AJAB

Asian J Agri & Biol. 2018;6(4):556-565.

Original Article

Response of forest tree species inoculated with MycoSilvi and soil ameliorant addition grown in silica sand

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Received:
May 16, 2018

Accepted:
November 02, 2018

Published:
December 31, 2018

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Abstract

Open mining of silica sand causes some environmental impacts such as declining levels of soil fertility, lowering soil pH, and increasing toxic acid mine waste such as Al, Fe, Cu, and Zn. Soil quality may be improved by the addition of soil ameliorant and MycoSilvi, an inoculum of arbuscular mycorrhizae enriched by mycorrhizal helper bacteria. This study aimed to analyze the growth responses of *Falcataria moluccana*, *Samanea saman*, and *Cassia siamea* seedlings by the addition of soil ameliorant and MycoSilvi grown in soil media from silica sand post mining. The experimental design used in this study was completely randomized design with split pot design that consists of two treatment factors (MycoSilvi and soil ameliorant) with five replications. The main plot was MycoSilvi that consists of two levels (without MycoSilvi and with MycoSilvi). The subplot was soil ameliorant that consists of six levels [(1) compost 0 g and lime 0 g, (2) compost 0 g and lime 3.6 g, (3) compost 0 g and lime 7.2 g, (4) compost 32.5 g and lime 0 g, (5) compost 32.5 g and lime 3.6 g, and (6) compost 32.5 g and lime 7.2 g]. The interaction of MycoSilvi and soil ameliorant significantly increased height, diameter, biomass, chlorophyll content, and mycorrhizal colonization of *F. moluccana*, *S. saman*, and *C. siamea*. The MycoSilvi and soil ameliorant (32.5 g of compost and 7.2 g of lime) was the best treatment for the growth and chlorophyll content of *F. moluccana*, *S. saman*, and *C. siamea* study species.

Keywords: Forestry plants, MycoSilvi, Silica sand post mining land, Soil ameliorant

Introduction

Mining activities in Indonesia continue to develop. Statistical data of the Ministry of Environment and Forestry of Indonesia (2016) indicate that the number of forest concession for the operation of the mine production between 2010-2015 was around 562 units, with a total area of 457,614 ha. One of the mining activities is silica sand mining. Silica sand (quartz sand) consists of crystals of silica (SiO_2) and other compounds that get carried away during the deposition process. Silica sand is used as a source of silica on glass-making manufacture and cement industry (Hamer and Hamer, 2004; Taylor, 2004).

Open mining including silica sand mining causes some environmental impacts such as development of acidic soils and waters, erosion of tailings by wind and water, and physical disturbances to the landscape (Hudson et al., 1999). Open mining activities also increase toxic elements to plants such as Al, Fe, Mn, Ni, Zn, Co and Cd (Wahab and Marikar, 2012).

Soil quality of post mining silica sand can be improved by the addition of soil ameliorant. According to Oklima et al. (2014) soil ameliorant is a substance added to soil that aimed to improve low soil

fertility condition especially to increase nutrient content for plant growth. Soil ameliorants that commonly use are lime and compost. Liming can raise pH of the soil that causes nutrient elements such as P, which is not tied by Al so that it can easily absorbed by the plants (Robson, 1989). According to Anastas and Crabtree (2009) the addition of compost changes pH, improves the stability of soil aggregates, provides food to the soil microorganisms and nutrients to the plants.

In addition to soil ameliorant, soil quality of post mining silica sand is also can be improved by the addition of biological fertilizers which contain living organisms. One type of biological fertilizer is Arbuscular Mycorrhizal Fungi (AMF), the symbiotic relationship of the mutualism between the fungi and the plant roots that can form an arbuscular structure (Smith and Read, 2008). AMF may absorb nutrient and water from the soil by expand of its external hypha when the roots of the plants are no longer able to take it. This phenomenon usually occurs in the plants on marginal or used mine soil. AMF can increase the absorption of nitrogen, potassium, and phosphate (Hartoyo et al., 2015). We develop an arbuscular mycorrhizal inoculum enriched with Mycorrhizal Helper Bacteria (MHBs), called MycoSilvi. Previous study showed that MHBs isolated from AMF spores can increase spores production and mycorrhizal root colonization (Budi and Christina, 2012).

Selected plants to revegetation post mining should be drought-resistant and fast growing species with dense canopy and root system (Caravaca et al., 2002; Mendez and Maier, 2008). These kinds of trees in the family of Fabaceae are recommended because it may form symbiosis with nitrogen-fixing bacteria such as *Rhizobium* sp. that are capable in forming rhizobia on the root system. In this research we select three species from Fabaceae as *Falcataria moluccana* (Miq.) Barneby & J.W. Grimes, *Samanea saman* (Jacq.) Merr, and *Cassia siamea* Lam.

The aim of this study was to analyze the growths of *F. moluccana*, *S. saman*, and *C. siamea* seedlings by the addition of soil ameliorant and MycoSilvi grown in post mining silica sand.

Material and Methods

Soil sampling and analysis

Soil was taken in the area of post silica mining PT Holcim Indonesia Tbk., located at Sukabumi District, West Java (06º55'18.7" S and 106 º47'10.0"E) on Saturday 30th July 2016. Soil was taken in the surface (0-20 cm depth) at two points, the distance of point sampling to another point approximately 50 m. The soil was then sieved (2 mm), air dried, and composited as a soil sample. About 1 kg of composite soil sample was analysed at the Laboratory of Soil Science and Land Resources, Faculty of Agriculture, Bogor Agricultural University. Chemical characteristics of soil post mining are shown in Table 1.

Table 1: Chemical characteristics of silica sand post mining soil

Chemical characteristics	Analysis result	Criteria*
pH (H_2O)	3.37	Very acid
C-organic (%)	1.20	Low
N-total (%)	0.11	Low
P-available (ppm)	2.95	Very low
P-total (ppm)	139.45	Very high
Ca (cmol/kg)	0.22	Very low
Mg (cmol/kg)	0.14	Very low
K (cmol/kg)	0.22	Low
Na (cmol/kg)	0.12	Low
CEC (cmol/kg)	10.06	Low
Al (cmol/kg)	8.12	High
Fe (ppm)	116.69	High
Cu (ppm)	2.44	Normal
Zn (ppm)	1.10	Normal
Sand (%)	24.9	
Silt (%)	36.46	
Clay (%)	38.63	

* Estevan et al. (2013)

Preparation of MycoSilvi inoculum

Bacteria isolated from surface sterilized AMF spores that were shown as Mycorrhizal Helper Bacteria (MHBs) from previously research (Budi and May, 2013) were rejuvenated on media of nutrient broth for 36 hours. *Pureria javanica* seedlings grown in zeolit medium were inoculated with AMF spores of *Glomus* sp. and then inoculated with approximately 5 ml MHBs containing 10^{-9} CFU/ml. The plants were maintained for 2 months in the green house. After harvesting, the number of AMF spores were determined. This AMF inoculum is called as MycoSilvi.

Seed germination of *S. saman*, *C. siamea*, and *F. moluccana*

F. moluccana seeds were immersed in water of 80°C for 15 minutes following cold water of 10°C for 24 hours. The seeds were germinated on the sterilized sand and charcoal media. *S. saman* seeds were immersed in water of 80°C for two minutes following warm water with a temperature of 30°C for 24 hours. The seeds were then germinated on the sterilized zeolite. *C. siamea* seeds were immersed in water of 80°C until the water cooling down. The seeds were then germinated on the sterilized zeolite. All seeds were placed in the green house.

Preparation of planting media and the addition of soil ameliorant

The soil was filled into the heat resistant plastic then sterilized on autoclave with temperature of 121°C for one hour. Sterilized soil was then added with soil ameliorant (lime and compost) or not as a control according treatment design. The soil was filled to the polybag with size 15 cm x 20 cm.

MycoSilvi inoculation

The seedlings were transplanted to the soil media in the polybag. 10 g MycoSilvi (approximately containing 50 AMF spores) were placed near the roots of seedlings. The seedlings were the placed in the green house for sixteen weeks.

Harvesting and parameter measurement

Plants were harvested sixteen weeks after planting and evaluated for their height, diameter, chlorophyll content, shoot and root dry weight, and mycorrhizal colonization.

- **Height**
  Plant height was measured by ruler from the base of the stem to the point of growing shoots of seedlings
- **Diameter**
  Plant diameter was measured by caliper at 1 cm above the base of the seedlings stems
- **Chlorophyll content**
  Chlorophyll content was measured by SPAD chlorophyll meter. Three leaves were collected per plant and three SPAD measurements were taken per leaf and averaged.
- **Shoot and root weights**
  Shoot and root weights were recorded after drying at 105°C to constant weight
- **Mycorrhizal colonization**
  Mycorrhizal colonization was evaluated after cleaned with 2.5% KOH, soaked with HCl 0.1 M, stained with trypan blue, and soaked with acid lactic liquid as destaining (Clapp et al., 1996). Percentage of mycorrhizal colonization was determined according to the method of O'connor et al. (2001).

Experimental design and data analysis

The experimental design used in this study was completely randomized design with split pot design that consists of two treatments (the addition of MycoSilvi and soil ameliorant).

Addition of MycoSilvi consists of two levels:
A0 = Without MycoSilvi
A1 = With MycoSilvi

Addition of soil ameliorant consists of six levels:
B0 = Compost 0 g and lime 0 g
B1 = Compost 0 g and lime 3.6 g
B2 = Compost 0 g and lime 7.2 g
B3 = Compost 32.5 g and lime 0 g
B4 = Compost 32.5 g and lime 3.6 g
B5 = Compost 32.5 g and lime 7.2 g

The interaction of two factors form 12 treatments with five replications. The treatments was tested on *F. moluccana*, *S. saman*, and *C. siamea*. All data were analyzed by using ANOVA and followed by further tests using Duncan Multiple Range Test (DMRT) at 5% level by using the SAS 9.1.3. Software.

Results

Recapitulation of variance analysis of seedlings growth at Table 2 shows that the interaction of MycoSilvi and soil ameliorant were significant increased height, diameter, chlorophyll content, biomass, and mycorrhizal colonization on *F. moluccana*, *S. saman*, and *C. siamea*.

Table 2: Recapitulation of analysis of variance of seedlings growth

Plant Species	No	Parameter	A	B	A x B
F. moluccana	1	Height (cm)	**	**	**
	2	Diameter (mm)	**	**	**
	3	Biomass (g)	**	**	*
	4	Chlorophyll content (µg/cm^2)	*	**	*
	5	Mycorrhizal colonization	**	**	**
S. saman	1	Height (cm)	**	**	*
	2	Diameter (mm)	**	**	*
	3	Biomass (g)	**	**	*
	4	Chlorophyll content (µg/cm^2)	*	**	*
	5	Mycorrhizal colonization	**	**	**
C. siamea	1	Height (cm)	**	**	**
	2	Diameter (mm)	**	**	**
	3	Biomass (g)	**	**	*
	4	Chlorophyll content (µg/cm^2)	*	**	*
	5	Mycorrhizal colonization	**	**	**

A = MycoSilvi, B = Soil ameliorant, ** = P≤0.0001 and * = 0.0001<P≤0.05

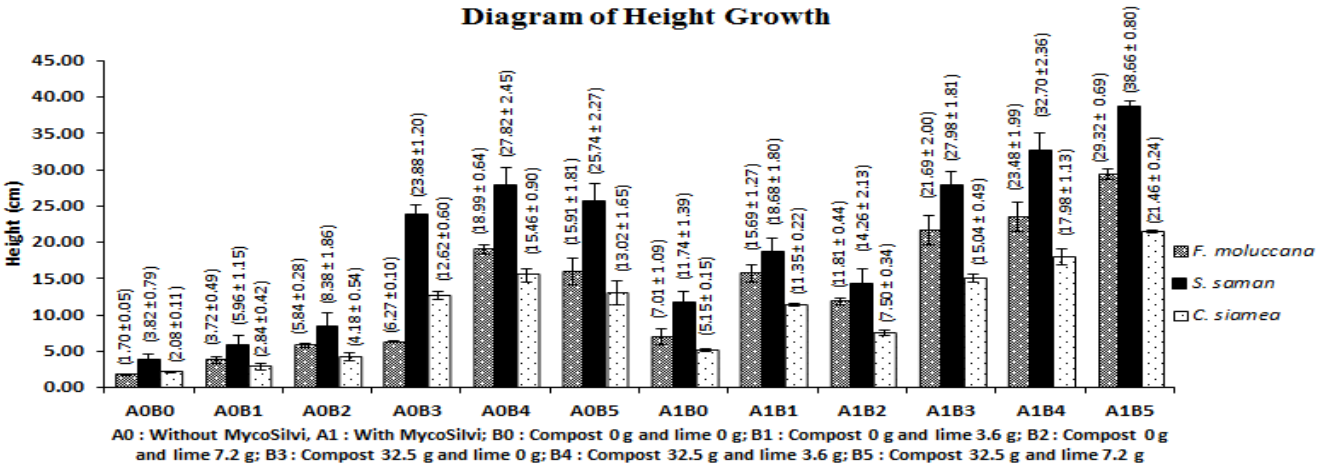

Fig. 1: The effect of interaction of MycoSilvi and soil ameliorant to height growth of seedlings

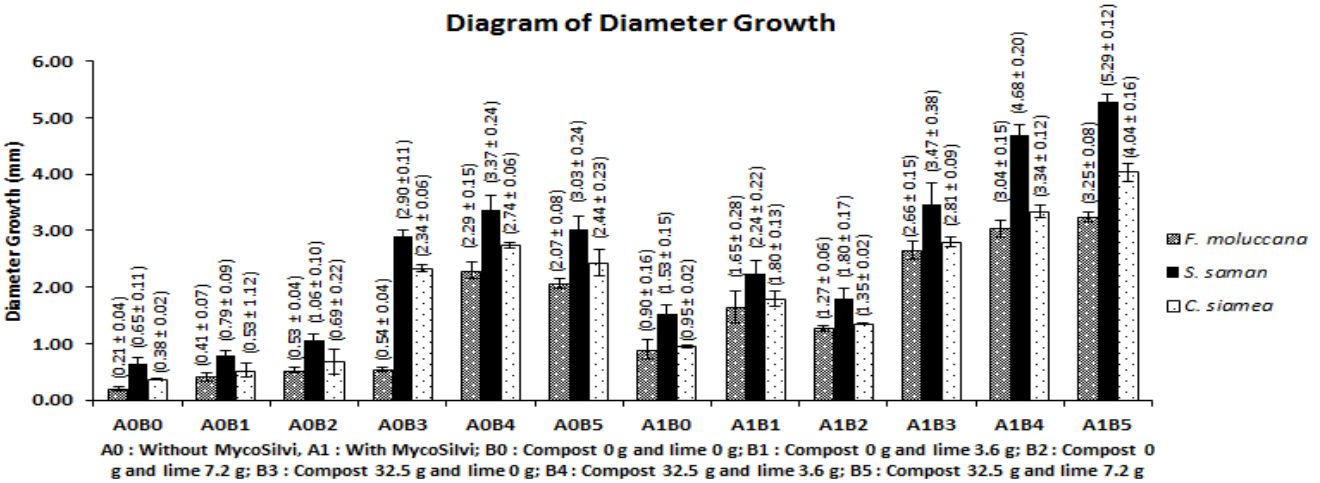

Fig. 2: The effect of interaction of MycoSilvi and soil ameliorant to stump diameter growth of seedlings

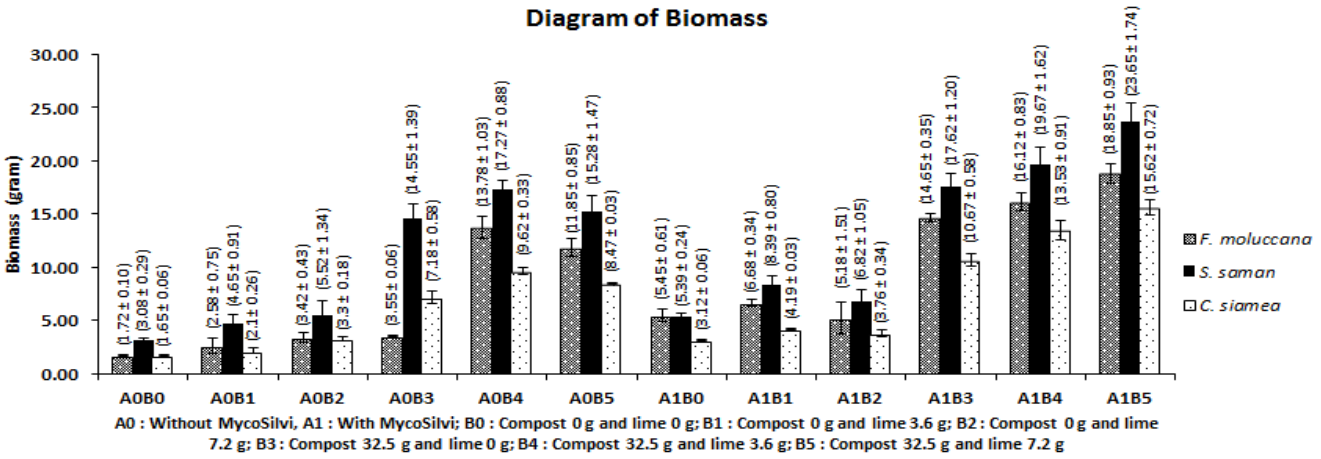

Fig. 3: The effect of interaction of MycoSilvi and soil ameliorant to biomass of seedlings

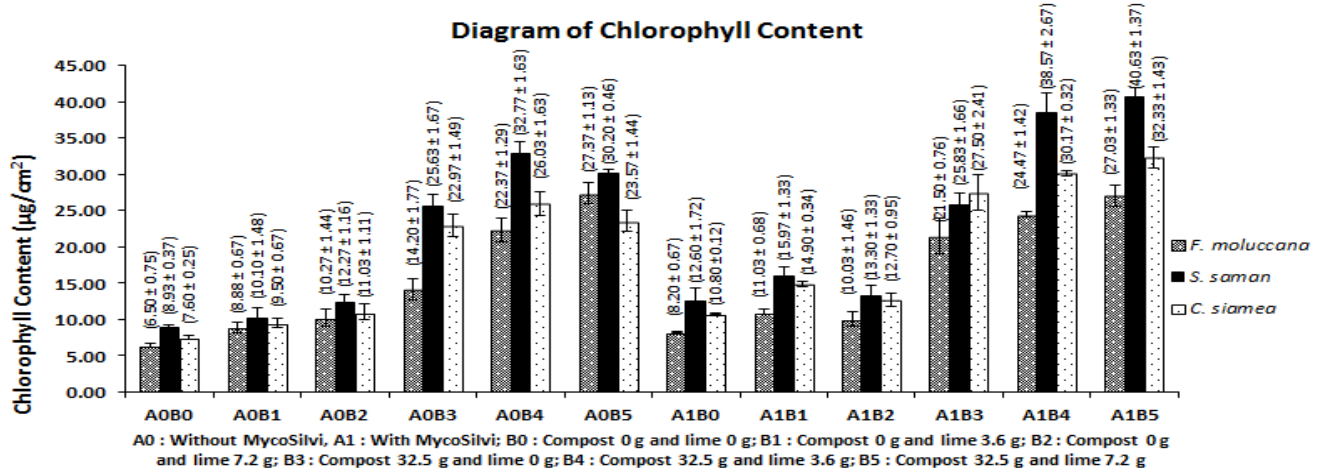

Fig. 4: The effect of interaction of MycoSilvi and soil ameliorant to chlorophyll content of seedlings

Plants growth

The interaction of MycoSilvi and soil ameliorant were significant increased height, diameter, and biomass growth on *F. moluccana*, *S. saman*, and *C. siamea* as shown in Fig. 1, 2, and 3

Chlorophyll content

The interaction of MycoSilvi and soil ameliorant were significant increased chlorophyll content of *F. moluccana*, *S. saman*, and *C. siamea* as shown in Fig. 4.

Mycorrhizal colonization

The interaction of MycoSilvi and soil ameliorant was significantly increased mycorrhizal colonization. DMRT result in Table 3 shows that the MycoSilvi and soil ameliorant (0 g of compost and 3.6 g of lime) (A1B1) increased mycorrhizal colonization higher than other treatments on *F. mouccana*, *S. saman*, and *C. siamea*.

Table 3: The effect of the addition of MycoSilvi and soil ameliorant on mycorrhizal colonization

Treatment	Mycorrhizal colonization			Criteria*
	F. moluccana	*S. saman*	*C. siamea*	
A0B0	0.00[f]	0.00[f]	0.00[f]	Not colonized
A0B1	0.00[f]	0.00[f]	0.00[f]	Not colonized
A0B2	0.00[f]	0.00[f]	0.00[f]	Not colonized
A0B3	0.00[f]	0.00[f]	0.00[f]	Not colonized
A0B4	0.00[f]	0.00[f]	0.00[f]	Not colonized
A0B5	0.00[f]	0.00[f]	0.00[f]	Not colonized
A1B1	90.00[a]	86.67[a]	73.33[a]	High
A1B4	70.00[b]	66.67[b]	56.67[b]	High
A1B2	50.00[c]	43.33[c]	40.00[c]	High
A1B5	40.00[cd]	36.67[cd]	33.33[c]	High
A1B3	30.00[de]	26.67[de]	20.00[d]	Medium
A1B0	26.67[e]	23.33[e]	20.00[d]	Medium

Numbers followed by same letter are not significant different (P = 0.05) by DMRT test
*O'connor *et al.* (2001)

Discussion

Characteristics of media

Table 1 shows that silica post mining soil at PT Holcim Indonesia Tbk in Sukabumi, West Java, had low nutrient content, very acid soil pH, and high Al and Fe content that are toxic to plants. Nutrients content such as C-organic, N-total, P-available, Ca, Mg, K, Na were very low so that the plants were not able to grow well. The soil also had low CEC. The low pH value caused the accumulation of Al element that lead to P-fixation which is unavailable for plants (Robson, 1989).

The addition of 3.6 g of lime can increase pH to acid condition and increase C-organic, N, P-available from very low to low category. The addition of 7.2 g of lime can increase pH to neutral condition and increase C-organic, N, P-available to low category. MycoSilvi can increase C-organic, N, P-available to low category but the pH condition still very acid. The addition of 3.6 of lime, compost, and MycoSilvi can increase pH to neutral condition and increase C-organic to medium and P-available to very high category. The addition of 7.2 of lime, compost, and MycoSilvi can increase pH to neutral condition and increase C-organic to medium category, N to low category, and P-available to very high category.

Plant height and diameter

Analysis of variance seedlings growth in Table 2 showed that the combination of MycoSilvi and soil ameliorant was significantly increase height and diameter growth. The interaction between MycoSilvi, compost, and lime were very effective for increasing the height and diameter of the plant.

The addition of 3.6 g of lime (A0B1) can increase height and diameter growth of *F. moluccana*, *S. saman*, and *C. siamea* as compare to control (A0B0). Height growth of *F. moluccana* increase 118.82% and diameter growth increase 95.24%. Height growth of *S. saman* increase 56.02% and diameter growth increase 21.54%. Height growth of *C. siamea* increase 36.54% and diameter growth increase 27.27%. This is due to 3.6 g of lime can increase soil pH from very acid to acid condition so that nutrients avaibility are increase.

The addition of 7.2 g of lime (A0B2) can increase height and diameter growth of *F. moluccana*, *S. saman*, and *C. siamea* as compare to control (A0B0). Height growth of *F. moluccana* increase 243.53% and diameter growth increase 152.38%. Height growth of *S. saman* increase 119.37% and diameter growth increase 61.54%. Height growth of *C. siamea* increase 100.96% and diameter growth increase 89.09%. This is due to 7.2 g of lime can increase soil pH from very acid to neutral condition so that so that nutrients are available to plants. According to Robson (1989) available form of nutrients can be absorbed by plants on pH conditions around the neutral (pH = 7). This is due to most of the nutrient elements are soluble easily in water on that condition.

The addition of 7.2 g of lime and MycoSilvi (A1B2) can increase height and diameter growth of *F. moluccana*, *S. saman*, and *C. siamea* as compare to plant without MycoSilvi (A0B2). Height growth of *F.*

moluccana increase 97.38% and diameter growth increase 56.00%. Height growth of *S. saman* increase 113.08% and diameter growth increase 185.70%. Height growth of *C. siamea* increase 176.30% and diameter growth increase 226.70%. This result shows that MycoSilvi can increase plant height and diameter growth. This is because MycoSilvi contains AMF that can increase the absorption of nutrient elements from the soil due to they have network of external hypha that able to expand the field of nutrient absorption.

The addition of 7.2 g of lime, compost, and MycoSilvi (A1B5) can increase height and diameter growth of *F. moluccana*, *S. saman*, and *C. siamea* as compare to without compost (A1B2). Height growth of *F. moluccana* increase 173.19% and diameter growth increase 186.79%. Height growth of *S. saman* increase 233.71% and diameter growth increase 303.48%. Height growth of *C. siamea* increase 257.56% and diameter growth increase 562.08%. This is due to compost contained the high C-organic, P-total, and N-total nutrient that required for plant growth. This treatment can increase soil pH to neutral condition, C-organic to medium category, and P-available to very high category. This result indicated that there is a synergistic effect of compost, MycoSilvi, and lime with appropriate dosage for improving plant growth in marginal soil medium.

High content of C-organic in compost was able to increase the content of organic matter in the planting media. According to Allison (1973) high organic matter can also improve the cation exchange capacity (CEC). Soil with High CEC can hold nutrient elements. N element have function in the formation of amino acids that are the constituents of proteins. Amino acids are used to form the protoplast where cell division for plant growth. N is also a major part of the chlorophyll molecules that are used in the process of photosynthesis. The P element plays an important role in the process of photosynthesis and respiration as a storage of energy in ATP and ADP form. The P element is needed in great numbers especially in young cells such as on the ends of the roots and shoots where cell division occurs quickly and high metabolism (Silva and Uchida, 2000).

Compost that were used also contains other nutrient elements needed for plant growth. K element plays an important role as the activator of the enzyme that can increase metabolism. It also plays a role in enhancing photosynthate translocations for plant growth. Ca element plays an important role as activator of the enzyme in the synthesis of protein and carbohydrate transfer. Ca can increase soil pH when liming. Mg element is a major part of the chlorophyll. Chlorophyll plays a role in the process of photosynthesis as a catcher of light energy that will form the ATP and NADPH.

DMRT result shows that the MycoSilvi and soil ameliorant (32.5 g of compost and 7.2 g of lime) on *F. moluccana*, *S. saman*, and *C. siamea* produced highest height and diameter growth than other treatment. Height growth of *F. moluccana* increase 1624.71% and diameter 1447.62% as compare to control. Height growth of *S. saman* increase 912.04% and diameter 713.85% as compare to control. Height growth of *C. siamea* increase 931.73% and diameter 846.67% as compare to control.

Biomass

The plant biomass consist of all the matters in plant which is derived from the results of carbon assimilation (Hopkins, 2006). Biomass also showed ability of plant to take nutrient elements of planting media to support its growth. Plant biomass are affected by the AMF's role in absorbing water and nutrient elements especially phosphate (Facelli et al., 2009). Results of analysis of variance seedlings growth in Table 2 shows that the combination of MycoSilvi and soil ameliorant were significantly effect on biomass parameter on *F. moluccana*, *S. saman*, and *C. siamea*.

The addition of 7.2 g of lime (A0B2) can increase biomass of *F. moluccana*, *S. saman*, and *C. siamea* as compare to control (A0B0). This treatment increase 98.84% of *F. moluccana* biomass, 79.22% of *S. saman* biomass, and 100% of *C. siamea* biomass as compare to the control plant. This is due to 7.2 g of lime can increase soil pH from very acid to neutral condition so that so that nutrients are available to plants. According to Robson (1989) available form of nutrients can be absorbed by plants on pH conditions around the neutral (pH = 7). This is due to most of the nutrient elements are available for plant growth.

The addition of 7.2 g of lime and MycoSilvi (A1B2) can increase biomass of *F. moluccana*, *S. saman*, and *C. siamea* as compare to plant without MycoSilvi (A0B2). This treatment increase 51.46% of *F. moluccana* biomass, 23.55% of *S. saman* biomass, and 13.94% of *C. siamea* biomass as compare to plant without MycoSilvi. This relates to the MycoSilvi that containing AMF that can absorb the nutrient elements and water to form plant biomass.

The addition of 7.2 g of lime, compost, and MycoSilvi (A1B5) can increase biomass of *F. moluccana*, *S. saman*, and *C. siamea* as compare to plant without compost (A1B2). This treatment increase 263.90% of *F. moluccana* biomass, 246.77% of *S. saman* biomass, and 315.43% of *C. siamea* biomass as compare to plant without compost. This is due to compost contained the high P-total, N-total, K, and Mg nutrient that required to produce plant biomass.

DMRT result shows that the MycoSilvi and soil ameliorant (32.5 g of compost and 7.2 g of lime) on *F. moluccana*, *S. saman*, and *C. siamea* produced highest biomass than other treatment. This treatment increase 995.93% of *F. moluccana* biomass, 667.86% of *S. saman* biomass, and 1447.62% of *C. siamea* biomass as compare to the control plant. This treatment also have the highest height and diameter growth on *F. moluccana*, *S. saman*, and *C. siamea*.

Chlorophyll content

Cells on the leaves have microscopic structures called chloroplasts which contain the green pigment or chlorophyll. Chlorophyll absorbs the incoming solar energy and sets into motion that truly remarkable cascade of energy-transforming reactions called photosynthesis (Hopkins, 2006). The content of chlorophyll in the leaves will affect the reactions of photosynthesis and the resulting photosyntat. The more chlorophyll content on leaves will maximize the photosynthesis so that photosyntat generated will be even greater.

The addition of 7.2 g of lime (A0B2) can increase chlorophyll content of *F. moluccana*, *S. saman*, and *C. siamea* as compare to control (A0B0). This treatment increase 58.00% of *F. moluccana* chlorophyll content, 37.40% of *S. saman* chlorophyll content, and 45.13% of *C. siamea* chlorophyll content as compare to the control plant. This is due to 7.2 g of lime can increase soil pH from very acid to neutral condition that could facilitate the avaibility of nutrient in the soil that easily absorbed by the roots plants especially N and Mg which is a main element of chlorophyll.

The addition of 7.2 g of lime and MycoSilvi (A1B2) can increase chlorophyll content of *F. moluccana*, *S. saman*, and *C. siamea* as compare to plant without MycoSilvi (A0B2). This treatment increase 1.56% of *F. moluccana* chlorophyll content, 8.39% of *S. saman* chlorophyll content, and 15.14% of *C. siamea* chlorophyll content as compare to plant without MycoSilvi. This relates to the MycoSilvi that containing AMF that can absorb the nutrient elements in the soil especially N and Mg which is a main element of chlorophyll.

The addition of 7.2 g of lime, compost, and MycoSilvi (A1B5) can increase chlorophyll content of *F. moluccana*, *S. saman*, and *C. siamea* as compare to plant without compost (A1B2). This treatment increase 159.16% of *F. moluccana* chlorophyll content, 205.49% of *S. saman* chlorophyll content, and 154.57% of *C. siamea* chlorophyll content as compare to plant without compost. This is due to compost contained the high N and Mg which is a main element of chlorophyll.

Results of analysis of variance seedlings growth in Table 2 showed that the MycoSilvi and soil ameliorant were significantly increase chlorophyll content. DMRT result shows that the MycoSilvi and soil ameliorant (32.5 g of compost and 7.2 g of lime) on *F. moluccana*, *S. saman*, and *C. siamea* produced highest chlorophyll content than other treatment. This treatment increase 321.08% chlorophyll content of *F. moluccana*, 354.98% chlorophyll content of *S. saman*, and 325.39% chlorophyll content of *C. siamea* as compare to the control plant. This treatment also have the highest height, diameter, and biomass on *F. moluccana*, *S. saman*, and *C. siamea*.

Mycorrhizal colonization

AMF consists of typical structure, such as vesicles, arbuscule, and hypha (Brundrett et al., 1996). Arbuscule is a smooth internal hypha branches found in the cells of the cortex in the plant root. Vesicles have function as a food storage organ containing fatty compounds. Internal hypha is the growing hypha that grow in the cell cortex which will form colony and develop into vesicles and arbuscular.

Results of analysis of variance seedlings growth in Table 2 showed that the MycoSilvi and soil ameliorant were significantly effect on percentage of mycorrhizal colonization at the end of observation. According to Rozas et al. (2017) percentage of mycorrhizal colonization can be determined by the existing of hypha structure, vesicles, and arbuscular. Percentage of mycorrhizal colonization is related to soil pH and content of P and N element. DMRT results in Table 3 showed that addition of MycoSilvi and soil ameliorant (0 g of compost and 3.6 g of lime) (A1B1) have highest percentage of mycorrhizal colonization than other treatment on *F. moluccana*, *S. saman*, and *C. siamea*.

Soil pH condition influence on activities of enzymes that play a role in AMF spore germination (Robson,

1989). Addition of MycoSilvi and soil ameliorant (0 g of compost and 3.6 g of lime) (A1B1) that produced the highest percentage of mycorrhizal colonization had acid pH condition. The pH condition of A1B1 treatment as much as 5.02 on *F. moluccana*, 5.12 on *S. saman*, and 4.93 on *C. siamea*. This is in accordance with statement of Robson (1989) that Acaulospora sp. is found in the conditions of pH 4.5-6.4. The appropriate pH conditions for the growth of Acaulospora sp. cause the treatment produced highest percentage of mycorrhizal colonization.

Percentage of mycorrhizal colonization was also influenced by the content of P and N. Treatment of MycoSilvi and soil ameliorant (0 g of compost and 3.6 g of lime) (A1B1) that produced highest percentage of mycorrhizal colonization with high category (> 30%) had N content with low category (0.1-0.2%) and P available content is very low (< 4 ppm). Treatment of MycoSilvi and soil ameliorant (32.5 g of compost and 0 g of lime) (A1B5) that produce the best treatment to increase plant height, diameter, biomass, and chlorophyll content produced percentage of mycorrhizal colonization lower than A1B1 due to high content of P element (11-15 ppm). This is in accordance with the research of Rozas et al. (2017) that the higher P-available content causes declining of hypha, vesicles, and percentage of mycorrhizal colonization. Research of Farzaneh et al. (2011) also showed that percentage mycorrhizal colonization getting smaller due to high P-available content.

Conclusion

The interaction of MycoSilvi and soil ameliorant significantly increased height, diameter, biomass, chlorophyll content, and mycorrhizal colonization of *F. moluccana*, *S. saman*, and *C. siamea* in soil media from silica sand post mining. The MycoSilvi and soil ameliorant (32.5 g of compost and 7.2 g of lime) was the best treatment to increased height, diameter, biomass, and chlorophyll content of *F. moluccana*, *S. saman*, and *C. siamea* in soil media from silica sand post mining.

Acknowledgment

The authors thank Directorate for Research and Community Services, Ministry of Research, Technology, and Higher Education, the Republic of Indonesia, for providing the grant accordance with the Letter Agreement of Implementation Research Grant No. 079/SP2H/LT/DPRM/11/2016. The authors also thank PT Holcim Indonesia Tbk., Sukabumi, West Java, for providing materials in this research.

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