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Original Research Paper

Optimization of Plant Species and Chelating Agents in Phytoextraction of Gold from Small-Scale Gold Mine Tailings

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ABSTRACT

The disposal of small-scale gold mine tailings into agricultural land has caused a decline in crop production in the District of Sekotong, Lombok Regency of Indonesia. One of the technologies that can be developed for remediation of soil contaminated by small-scale gold mine tailings is phytoextraction. This study was conducted in two steps. The first step was aimed to study the effect of various doses of fertilizer and soil amendment on the production of biomass of two plant species studied previously, i.e. *Paspalum conjugatum* and *Cyperus kyllingia*. Each seedling of the two plant species was grown for 9 weeks on the tailing disposal dam in a 1 × 1 m plot. Treatments tested were (1) plant species (two species), and (2) dose of NPK fertilizer (50, 100, and 150 kg/ha), and (3) dose of organic matter (5, 10, and 15 t/ha). The second step was to study the effect of the type and dose of chelating agents on the best plant species resulted from the first step of this study. The treatments tested consisted of three types of chelating agent (ammonium thiosulphate, sodium thiosulphate and sodium cyanide) and three doses of each chelating agent (1, 1.5, 2 g/kg). At the time of harvest (9 weeks), shoot and root of each plant were separated for the analysis of Au accumulation. The results showed that type of plant, dose of organic matter, and optimal applied fertilizer that generated the highest growth and biomass production of the accumulator plants was a combination of *P. conjugatum,* 10 kg NPK fertilizer/ha, and 10 t organic matter/ha. The type and dose of chelating agents applied to the combined treatment affected Au accumulation. The best combination of type and dose of chelating agent was 2 g of ammonium thiosulphate /ha. Overall, the combination of plant species, fertilizer dose, organic matter dose, type of chelating agent, and dose of chelating agent was *P. conjugatum* with 100 kg NPK fertilizer/ha, 10 t organic matter/ha and 2 g ammonium thiosulphate/kg.

INTRODUCTION

The disposal of small-scale gold mine tailings to agricultural land has caused a decline in crop production in the District of Sekotong, Lombok Regency since 2010. The decline in crop production is mainly due to the relatively high content of heavy metals such as Hg, Cu, Pb and Zn in the gold processing tailings discharged to the lands. Despite containing heavy metals that are toxic to crops, the discharged tailings still contain gold. Results of a survey conducted by Krisnayanti et al. (2012) in West Lombok regency showed that the amalgamation and cyanidation tailings still containing 1.2-6.3 mg Au/kg. This low level of Au is no longer profitable for further gold extraction and processing in a conventional mining method.

In relation to the facts that contamination of small-scale gold mine tailing containing toxic heavy metals that interfere with the growth and production of food crops, and on the other hand the tailings still contain gold, although in low concentrations, it is necessary to restore the land with adequate compensation. One of the technologies that can be developed for remediation of soil contaminated by small-scale gold mine tailings is phytoextraction that can play a double role, namely phytoremediation and phytomining (Anderson et al. 2005). Benefits of phytoremediation is, a toxic heavy metal absorption by metal accumulator plants in order not to contaminate soil, water and plants, while the benefits of phytomining is Au absorption by plants and then the content of Au in the plant can be harvested and processed into bio-ore of gold.

Phytoremediation is the use of green plants to extract or remove soil contaminant metals (Raskin et al. 1994, Tangahu et al. 2011). Phytomining is the production of ‘metal plants’ by growing hyper-accumulator plants that can accumulate metals in high concentrations (Brooks et al. 1998), and then harvest and burn the biomass to produce ‘bio-ore’ (Anderson et al. 1999, Nedelkoska & Doran 2000). Some researchers in New Zealand have developed a system in which Au and Hg can be absorbed by similar plants in some small-scale gold mining (Moreno et al. 2005). Therefore, phytomining usually refers to the recovery of metals. Hyper-accumulator plants are plant species that can absorb large amounts of metal elements compared to the non-accumulator plants, without visible symptoms of damage or death of the plant (Baker & Brooks 1989).

A hyper-accumulator plant can accumulate more than 10 mg/kg Hg; 100 mg/kg Cd; 1,000 mg/kg Co, Cr, Cu and Pb; and 10,000 mg/kg Zn and Ni. Up to now, about 400 species of plants and at least 45 families have been reported as metal hyper-accumulator plants (Baker et al. 2000). Besides that, the hyper-accumulator plant should be able to grow outside its original area, has a root system that is stable, and is capable of transporting metal elements to its shoot (Thangavel & Subhuram 2004). Therefore, the plant used in the phytoextraction should be local (indigenous) around the contaminated location, because these plants have adapted to the polluted conditions (Wolfe & Bjornstad 2002).

Studies conducted in New Zealand showed that gold and mercury were found in the same plants grown in soil contaminated by gold mine tailings (Moreno et al. 2005). Results of a study conducted in West Lombok by Muddarisna et al. (2013) showed that *Paspalum conjugatum, Cyperus kylingia*, and *Lindernia crustacea* were three species that had the potential to be used as mercury phytoremediator on agricultural land contaminated with small-scale gold mine tailings containing mercury. Because mercury is the main reagent that is widely used in small-scale gold mining process, the plant is also expected to be able to accumulate gold and other heavy metals. In the further study, the three species were grown for nine weeks on gold amalgamation and cyanidation tailings with the addition of ammonium thiosulphate or sodium cyanide as a chelating agent. The results showed that the highest accumulation of Au (859.8 µg/ kg) was found in the shoot of *Paspalum conjugatum* with the addition of ammonium thiosulphate (Handayanto et al. 2014). Adding ammonium thiosulphate or sodium cyanide increased the accumulation of Au in the shoot by 108% and 34% compared with the treatments without the addition of chelating agents. The ability of two selected species was further tested in the field in the respectable tailing dam near the gold cyanidation process location. Two of the three species of the best (at most absorbing Au), the results of the first year of study, are *Paspalum conjugatum* and *Cyperus kyllingia* combined with the application of ammonium thiosulphate or sodium cyanide with growth of nine weeks old. The results showed that the highest concentration of Au (601.9 µg/kg) was found in the shoot of *Paspalum conjugatum* with application of ammonium thiosulphate. Results of the studies indicated that the amount of accumulation of Au was below 1 mg/kg, although the amount of accumulated Au was already quite high when compared to the uptake of Au by plant under normal conditions. According to Sheoran (2013), in normal conditions (without the addition of chelating) plant is only capable of absorbing Au ions around 0.001 mg/kg. However, in phytoextaction/ phytomining of gold, ideally should gain accumulation of 3 mg Au/kg of dry biomass of the plants (Anderson et al. 2005).

Based on the above, this study aimed at the optimization of plant species and chelating agents on phytoextraction of gold from small-scale gold mine tailings. This study was conducted in two steps. The first step was aimed to study the effect of various doses of fertilizer and soil amendment on the production of biomass of two plant species studied previously, i.e. *Paspalum conjugatum* and *Cyperus kyllingia*. The second step was to study the effect of the type and dose of chelating agents on the best plant species resulted from the first step of this study.

MATERIALS AND METHODS

Location of study: A field experiment was conducted from March to August 2015 on an agricultural land dumped with tailings of gold cyanidation processing unit at Sekotong District of West Lombok (115°46'-116°20'E and 8°25'-8°55'S). The tailing dumping area was about 20 m × 30 m with the tailing depth varied from 20 cm to 40 cm. The characteristics of the tailing were similar to that used in previous study (Handayanto et al. 2014) as follows: sandy clay texture, pH 7.7, organic-C 1.19%, total N 0.001%, available P 2.9 mg/kg, exchangeable K 0.001 cmol/kg, 0.64 cmol/kg, exchangeable Na, exchangeable Ca 1.99 cmol/kg, exchangeable Mg 0.84 cmol/kg, CEC 11.57 mol/kg, 31% base saturation, Hg 1.090 mg/kg and Au 1.68 mg/kg.

Experiment 1: Effects of fertilizers and organic matter application on biomass production of accumulator plants: Each seedling of local plant species (*Paspalum conjugatum* L. and *Cyperus kyllingia* L. Endl.) which have been previously acclimatized for 2 weeks, were grown for 9 weeks on the tailing disposal dam in a 1 × 1 m plot. Treatments tested were (1) plant species (two species), and (2) dose of a combine NPK fertilizer (50, 100 and 150 kg/ha), and (3) dose of organic matter as a soil amendment (5, 10, and 15 t/ha). Two control treatments, i.e. each plant species without application of fertilizer and organic matter, were also included. The combined NPK fertilizer, which is produced by PT. Petrokimia Gresik, SNI 02-2803-2000 as Phonska, contains 15% N, 15% P_2O_5, 15% K_2O and 10% S. The organic matter used in this study is a mixture of compost and cow dung locally made by farmers. The characteristics of the organic matter used were as follows: 5.6% moisture content, 4.27% organic C, 0.24% total N, pH 7.4, and 22.8 mg available P/kg. Twenty treatments were arranged in a randomized block design with three replications. Planting was made with a line spacing of 15 cm, and 20 cm between rows. During the experiment, the soil water content was maintained on a regular basis so as not to hamper growth. At the time of harvest (9 weeks), shoot and roots of each plant were separated, washed, weighed and dried in an oven for 48 hours at 60°C for calculation of biomass production and analysis of Au content. The content of Au was determined by Graphite Furnace Analyser combined with Atomic Absorption Spectrophotometer, type A Analyst 50, Perkin Elmer, UK. For the analysis of Au, 0.2 g subsample of each ground plant sample was placed into a 10 mL borosilicate test tube. The tube was then heated at 550°C for one night in a muffle furnace. On the following day the ash was dissolved in a water bath containing 5 mL of aqua regia (a mixture of nitric acid hydrochloric concentration with a proportion of 3:1), and then was made to 10 mL with distilled water. The data were then performed an analysis of variance followed by the least significant different test at 5%. A treatment produced the highest plant biomass was then used in experiment 2.

Experiment 2: Effects of type and dose of chelating agents on Au accumulation by a selected accumulator plant: One of the twenty treatments of experiment 1 that produced the highest plant biomass, i.e. *Paspalum conjugatum* with application of 100 kg NPK fertilizer/ha and 10 t organic matter/ha, was then treated with type and dose of chelating agents and grown in the tailing disposal dam similar to that of experiment 1 for nine weeks. Plot size was also similar to that of experiment 1. The treatments tested consisted of three types of chelating agent (ammonium thiosulphate, sodium cyanide and sodium thiosulphate) and three doses of each chelating agent (1, 1.5 and 2 g/kg). A treatment without application of chelating agents was also included as a control. Ten treatments were arranged in a randomized block design with three replications. As in experiment 1, planting was made with a line spacing of 15 cm, and 20 cm between rows. All plots were given 100 kg NPK fertilizer/ha and 10 organic matter/ha based on the results of experiment 1. At 8 weeks after planting, chelating agents, each in 150 mL was added to respected treatments. During the experiment, the soil water content was maintained on a regular basis so as not to hamper the growth. At the time of harvest (9 weeks), shoot and root of each plant was separated, washed, weighed and dried in an oven for 48 hours at 60°C for calculation of biomass production analysis of Au content using methods similar to that used in experiment 1. The ability of plants to translocate metal from root to shoot measured using the translocation factor (Yoon et al. 2006). The data were then performed an analysis of variance followed by the least significant difference test at 5%.

RESULTS AND DISCUSSION

Experiment 1

Plant biomass production: Results of the first experiment showed that the application of NPK fertilizer and organic materials significantly affected biomass production of the two plant species grown for 9 weeks. The highest biomass production (based on dry weight of shoot and root), was found in *P. conjugatum* with the application of 100 kg NPK fertilizer/ha, and 10 t organic matter/ha, while the most stunted growth was observed for *C. kyllingia* with application of 50 kg NPK fertilizer/ha and 5 t organic matter/ha (Fig. 1). Application of high doses of NPK fertilizer and organic matter, i.e. 150 kg NPK/ha and 15 t organic matter/ha, were not significantly different from the results obtained by *P. Paspalum* with 100 kg NPK fertilizer/ha and 10 t organic matter/ha. For the *C. kyllingia*, application of 150 kg NPK fertilizer/ha and 15 t organic matter/ha gave the highest biomass production on *C. kyllingia*. However, the highest yield of *C. kyllngia* was still lower than the highest yield obtained *P. conjugatum* with 100 kg NPK/ha and 10 t organic matter/ha. Based on these results, the treatment chosen for the experimental phase 2 was the treatment of *P. conjugatum* with 100 kg NPK fertilizer/ha and 10 t organic matter/ha.

Au accumulation in *P. conjugatum* and *C. kyllingia* along with the shoot and root biomass production of the tested plants, the highest Au concentration was found in the shoot of *P. conjugatum* with 100 kg NPK fertilizer/ha and 10 t organic matter/ha. In general, the concentration of Au in the shoot of *P. conjugatum* on all treatments of NPK fertilizer and organic matter was higher than the *C. kyllingia* (Fig. 2). However, the concentration of Au in the roots was very low and no significant different between treatments. Compared with the results of several other studies, the accumulated Au obtained in this plant was relatively small and less than 1 mg/kg. This value was much lower than the results reported by Msuya et al. (2000) that *Raphanus sativus* L., *Allium cepa* L., *Beta vulgaris* L. and *Daucus carota* L. grown on media containing silica sand containing 3.8 mg Au/kg resulted in Au concentration greater than 200 mg/kg dry weight plant.

Experiment 2

Plant biomass: Results of the second experiment showed that the application of the type and dose of chelating agents significantly affected the production of biomass (based on dry weight of shoots and roots) of *P. conjugatum* (with the application of 100 kg NPK fertilizer/ha and 10 t organic matter/ha) during the 9 weeks of growth. The highest biomass production was found in *P. conjugatum* with the application of 100 kg NPK fertilizer/ha, 10 t organic matter/ha along with the supply of 2 g ammonium thiosulphate/kg. The most stunted growth was found in *P. conjugatum* with 100 kg

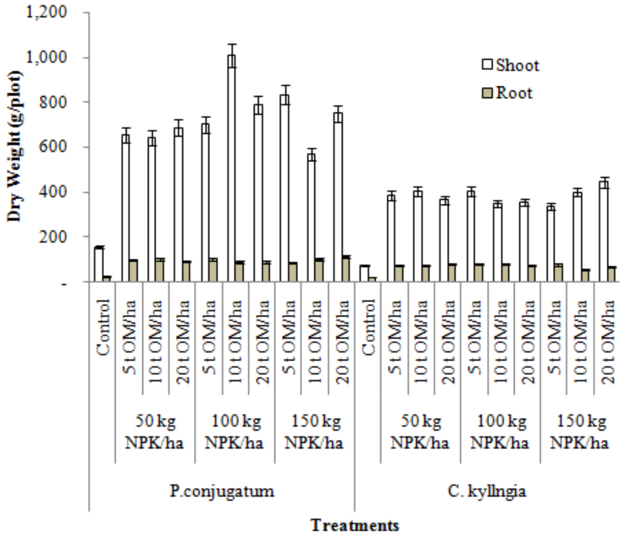

Fig. 1: Shoot and root dry weight of *P. conjugatum* and *C. kyllingia* grown for 9 weeks on small-scale gold mine tailing with various rates of NPK fertilizer and organic matter (OM) application.

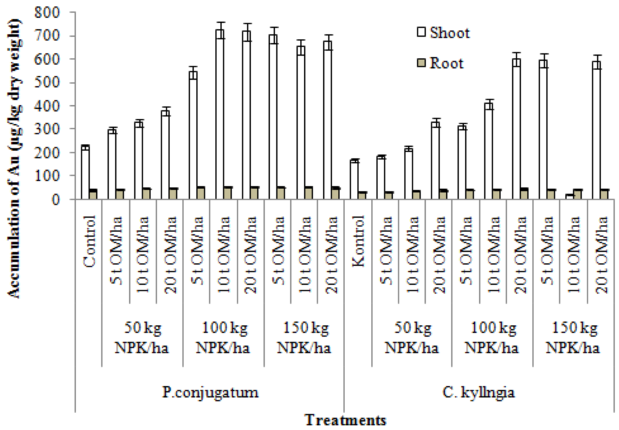

Fig. 2: Au accumulation in shoot and root of *P. conjugatum* and *C. kyllingia* grown for 9 weeks on small-scale gold mine tailing with various rates of NPK fertilizer and organic matter (OM) application.

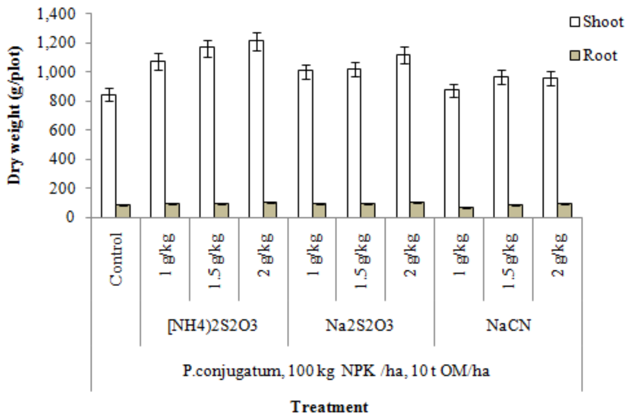

Fig. 3: Effects of types and doses of chelating agents on shoot and root dry weights of *P. conjugatum* (+100 kg NPK fertilizer/ha and 10 t organic matter/ha) that was grown for 9 weeks on small-scale gold mine tailing.

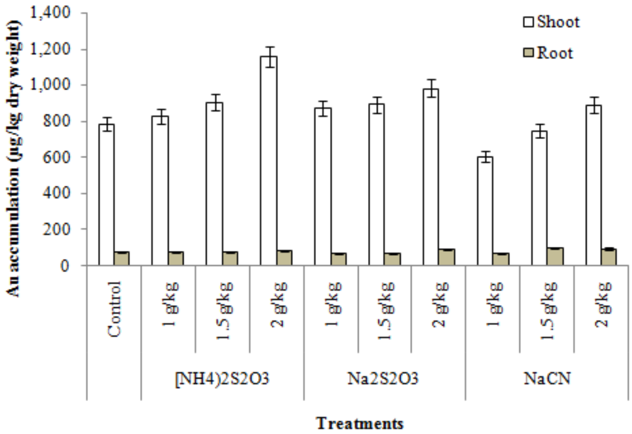

Fig. 4: Effects of types and doses of chelating agents on Au accumulation by *P. conjugatum* (+100 kg NPK fertilizer/ha and 10 t organic matter/ha) that was grown for 9 weeks on small-scale gold mine tailing.

NPK fertilizer/ha, 10 t organic matter/ha accompanied by application of 1 g sodium cyanide/kg (Fig. 3). Application of sodium cyanide at a dose of 2 g/kg resulted in the relative low production of biomass but it was significantly different than the application of 1 g/kg of ammonium thiosulphate. Compared to the control treatment, application of ammonium thiosulphate, sodium thiosulphate and sodium cyanide at a dose of 2 g/kg significantly increased 43%, 32%, 12% shoot dry weight and 18%, 15%, 7% root dry weight, respectively.

Au accumulation in *P. conjugatum*: Application of types and doses of chelating agents also gave significant effect to the accumulation Au in *P. conjugatum* shoot (with the application of 100 kg NPK/ha and 10 t organic matter/ha) at 9 weeks after planting. The increased doses of chelating material significantly increased the accumulation of Au in the shoot of *P. conjugatum*. Compared to the control treatment, application of ammonium thiosulphate, sodium thiosulphate, or sodium cyanide at a dose of 2 g/kg could significantly increase the accumulation of Au in the shoot of *P. conjugatum* by 47%, 24% and 13%, respectively. Results of a study previously conducted by Handayanto et al. (2014) showed that the addition of ammonium thiosulphate or sodium cyanide increased the accumulation of Au in the shoot of *P. conjugatum* by 108% and 34% compared to the treatment without the addition of chelating agent, although the absorption was still below 1 mg/kg. Ideally, gold phytomining should obtain Au accumulation of 3 mg/kg of dry biomass of plants (Anderson et al. 2005). In this experiment, the highest accumulation of Au (1.1 mg/kg) was found in *P. conjugatum* with the application of 100 kg NPK/ha, 10 t organic matter/ha along with the supply of 2 g ammonium thiosulphate dose/kg. The lowest accumulation of Au (0.6 mg/kg) was found in *P. conjugatum* with the application of 100 kg NPK/ha, 10 t organic matter/ha accompanied by application of 1 g sodium cyanide/kg (Fig. 4). The higher influence of thiosulphate compounds (ammonium and sodium) on the accumulation of Au compared to sodium cyanide because the thiosulphate compound is a lixiviant suitable for the extraction of gold from the media with a pH of 5-9, whereas cyanide is more suited to the medium with a pH > 10 (Sparrow & Woodcock 1995). Because gold cyanidation tailing used in this study had a pH value of 7.7, then Au was more driven by the uptake of thiosulphate compared by cyanide, as proposed by Wilson-Corral et al. (2011). Moreover, according to Habashi (1999), Au dissolution rate increases linearly with increasing concentration of cyanide to reach the maximum point. If it has reached the maximum, the additional cyanide will not improve dissolution of Au. Based on the above explanation, it was thought that the cyanide concentration of 2 g/kg used in the study exceeded the maximum concentration of cyanide for the leaching Au, which generally ranges from 0.25 to 0.75 g/kg (Mardsen & House 1992). According to Boyle & Smith (1994), in the process of dissolving gold using cyanide, other metallic elements such as copper, zinc, nickel, iron, cobalt and mercury also dissolved in alkaline cyanide solution. Reaction with other metals will increase, thereby reducing the need for cyanide extraction efficiency of Au.

CONCLUSION

Type of plant, dose of organic matter, and optimal applied fertilizer that generated the highest growth and biomass production of accumulator plants was a combination of *P. conjugatum*, 10 kg NPK fertilizer/ha, and 10 t organic matter/ha. Type and dose of chelating agents applied to the combined treatment affected the Au accumulation. The best combination of type and dose of chelating agent was 2 g of ammonium thiosulphate/ha. Overall, the combination of plant species, fertilizer dose, organic matter dose, type of chelating agent, and dose of chelating agent was *Paspalum conjugatum* with 100 kg NPK fertilizer/ha, 10 t organic matter/ha and 2 g of ammonium thiosulphate/kg.

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