Original Research Article

Effect of Type and Dose of Arbuscular mycorrhizal Fungi to Growth and Yield of Citronella Grass (*Cymbopogon nardus* L.) in Former Mining Area Land in Sawahlunto

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ABSTRACT

Aims: The study aimed to determine the type and dose of arbuscular mycorrhizal fungi (AMF) to growth and yield of citronella grass (*Cymbopogon nardus* L.) in former mining area land in Sawahlunto, West Sumatera, Indonesia.
Study design: Factorial completely randomized design
Place and Duration of Study: The research was conducted in shade net house of Faculty of Agriculture, Andalas University, Padang, West Sumatera, Indonesia from September 2021-January 2022.
Methodology: Two factors were used in the study. First factor was types of Arbuscular mychorrhizal fungi (AMF), *Glomus* sp., *Gigapora* sp. dan multispores (*Glomus* sp. + *Gigaspora* sp.) and the second factor was AMF doses that consisted of four degrees, 5 g, 10 g, 15 g and 20 g/plant. The experiment result was analyzed by using Analysis of variance (ANOVA) in 5% and continued by using Duncan's New Multople Range Test (DNMRT) in 5%.
Results: Type of AMF multispores in 20 g/ plant could number of tiller. Dose 20 g/plant could plant height and number of leaves. AMF multispores only increased number of tiller and net assimilation rate.
Conclusion: Type and dose of AMF affected the agronomy parameter of citronella grass in former mining area land.

Keywords: Arbuscular mycorrhizal fungi (AMF), Cymbopogon nardus L., mining area land

1. INTRODUCTION

Citronella grass (*Cymbopogon nardus* L.) is an valuable aromatic plant by producing essential oil, non-oil and gas commodity from estate crop sub-sector [1]. Essential oil was known due to it consisted of essence of aromatic plant. Aromatic plant is the plant which has scent that needed for perfume industry, cosmetics, pharmacy industry or medicine and food and beverage industries [2].

Citronella grass is widely spread in Indonesia. According the data of Ministry of Agriculture of Republic of Indonesia, citronella grass area in Indonesia in 2014 was 25,000 ha and the essential oil production was 3,152 thousands ton per year [3]. The production center of citronella grass in Indonesia was West Java, Central Java, Aceh and West Sumatera. In West Sumatera, the producer area of this plant was Solok, Sawahlunto, Pasaman and Mentawai Island [4].

Citronella grass is herbaceous and tropical plant. The characteristic of this plant was it can grow in fertile and marginal soil, pH 3-6, rapid growth, adaptive, dense number of roots so they can hold soil, lush leaves and potential for multiple valuable commodity due to it can convert land and producing essential oil [5]. The integration of cultivation and processing industry of citronella grass has positive impact for land reclamation and also for rural community economic empowerment [6].

West Sumatera is one of biggest mining product contributor (Particularly coal) and Sawahlunto is the biggest one in West Sumatera. In this region, 54% area is mining area. In 2018, licensed active mining was 14,987 ha and land opening area was 6,209. From this land opening area, 1,989 ha was re-vegetation and rest of them, 68% of land was still abandoned and not used yet by public [7].

The former mining area was opportunity to expand agricultural land. From technical side, former mining area can be used for crop cultivation if the land improvement was conducted. From physical, chemical and biology of soil, the used plant In this land should have high adaptation ability in marginal land [8] [9].

Citronella grass is a plant that can grow up 1,800 m a.s.l eventhough it grow well in 250 m a.s.l. This plant even can grow in poor nutrient land. With high adaptation ability in marginal land, this plant can be classified as pioneer plant and can be used for land conservation [10].

As pioneer plant, the growth in former mining area land was not well and the production was low because the land damage after coal mining was significant. One of alternative was to maximize citronella grass growth for optimal production and sustainable is arbuscular mycorrhizal fungi (AMF) use.

Arbuscular mycorrhizal fungi (AMF) is a soil microorganism that helping in nutrient cycle. AMF is symbiotic association between plant root and fungi [11]. Generally, root plant could optimize water and mineral absorption so that it could maximize root absorption ability to absorb water and nutrient in growth and and development in former coal mining area land. AMF works by infecting host plant root system in producing external hypha tissue that grow expansively and penetrate sub soil layer. This condition can increase root capacity in water and nutrients absorption. Several genus of mycorrhizal were reported live in marginal land such as *Glomus* sp. And *Gigaspora* sp. [12]. The research aimed to determine the type and dose of arbuscular mycorrhizal fungi (AMF) to growth and yield of citronella grass (*Cymbopogon nardus* L.) in former mining area land in Sawahlunto, West Sumatera, Indonesia.

2. MATERIAL AND METHODS

The research was conducted in shade net house of Faculty of Agriculture, Andalas University, Padang, West Sumatera and Laboratory of BPSMB, Padang, West Sumatera, Indonesia from September 2021-January 2022. The material was citronella grass clone Mahapengiri G1, AMF genus *Glomus* sp. And *Gigaspora* sp. that were obtained from Soil Microbiologist of Faculty of Agriculture, Andalas University, planting media (former mining area land soil of Bukit Asam company, located in Kandi, Talawi district, Sawahlunto, West Sumatera, Indonesia), poly bag 45 cm x 45 cm in size, manure 94.25 g/ poly bag, urea fertilizer 0.7065 g/ poly bag, SP-36 fertilizer 0.283 g/ poly bag, KCl fertilizer 0.424 g/ poly bag.

2.1 Method

Completely randomized design in 3 x 4 treatment and three replications in factorial was used in the research. The first factor was type of AMF that consisted of three degrees, single *Glomus* sp. inoculant (A1), single *Gigaspora* sp. inoculant (A2) and multispores inoculant of *Glomus* sp. and *Gigaspora* sp. The second factor was AMF dose that consisted of four degrees, 5 g AMF/ plant (B1), 10 g AMF/ plant (B2), 15 g AMF/ plant (B3) and 20 g AMF/ plant (B4). The treatments were replied three times. The soil sample was analyzed in Laboratory of Soil, Faculty of Agriculture, Andalas University.

The soil for plant media was obtained from former mining area land soil of Bukit Asam company, located in Kandi, Talawi district, Sawahlunto, West Sumatera, Indonesia. The depth of soil was 30 cm from soil surface. The obtained soil was sterilized previously before filling to poly bag.

The clone Mahapengiri G1 was obtained from citronella grass public estate in Simawang, Tanah Datar regency, West Sumatera, Indonesia. Micorrhiza application was conducted by sowing in planting hole. The holes the were covered by soil. A plant for a hole. The manure was combined with urea, TSP and KCl fertilizer. They were applied 2/3 of recommendation dose. The manure was applied a week before planting and an-organic fertilizer was applied four weeks after planting.

The harvesting was conducted in 16 weeks after planting. The harvesting was conducted by cutting leaves 5 cm under leaves midrib by using scissor. The harvesting was conducted at 6 am to 10 am.

2.2 Parameter and data analysis

The parameters were plant height, number of leaves, number of tiller, net assimilation rate and relative growth rate. The data was analyzed by using Analysis of variance (ANNOVA) and continued by using Duncan's New Multiple Range Test (DNMRT) in 5%.

3. RESULTS AND DISCUSSION

3.1 Plant height

Single dose of AMF treatment affected plant height. Otherwise, type of AMF did not affect the plant height (Table 1).

Table 1. Plant height of citronella grass in several type and dose of AMF treatments in 12 weeks after planting

Type of AMF	Dose of AMF (g/plant)				Average
	5	10	15	20	
	…………(g)………				
Glomus sp.	40.10	46.76	72.83	92.60	63.07
Gigaspora sp.	45.60	47.93	77.00	92.43	65.74
Glomus sp.+*Gigaspora* sp.	42.50	50.60	85.46	102.16	70.18
Average	42.73C	48.43C	78.43B	95.73A	

Note : Numbers were followed by lowercase was significantly different according DNMRT test in 5%

Dose 20 g/ plant was the best treatment for plant height. Plant height was affected by nutrients adequacy that required by plant for growth process. AMF application could repair plant nutrients so that it increased plant growth. The nutrient absorption that required by plant run effectively so that plant growth metabolism was effective particularly in vegetative stage. It represented by better plant height. This condition occurred due to AMF infected host plant root system by forming hypha braid intensively so that plant could increase water and nutrients absorption. mycorrhizal could absorb phosphorus from phosphorus mineral sources that was hard to dissolve due to producing organic acids and phosphatase enzyme. mycorrhizal also could increase phosphorus absorption by external hypae presence [13].

AMF performed mutualism symbiosis with host to increase host plant growth could not be separated from AMF ability to produce growth regulating hormone. AMF producing growth regulating hormone such as auxin, cytokinin, gibberellins for its host [14].

3.2 Number of leaves

There was no interaction between type and dose of AMF to number of leaves. Single treatment dose of AMF affected the addition of number of leaves. Otherwise, type of AMF did not affect number of leaves (Table 2). Dose 30 g/ plant showed the best result for number of leaves (22.6). The increasing of number of leaves was seen from dose 5 g/plant to 15 g/plant and reached the highest result in dose 20 g/plant. This result was affected by optimal nutrient availability for plant growth. Dose 20 g/ plant stimulated vegetative growth particularly in former mining area land by absorbing nutrient in this soil. AMF increased plant growth in poor nutrient soil was more effective than fertile soil and showed the positive impact than fertile soil. Generally, association between mycorrhizal and plant in poor nutrient soil was better than in fertile soil [15].

Table 2. Number of leaves of citronella grass in several type and dose of AMF treatments in 12 weeks after planting

Type of AMF	Dose of AMF (g/ plant)				Average
	5	10	15	20	
	……..…(helai)….......				
Glomus sp.	7.00	10.67	15.34	16.00	12.25
Gigaspora sp.	9.00	12.67	16.34	23.34	15.33
Glomus sp.+*Gigaspora* sp.	9.34	12.67	18.67	28.34	17.26
Average	8.45C	12.01CB	16.78B	22.56A	

Note : Numbers were followed by lowercase was significantly different according DNMRT test in 5%

mycorrhizal could support maintaining plant growth stability in polluted condition. In particular, micorrhiza played important role in increasing ion absorption with low mobility level such as phosphate (PO4 3-), ammonium, and immobile nutrient such as sulfur (S), copper (Cu), zinc (Zn) and boron (B) [16]. The mechanism in nutrient absorption increasing could be explained as result of thick hyphal sheath formation so that it increased root adsorb layer to nutrient in soil. Root adsorb layer increasing caused energy availability and it increased root metabolism activity. This condition was caused by oxygen consumption two times so the root could expand mineral salt absorption by expanding ion supply exchange. Nutrient supply increasing from root that associated with AMF would increase photosynthate result that would be translocated to part of plant that required it during vegetative stage [17].

3.3 Number of tiller

In this parameter there was no interaction between type and dose of AMF to number of tiller. But, in single treatment, both of them affected the number of tiller (Table 3)(Figure 1). Type of AMF multispores showed the best result (34.04). Dose AMF 20 g/ plant showed the best result that other doses. Tiller was stem that appeared from main stem. Number of tiller was important parameter in essential oil production due to it affected oil content weight. Number of tiller affected fresh weight so it increased yield component, percentage of essential oil of citronella grass [18].

AMF could increase growth and number of tiller addition of citronella grass in marginal land (coal former mining area) [19]. Outer AMF structure, there was external hypha. For mycorrhizal, external hypha played role for supporting reproduction and transportation of carbon and other nutrients into spores, beside its function as nutrient absorption from soil for plant. External hypha growth occurred if internal hypha grew from cortex through epidermis. External hypha growth occurred until there was no possibility to grow anymore. AMF external hypha increased contact surface area with soil so it increased root absorption area up to 47 times to facilitated to do access to nutrients in soil [20].

Table 3. Number of tiller of citronella grass in several type and dose of AMF treatments in 12 weeks after planting

Type of AMF	Dose AMF (g/plant)				Average
	5	10	15	20	
	….......(stem) ……..				
Glomus sp.	1.67	4.00	6.34	9.67	22.01 b
Gigaspora sp.	2.00	5.00	7.67	11.67	26.34 ab
Glomus sp+*Gigaspora* sp	3.00	5.67	10.34	14.34	34.04 a
Average	9.01C	16.67BC	22.67B	34.04A	

Note : Numbers were followed by lowercase was significantly different according DNMRT test in 5%

According Table 3, type of AMF multispores produced best number of tiller than single type of AMF. This result indicated that AMF multispores was more effective to produce tiller and also effective in plant vegetative growth.Wide spread of *Glomus* sp. spore and high adaptation to environment, if combined to ability of *Gigaspora* sp. in symbiosis with host plant and optimal nutrient absorption affected vegetative and generative growth of host plant [21]. Previous research reported that multispores showed higher root infection ability than single spore. Dose 20 g/plant AMF in multispores could increase maize growth in former coal mining area land [22].

Figure 1. Number of tiller in several treatments of type and dose of AMF in 12 weeks after planting (a. *Glomus* sp., dose 5 g/plant; b. a. *Glomus* sp., dose 10 g/plant; c. a. *Glomus* sp., dose 15 g/plant; d. *Glomus* sp., dose 20 g/plant)

3.4 Net assimilation rate (NAR)

There was interaction between type and dose of AMF to net assimilation rate of citronella grass in this study. The single treatment also affected this parameter (Table 4). Net assimilation rate was plant ability to produce dry material from assimilation per leave area unit and time that determined by water and nutrients availability. The result showed that dose 15 g/ plant could produce better NAR. It indicated that AMF was optimal in increasing water and nutrient absorption capacity for plant.

Table 4. Net assimilation rate (NAR) of citronella grass in several type and dose of AMF treatments in 14 weeks after planting

Type of AMF	Dose of AMF (g/plant)			
	5	10	15	20
	…..................(batang)……... ……...…			
Glomus sp.	0.000504ab B	0.000524b B	0.000559b AB	0.000654b A
Gigaspora sp.	0.000529a A	0.000509b A	0.000492b A	0.000558b A
Glomus sp. +*Gigaspora* sp.	0.000416b C	0.000648a B	0.000734a AB	0.000777a A

Note : Numbers were followed by lowercase was significantly different according DNMRT test in 5%

Type of AMF also affected NAR increasing. Generally, AMF multispores could increase NAR significantly than other single treatment except dose 5 g/plant. AMF multispores dose 20 g/plant could increase NAR. It was caused by AMF multispores could increase vegetative and generative growth of citronella grass optimally. It could be seen in plant height, number of leaves and number of tiller.

AMF multispores also increased water and nutrient absorption effectively so more photosynthesis material was obtained. More photosynthesis material was obtained, more assimilate was translocated to part of plant that required it during vegetative growth for tiller cells formation, leaves and root. This affected plant dry weight. Previous research reported that AMF affected plant height, number of leaves and fresh weight of sorghum [23].

3.5 Relative growth rate (RGT)

In this parameter, the interaction between type and dose of AMF affected relative growth rate (RGT). Each treatment showed the similar result (Table 5). According the result, in type of AMF *Glomus* sp., RGT increasing was significantly seen in dose 5 g/plant up to 15 g/plant. But, in 20 g/plant, the increasing was not significant. The similar result occurred in AMF type multispores. In AMF *Gigaspora* sp., RGT increasing was significant in dose 10 g/plant, but if the dose was increased up to 20 g/plant, RGT did not increase anymore.

Table 5. Relative growth rate (RGT) of citronella grass in several type and dose of AMF treatments in 14 weeks after planting

Type of AMF	Dose of AMF (g/plant)			
	5	10	15	20
	…..................(Stem)……………..….			
Glomus sp.	0.0069b C	0.0109b B	0.0132a A	0.0144a A
Gigaspora sp	0.0108a B	0.0132a A	0.0141a A	0.0141a A
Glomus sp+*Gigaspora* sp	0.0124a A	0.0128ab A	0.0132a A	0.0140a A

Note : Numbers were followed by lowercase was significantly different according DNMRT test in 5%

RGT was material increasing per unit time. Generally, RGT was plant dry weight addition in certain interval so RGT increasing could not separated from dry weight increasing. AMF application positively affected dry weight including root dry weight and crown dry weight. AMF had ability to optimize water and nutrient absorption from soil so it increased plant dry weight. Higher dry weight of plant caused RGT increasing [24].

AFM had ability to provide nutrients that could not be absorbed by plant to be absorbed nutrient by plant. mycorrhizal had ability to release soil phosphorus from undissolved form to dissolved form so available phosphorus increased. mycorrhizal could dissolve phosphorus by producing phosphatase enzyme and Fe and Al chelating compound. Increasing of phosphate was continued by nitrogen, zinc, copper and sulphur [13].

The increasing of dry product was affected by absorbed photosynthesis material by AMF helping by stimulating tertiary root growth. The expanding of mycorrhizal external hypha affected tertiary roots of host plant. More and more tertiary roots caused the absorption of water and nutrients was more maximum. This condition increased photosynthesis process of plant [25].

4. CONCLUSION

There was interaction between type and dose of AMF to growth and yield pf citronella grass in former coal mining area land. The best type of AMF was AMF multispores (*Glomus* sp. + *Gigaspora* sp.). This combination gave the best result for growth and and yield of citronella grass that planted in former coal mining area land soil. Type of AMF multispores in 20 g/ plant could number of tiller. Dose 20 g/plant could plant height and number of leaves. AMF multispores only increased number of tiller and net assimilation rate.

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