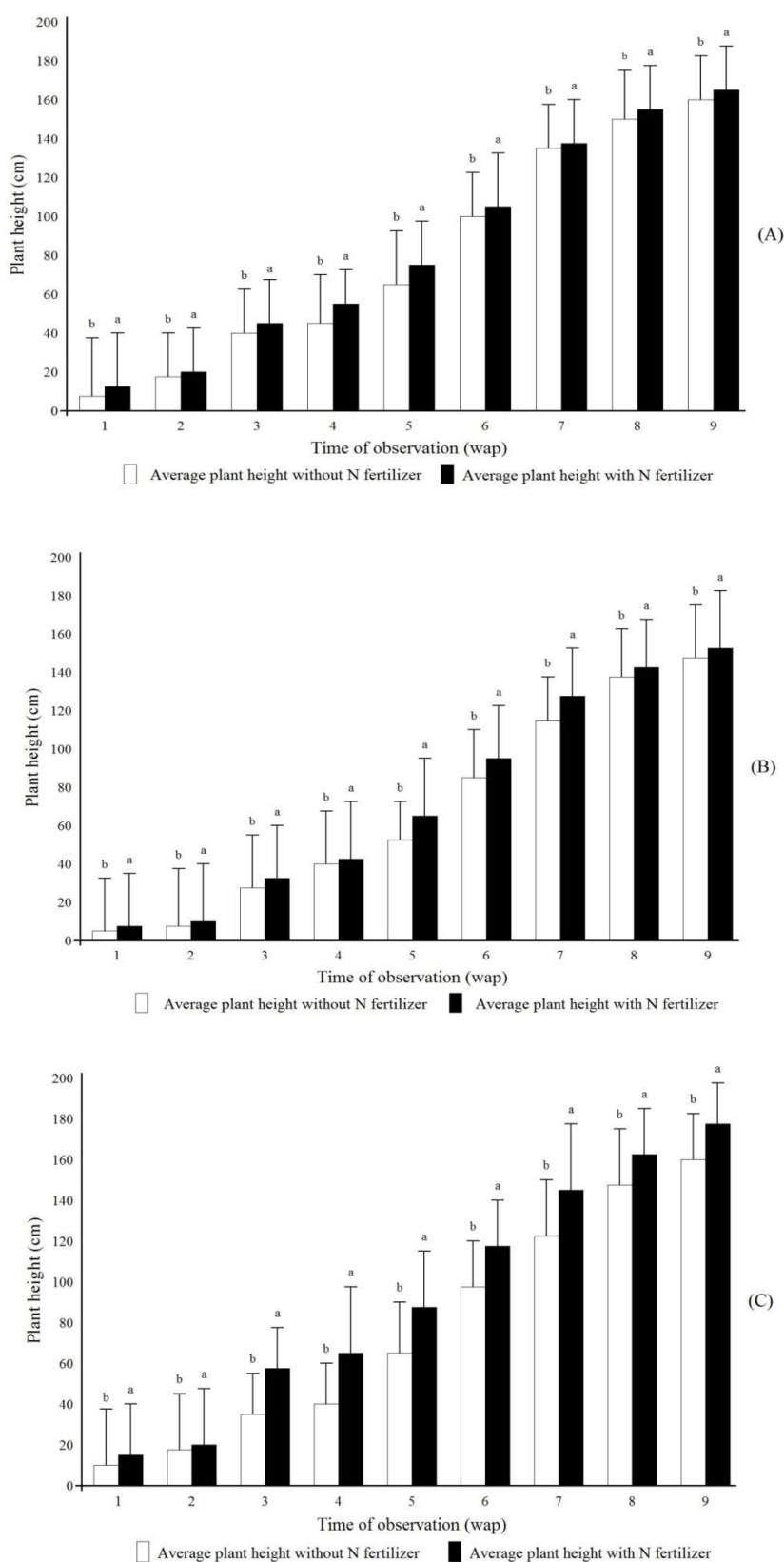


Figure 1. Effects of application of N fertilizer on plant height in growing media of amalgamation tailings (A), cyanidation tailing (B), and inceptisol (C). Bar charts with different letters and in parentheses are significant in DMRT at 5% level

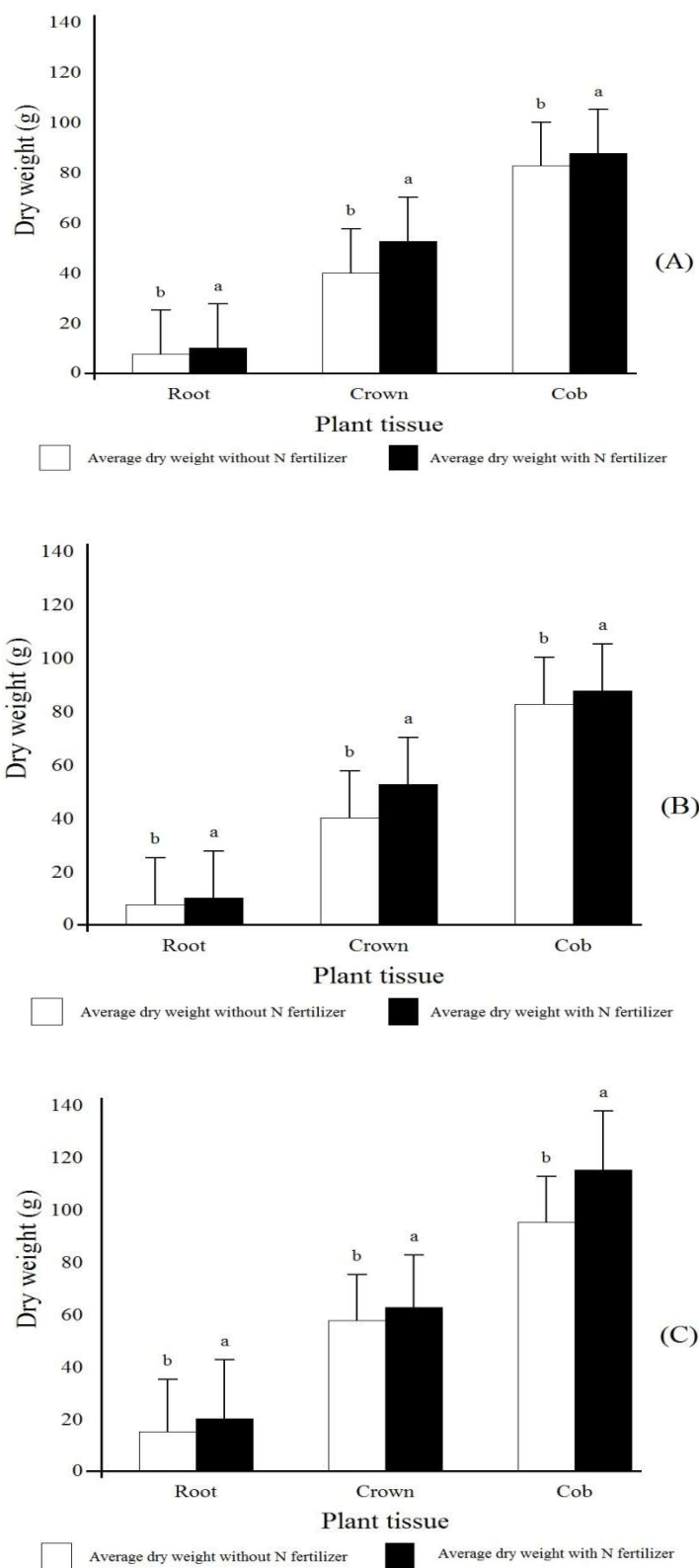


Figure 2. Effect of N fertilizer to plant tissue's dry weight on the growing media of amalgamation tailing (A), cyanidation tailing (B), and inceptisol (C). Bar charts with different letters in parentheses are significant in DMRT at 5% level

Table 4. Total Hg (THg) concentration on the plant tissues

Growing Media	Plant Tissue (ppm)					
	Root		Shoot		Seed	
	DMRT 0.02		DMRT 0.003		0	
	N1	N2	N1	N2	N1	N2
TL1	0.27 b (b)	0.43 a (b)	0 b (b)	0.01 a (b)	0	0
TL2	0.33 b (a)	0.57 a (a)	0.01 b (a)	0.03 a (a)	0	0
KT	0 a (c)	0 a (c)	0 a (b)	0 a (c)	0	0

Note: Different letters behind treatment and parentheses show significantly different ( $P > 0.05$ ) effect of giving N fertilizer on absorption and the effect of planting media on Hg uptake in plant tissue. DMRT factor TxN interaction at level 5% = 0.02

While most of the plants that uptake Hg tends to accumulate it on the roots (Lenka et al., 1992), some plants are even able to accumulate moderate amounts of Hg in the shoots (Dushenkov et al., 1995) either due to translocation or direct absorption of the vapour form. Suszcynsky and Shann (1995) stated that plants exposed to HgO can uptake and accumulate it in plant's shoots, but there is no translocation to the roots. It is expected that the toxic metal ions enter the plant cells by the same uptake mechanism as micronutrients, that is competing with the elements for absorption. As a class B metal, Hg prefers to binds with sulphur and nitrogen ligands, and it will enter to the cell through ionic channels that will compete with other heavy metals such as cadmium or trace elements like zinc, copper and iron (Blazka and Shaikh, 1992). The absorption mechanisms between the amount of Hg content in the soil and its uptake by plants is not linear and depends on some soil characteristic variables such as cation-exchange capacity, soil pH, soil aeration, and plant species. The mercury uptake can be reduced when the soil's pH is high and/or there is an abundance of lime and salts (Patra and Sharma, 2000; Patra et al., 2004). The provision of compost in this study has a big role in increasing the absorption of Hg by plant tissue. This can occur through two mechanisms which are the provision of compost into the soil will increase N availability and the ability of the soil to provide other nutrients for optimal plant growth, and will increase dissolution and mobility Hg into the soil through increasing chelate and chelating processes; thus more Hg is available and can be easily absorbed by plant roots.

## Conclusions

Introducing of nitrogen fertilizer on contaminated soil with amalgamation and cyanidation tailings increased the growth and dry weight of maize and

total mercury concentration in the plant while most of the mercury in plant tissue was concentrated in the roots. Thus, the maize seeds were below the threshold limit value so that maize kernels were safe for consumption. NK33 variety of maize can be categorized as a Hg tolerant plant. The treatment of adding amalgamation and cyanidation tailings increased the soil physical and chemical fertility of an inceptisol but has caused mercury pollution.

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