Rehabilitation of Artisanal Mining Gold Land in West Lombok, Indonesia: 2. Arbuscular Mycorrhiza Status of Tailings and Surrounding Soils

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Abstract

Artisanal mining plays an important role in the Indonesia economy; however, it has created serious environmental destruction. The prohibition of artisanal mining is not a wise policy and never works. A more valuable strategy is to encourage artisanal mining; however, the associated poor health, safety, and environmental conditions must be improved. Therefore, effective rehabilitation of the environment post mining is essential. Phytoremediation is considered to be one method to achieve this rehabilitation. In this method, the interaction of plant roots with mycorrhiza is one of the key determinants of successful rehabilitation. A study to identify the indigenous mycorrhiza present in soil was carried out at an artisanal gold mining region at Sekotong, West Lombok, Indonesia. Soil samples were collected from the rhizosphere of a selection of indigenous plant species for the identification of the associated mycorrhizal fungi. Rhizosphere samples were collected from a normal agricultural soil as well as waste rock and mine tailings. The plants studied were *Acassia sp, Gmelina arborea, Leucaena glauca, Tectonia grandis, Manihot utilissima, and Zea mays*. There was an abundance of mycorrhizal fungi species associated with all observed plants, with the dominant genus being *Glomus*. The spore density varied from 77 – 240 spores/100g in natural soils, with the percentage of infected roots varying from 10 to 40%, and decreasing as the soil was disturbed. Some of these mycorrhizal fungi showed a relatively good ability to grow in the heavy metal contaminated gold mine tailing.

Key words: Artisanal mining, Land degradation, Indigenous species, Mycorrhiza

1. Introduction

Indonesia is one of the main countries where artisanal gold mining is operated. Artisanal mining is defined as an informal and unregulated system of small-scale mining. Most of these activities are illegal; therefore, in the case of gold mining, artisanal mining is called "PETI", an abbreviation of the Indonesian term "Pertambangan Emas Tanpa Izin" (illegal gold mining). A report by the International Institute for Environment and Development in 2001 quantified the number of locations of illegal small-scale mining in Indonesia at 713. These were located in Sumatra, Java, Kalimantan, and Sulawesi (Aspinal, 2001), however artisanal mining has subsequently spread to West Nusa Tenggara.

Artisanal mining usually employs very simple technologies, and there is no planning for rehabilitation after the closure of the mining operation. The most visible outcome of artisanal mining is environmental destruction. This includes heavy metal pollution of land and water surrounding the artisanal mining operation as a result of the disposal of waste rocks and tailings. In some cases, the separation of gold ore is not done at the mining location, and therefore environmental pollution can affect further areas.

Despite its negative effects, artisanal mining plays an essential role in developing societies. Artisanal mining is a significant source of revenue in countries that operate the system. Artisanal mining provides livelihoods for rural communities. Mining provides employment for significant numbers of workers, not only in the mining activities, but also in all related support activities. Upon consideration of these roles, Sir Mark Moody Stewart, the President of the Geological Society of London, during a November 2003 conference on sustainable mining in London, said that "Artisanal mining should be encouraged; however, the associated poor health, safety, and environmental conditions must be improved."

Conventional engineering systems to rehabilitate degraded mining land exist, but are expensive and rely on technology that may be unavailable in the worst affecting artisanal mining areas. Rehabilitation can be done *in situ* and/or *ex situ* through the use of physical, chemical, or biological techniques. *In situ* improvement is favored because of the lower cost and reduced impact on the environment. The use of vegetative technologies, known as "phytoremediation", is recognized as a potentially cost-effective and ecologically sound alternative to degraded land rehabilitation. When used appropriately, phytoremediation can facilitate natural soil formation processes, and can bring about pollutant degradation or removal (Munshower, 1993; Cunningham *et al.*, 1995; Brooks, 1998).

The existence of soil microorganisms, especially those that interact with plant roots, is one of the important keys in determining the success of phytoremediation. In post mined land, because of the disturbance of top soils, there are changes in the microbial community. Severe disturbance and contamination can lead to the complete extinction of normal soil microorganisms. Experience has shown that soil microbial communities in tailings can become severely stressed. Heterotrophic populations become severely diminished, while acidophilic autotrophic bacteria such as *Acidothiobacillus spp.* and *Leptospirillum ferrooxidans* thrive (Southam and Beveridge, 1992). Heterotropic bacteria are very important for plant survival; the interaction between plant and microorganism leads to increased plant nutrient availability and absorption and therefore to increased growth (Mummey et al., 2002; Mendez et al., 2007; Moynahan et al., 2002; Rosario et al., 2007).

The role of mycorrhiza in phytoremediation has attracted research attention (Gaur and Adholeya, 2004; Hidelbrandt *et al.*, 2007). There are at least two mechanisms by which the occurrence of mycorrhiza in plant root promotes the work of phytoremediation. First, as suggested by Galli *et al.* (1994), mycorrhizal colonization in roots plays a role in protecting the plant root from heavy metals, and the second, which is widely known as the mycorrhizal colonization of roots, increases root surface area for nutrient absorption. The extramatrical of fungal hyphae can extend several cm into the soil and encourage the uptake of plant nutrients to the host roots. In their review paper, Gaur and Adholeya (2004) summarized that abuscular mycorrhiza are able to improve plant growth in heavy metal-contaminated sites by improvement of soil properties, increasing plant access to relatively immobile mineral nutrients, and binding heavy metals into roots thus restricting their translocation into shoot tissues.

Many researchers have reported the occurrence of various mycorrhiza in plant roots growing in heavy metal contaminated soils (Shetty *et al.*, 1995; Chaudhry *et al.*, 1999). Ramman *et al.* (1993) reported the presence of abuscular mycorrhiza taxa such as *Glomus* and *Gigaspora spp* in the rhizospheres of fourteen plant species growing in magnesite mine spoil in India. The existence of *Glomus mosseae* in heavily contaminated soils have also been reported by Weissenhorn and Leyval (1995). Pawlowska *et al.* (1996) reported a re-covering of spores of *Glomus aggregatum*, *G. fasciculatum*, and *Entrophospora spp*. on spoil mounds rich in Pb, Cd and Zn. The evidence of extomycorrhizal fungi in plants growing in heavy metal contaminated soils has been reported by Galli *et al.* (1993). It is suggested that the role of this ectomycorrhizal fungi is to protect the roots from toxic heavy metals. Donneley and Fletcher (1994) suggested that the ability of ectomycorrhizal fungus to protect roots from heavy metals is by mycelia, which affords a physical barrier or mantle to inhibit the movement of heavy metals into the xylem.

Although the effectiveness in the protection and promotion of nutrient absorption differs between species, all researchers have agreed that the colonization of plant roots by arbuscular mycorrhiza plays an important role in nutrient absorption (Marschener, 1998). The protection and enhanced capability afforded by greater uptake of plant nutrients will result in a higher plant biomass, a pre-requisite for successful remediation.

One of the key factors for the success of phytoremediation technology is the use of local indigenous species. These indigenous species have well adapted to the soil and climate conditions of the host site, hence these indigenous

plants will ensure the greatest chance for remedial success. It has been shown that abuscular mycorrhizal fungal adapted to local conditions can stimulate plant growth better than non indigenous isolates (Sylvia and Williams, 1992). The best way to initiate species selection is by observing the indigenous species that have grown on disturbed adjacent areas. It is not wise to use species from another place which may have different chemical, physical and biological conditions.

The aim of the work described in this manuscript is to identify the local indigenous mycorrhizal species in soil adjacent to an artisanal mining area. It is expected that the findings of this study will be used to promote the successfull remediation of degraded land at this mining area.

2. Materials and Methods

2.1 Fieldstudy location

A field study was conducted in the Sekotong sub-district, West Lombok, Indonesia. Mining activity at this location was officially closed in 2006. However artisanal mining activity was illegally re-opened in 2009. The occurrence and identity of plants was assessed in the field location, and soil samples collected for mycorrhizal identification.

2.2 Sample collection

Approximately 2 kg of soil was collected from the rhizosphere of each of the plant species Acacia (Acacia spp), Gmelina (Gmelina arborea), Leucaena (Leucaena glauca), Teak (Tectona grandis), Cassava (Manihot utilisima), Maize (Zea mays), and Rice (Oryza sativa), growing in non-contaminated soil. These crops were selected because they grow extensively in the study area. Revegetated waste rock samples were taken from the rhizosphere of Acacia, Gmelina, Leucaena, and Teak. Revegetated tailings samples were taken from the rhizosphere of Acacia, Cassava, Maize and Rice.

2.3 Soil and tailing analysis

All samples were processed for laboratory analysis and mycorrhizal fungi characterization. Soil particle size analysis was performed by the pipette method (Soil Survey Laboratory Staff, 1992). Soil pH was measured in 1:2.5 ratio soil solutions (with de-ionized water) with a pH meter (Jenway 3305).

Organic C content was determined by the wet oxidation method of Walkley and Black (Soil Survey Laboratory Staff, 1992). Total N content, NH_4^+ , and NO_3^- were measured by the Kjeldhal method (Bremner and Mulvaney, 1982). The CEC was extracted with 1M NH_4OAc (buffered at pH 7.0), and exchangeable base concentrations were measured using AAS (Shimatzu).

The heavy metals Fe, Zn, Cu, Ni, Cd, and As were extracted by 0.1M HCl (Jones and Meuhlchen, 1994) and the concentration of these metals in the substrate was measured with AAS (Shimatzu).

2.4 Identification of mycorrhizal fungi

The root samples were stained according to the methods described by Brundrett *et al.* (1984), and the percentage of infected roots was evaluated by the "grid-intersect" method of Givanetty and Mosse (1980).

Mycorrizal spore analysis was done by the method of Daniel and Skipper (1982). The spores were then examined and counted using a dissecting microscope. Taxonomy of AM was used to classify the spores according to the method of de la Cruz (1991)

3. Results and Discussion

3.1 Soil and tailing properties

The field study area soil profile has been described elsewhere (Siswanto *et al.*, 2010), as Lithic dystropept with sandy clay loam to clay loam soil texture, slightly acidic, with low soil organic matter content, low nitrogen and phosphorus content, but high potassium content (Table 1). The concentration of heavy metals in the soil was at a level below the limit of detection of the analytical method used.

The results presented in Table 1 show that waste rock has a lower organic C, N and P content compared to the natural soil, but has a higher content of K and heavy metals. A similar tendency occurred for tailing, except it had a significantly higher heavy metal content.

<Table 1>

3.2 Mycorrhizal fungi

The results of mycorrhizal spore counting, expressed as spore density, are presented in Table 2. The percentage of infected roots is shown in Table 3, and a visual figure of genus *Glomus* and *Gigaspora* observed in this works is is presented as Figure 1.

<Figure 1>

The results in Table 2 show that in general, spore density in the waste rock and mine tailings was lower than that of the natural soil. In the soil, the spore density varied from 77 spores/100g soil to 240 spores/100g soil. The highest spore density (240 spores/100g) was observed in the rhizosphere of cassava, and the lowest (77 spores/100g) was in the rhizosphere of maize. In the mine tailings, the spore density varied from 34 spores/100g soil to 78 spores/100g soil. Again, the highest spore density occured in the rhizosphere of cassava.

<Table 2>

The lower spore density in the rhizopshere of plants growing in waste rocks and tailings indicates that soil disturbance decreases the abundance of mycohizal fungi spores. This is a likely an indicator of decreasing environtmental quality. Sylvia and Williams (1992) found that mining decreases the abundance and the diverty of mycorrhizal fungi in the rizosphere. A decrease in abundance and diversity of mycorrhizal fungi due to heavy metal contaminant had also been shown by Koomen *et al.*, (1998).

The results presented in Table 3 show that the mycorrhizal fungi in the Sekotong artisanal mining area was dominated by genus *Glomus*. All plants observed in this study were associated with this mycorrhizal fungi; and with the exception of maize, all roots of the plants observed were infected by *Glomus mossaeae*. Other mycorrhizal fungi found were genus *Gigaspora*, *Scutellospora*, and *Acaulospora*.

<Table 3>

The abundance of the mycorrhizal fungi varies from low (in maize roots) to high (cassava and acacia), and decreased as the soil was disturbed. The mycorrhizal fungal species *Glomus aggregatum*, *Glomus geosporum*, and *Acaulospora scrobiculata* were not observed in the tailings. This indicates that these three mycorrhizal species, at least under the conditions apparent at the Sekotong study area, do not tolerate the environmental disturbance that is associated with mining

4. Conclusion

It is widely understood that heavy metal contamination at mine sites cannot be chemically degraded. The traditional technology used to neutralize the negative effects of heavy metals is to physically remove the contaminants, either by excavation and subsequent disposal to a landfill site, or by soil washing. However, these methods are costly and simply move the problem to a new location. An alternative method to decrease the negative impact of heavy metals is to imobilize the contaminants in the soil or soil-like medium. Phytoremediation, the use of plants either to remove or to immobilize contaminants and to stabilize soil is becoming increasingly important as an option to rehabilitate degraded land. The system shows promise for use at artisanal mine sites in developing areas of the world. The abundance and diversity of mycorrhizal fungi is recognised as a key component of the soil-plant system in phytoremediation. The occurrence of mycorrhizal fungi can promote plant growth by increasing nutrient absorption (Marschner, 1998) as well as protecting plants from the harmful effect of the toxic metals (Galli *et al.* 1994). However, disturbance of soil will change the abundance and diversity of the mycorrhizal fungi population. Under some conditions, mycorrhizal fungi populations can become severely diminished was diminished (Tables 2 and 3). Fortunately, as this research shows, there are some mycorrhizal fungal species tolerant to the disturbed conditions apparent at artisanal mine sites.

The results in Table 3 show that mycorrhizal fungi belonging to the genus *Glomus* were observed in the roots of most plants growing in gold tailings. These local and indigenous mycorrhizal fungi appear to be tolerant of the chemical and physical conditions at the study site, and could be developed for use as an inoculant in the re-vegetation of the Sekotong artisanal gold mining area.

Acknowledgements

The work was funded by the Indonesian Department of National Education under the scheme of "Research for International Collaboration". Thanks is expressed to Massey University for providing facilities to the third author during Programme of Academic Recharge for the completion of the manuscript.

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Table 1. Some important properties of natural soil, waste rock and mine tailings of the artisanal gold mining area at Sekotong

Properties	Natural soil	Waste rock	Tailings
Clay (%)	24.5	-	26.9
Sand (%)	30.3	-	29.5
рН	6.4	4.7	5.4
C (%)	0.95	0.03	0.32
N (%)	0.10	0.01	0.03
P (mg/kg)	0.98	0.99	0.92
S (mg/kg)	8.92	15.74	18.75
CEC (cmol/kg)	14.25	3.46	19.64
K (cmol/kg)	3.25	0.14	1.53
Ca (cmol/kg)	3.04	1.18	2.53
Mg (cmol/kg)	1.26	0.27	0.30
Na (cmol/kg)	0.89	0.17	0.17
Fe ($\mu g/kg$)	ud	0.002	130
Cu (µg/kg)	ud	0.006	560
Zn ($\mu g/kg$)	ud	0.001	68
Ni (µg/kg)	ud	0.001	67
Cd ($\mu g/kg$)	ud	ud	ud
As (µg/kg)	ud	ud	ud

Ud: undetected

Host plant	Spore density (no of spores/100 g dry materials)			
	Natural soils	Waste rock	Tailings	
Acacia	167	98	46	
Gmelina	98	57	46	
Leucaena	110	48	34	
Teak	78	87	45	
Cassava	240	np	78	
Maize	77	np	np	

Table 2. Mycorrhiza spore density in observed plants for the artisanal mining area of Sekotong, West Lombok

np : there was no plant species observed on this medium

Table 3. Host plants and the associated mycorrhizal species in the artisanal mining area of Sekotong, West Lombok

Host plant and	Abune	Abundance	
the associated mycorrhiza	Natural soils	Waste rock	Tailings
Acacia	high	medium	low
Glomus mossaeae	+	+	+
Gigaspora sp	+	+	+
Scutellospora calospora	+	+	+
Gmelina	medium	low	low
Glomus leptotichum	+	+	+
Glomus mossaeae	+	+	+
Glomus sp	+	+	+
Leucaena	medium	low	very low
Glomus deserticola	+	+	+
Glomus aggregatum	+	+	-
Glomus geosporum	+	+	-
Acaulospora scrobiculata	+	+	_
Teak	medium	low	no plant
Glomus leptotichum	+	+	np
Glomus mossaeae	+	+	np
Glomus fasciculatum	+	+	np
Cassava	high	no plant	low
Glomus Intraradices	+	np	+
Glomus mosaeae	+	np	+
Glomus deserticola	+	np	+
Acaulospora scrobiculata	+	np	-
Maize	low	no plant	no plant
Glomus etunicatum	+	np	np
Glomus caledonium	+	np	np

Abundance was measured by % colonization: very low (<10%); low (10-20%); medium (20-30%); high (>30%). (+) indicates that this micorrhiza species exist in the associated plant and growth medium, and (-) indicates that there was no mycorrhize species in the associated plant and growth medium. No plant (np): there was no plant species found in this growth medium.

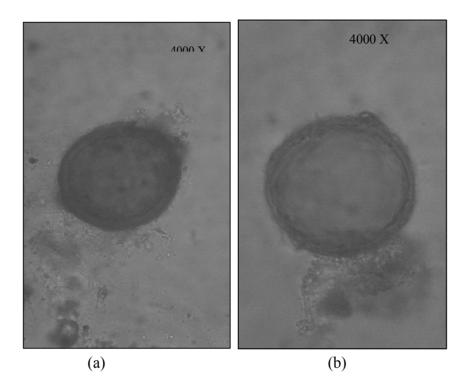


Figure 1. Visual observation of genus *Glomus* (a) and *Gigaspora* (b) in plant roots for Sekotong artisanal gold mining