SOIL FERTILITY STATUS AND SOYBEAN [*Glycine max* (L) Merr] PERFORMANCE FOLLOWING INTRODUCTION OF INDIGENOUS MYCORRHIZA COMBINED WITH VARIOUS NUTRIENT SOURCES INTO SANDY SOIL

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ABSTRACT

A series of experiment aimed at obtaining soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil was conducted in a glasshouse. The experiment tested four treatments, namely; inoculation of soil with mycorrhiza, inoculation of soil with mycorrhiza and cattle manure, inoculation of soil with mycorrhiza and rock phosphate, and inoculation of soil with mycorrhiza and inorganic fertilizers. Soil without any inoculation was provided as control. The treatments were arranged in a Completely Randomized Design with four replications. Soil fertility status was based on the concentrations of N, P, K, and organic-C as well as soil pH. Plant performance was determined based on its ability to uptake nutrients (N, P, K, and Ca), its growth and yield. Then, the activity of mycorrhiza was measured based on total population and percentage of root infection. Overall, results of the present study showed that, compared with other treatments, inoculation of mycorrhiza into soil and amended with cattle manure significantly performed higher concentrations of N, P, K, and organic-C. This soil condition caused soybean to absorb significantly higher nutrients, grew well with higher yield compared with plant performance in other treatments. Therefore, results of this study implies that the application of mycorrhiza into soil amended with organic matter is promising to sustain soil productivity under soybean cropping system.

Key words: Arbuscular Mycorrhiza , soil fertility, soybean yield, dryland

INTRODUCTION

Shortage in availability of water, nutrients, and organic matter was a core problems in improving plant production in sandy soil of northern Lombok (Suwardji *et al*., 2007). Sandy soil with low organic matter content has low capacity in holding water and nutrients to support optimal plant growth (Suzuki and Noble, 2007; Bastida *et al*., 2010). This character of sandy soil is considered as a specific problem in managing sandy soil in northern Lombok, especially to grow soybean. To overcome this particular problem, a strategy in managing soil, in long term, to gain improvement and stability of soil organic matters which then lead to the improvement of soil characteristics especially in holding capacity of water and nutrients in the state of ready to be used by plants (Parman *et al*., 2000). In addition, soil management by improving role of indigenuous arbuscular mycorrhiza (AM) in sandy soil is one of best possible alternative solutions to improve plant productivity (Herrera *et al*., 1993 and Astiko, 2009).

Optimizing role and beneficial characteristic of AM through application of biofertilizer to improve plant productivity in dry land is prospective enough (Sastrahidayat *et al.*, 2001 dan Astiko *et al*., 2005). Application of AM by introducing the organisms into soil is expected to be able to improve plant productivity significantly through role of AM in improving plant capacity to absorb N, P, K, Ca and other micro nutrients. Besides, with its external hypha, AM will improve plant resistance on drought, protect plant root from soil-born pathogen infection, stimulate activity of beneficial microorganisms, and improve soil texture and structure (Gianinazzi and Vosátka, 2004; Feldmann *et al*., 2009; Ijdo *et al*., 2011).

Results of many previous studies have proven that nutrient absorption, growth and result of plant inoculated by AM were much higher than control (Azcón-Aguilar and Barea,

1997; Nogueira *et al.*, 2007; Fisher and Jayachandran, 2008). Furtheermore, it was reported that the use of indigenous AM had advantages as the microorganism was able to establish and develop well in situ environment as well as its ability to compete with existing soil microbes compared with introduced AM (Turrini *et al.*, 2008).

Incorporation of AM fungus on soybean in sandy soil of dry-land Northern Lombok was expected to have positive impact on soil characteristic improvement, nutrient absorption, and finally plant growth and yield. This hypothesis was constructed based on results of previous research in Vertisol soil (Astiko *et al*., 2005) proving that inoculation of AM on soybean improved absorption of P and plant yield was higher tan that of plant without AM. The improvement of P absorption was due to AM activity in improving nutrient availability and root proliferation (Smith *et al.*, 2010). This increase of plant yield was suggested due to the ability of AM to increase efficiency in wáter use, nutrient absorption, and to maintain turgor of plant cells. The external hypha of AM fungus were expected to be able to absorp soil pore wáter when plant roots anable to do so. In addition, wide distribution of external hypha caused the amount of wáter taken improved on the soil with low wáter content (Drew *et al*., 2003; Smith and Read, 2008). Base don the above mentioned, the aim of this study was to assess soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil.

METHODS

Experiment Preparation and Maintainance

A series of pot experiments using sandy soil taken Northern Lombok was conducted in a glasshouse of Faculty of Agriculture, Mataram University. The experiment was conducted

in two cropping cycles, namely; the first cropping cycle in which soybean was grown and fertilized according to treatments, and the second cropping cycle in which soybean was grown on soil that had been used for the first cropping cycle without fertilization. The study tested four treatments, namely; soil was inoculated with AM (F1), soil was inoculated with AM and amended with cattle manure (F2), soil was inoculated with AM and amended with rock phosphate (F3), and soil was inoculated with AM and amended with inorganic fertilizers (F4). Soil without AM inoculation and amendment was provided as control (F0). The treatments were completely randomized designed with four replications. The AM inoculums was prepared by mixing spores, powder medium, and root residues. The AM inoculum (20 gram per pot) then was inoculated at sowing time by layering the inoculum at 10 cm depth. Soybean seeds of Kaba var. were sown (2 seeds per pot) at 3 cm depth and 14 days after sowing (das), only one seedling per pot was left to grow further. Nutrient sources applied according to treatments were cattle manure, rock phosphate, Urea, and SP36. Rock phosphate and cattle manure were applied at rate of 1.2 and 2.0 g per plant respectively, and inorganic fertilizers of Urea and SP-36 were applied at rate of 0.1 g and 0.2 g per plant, consecutively. The nutrient sources were buried 5 cm around the plant at depth of 7 cm. The plants were harvested 100 das.

Parameters Observation

Concentrations of N, P, K, and organic-C in soil, and soil pH were measured before sowing, 60 das and 100 das. Plant biomass (root and shoot dry weight) were measured at 60 das and 100 das after being oven dried at 60 ^{o}C for 48 hours. Nutrient uptake (N, P, K, and Ca) was measured at 60 das. Plant yield components recorded were weights of cobs, total

grains, and 100 grains at 100 das. Population of AM and percentage of root infection were measured at 60 das.

Soil, Plant Analysis and Mycorrhiza Observation

While analyses for N, P, and organic-C were done by using Kjeldhal method, spectrophotometer, and colorimetric method according to Walkley and Black, respectively, K and Ca were analyzed by using AAS. Mycorrhiza population was observed using wet sieving technique according to Brundrett *et al*. (1996). The supernatan caught at 38 µm-sieve was added with 60% of sucrosa solution and subsequently centrifused at 3000 rpm for 10 minute (Daniel and Skipper, 1982). The harvested spore were stored on the Whatman paper with permanent ink marked of 0,5 x 0,5 cm. Counting of mycoriza population was done using stereo mikroscope (extended 40 x). Calculation of roots percentage infections was conducted using modification of clearing and staining method (Kormanik and McGraw, 1982), counted with the *Gridline Intersect* technique (Giovenneti and Mosse, 1980) under stereo-microcope observation.

Statistical analysis

Data were statistically analyzed for analysis of variance (ANOVA) and then continued using least significance different (LSD) at $p = 0.05$.

RESULTS AND DISCUSSION

Soil chemical properties

In general, compared with control, all treatments significantly increased concentration of total N, available P and K, and organic-C both at 60 das and 100 das (Table 1). The highest

increases of nutrient concentrations were observed in sandy soil inoculated with arbuscular mycorrhiza (AM) and amended with cattle manure (CM). This indicated that AM performed well when combined with CM compared with other combinations. Compared with control, inoculation of AM amended with CM increased N, P, K, and org-C as much as 39%, 105%, 27%, and 85%, respectively on 60 das and 12%, 60%, 10%, and 11%, consecutively on 100 das at the first cropping cycle. Interestingly, these effects were also observed at the second cropping cycle as the total N, P, K, and org-C increased as much as 43%, 120%, 37%, and 36%, respectively on 60 das, and 44%, 53%, 14%, and 36%, respectively on 100 das. Data presented in Table 1 also indicated that at the second cropping cycle the treatment of AM plus CM could improve nutrient concentration of P and N as much as 4 and 48% on 60 das, respectively, and on 100 das the treatment increased N and Org-C as much as 12 and 20%, consecutively.

Results of this study were in accordance with those of done by Jeffries *et al*. (2003) and Gianinazzi *et al*. (2010) reporting that AM inoculation with organic matter amendment can recover soil fertility on an ecosystem. Furthermore, Douds *et al*. (2006) reported that AM inoculation could increase soil nutrient content. This increase was suggested due to activity of enzyme present in external hypha of AM in the rhizosphere able to catalize and hydrolyze unavailable nutrient complex into available (Widiastuti *et al.*, 2003).

Table 1. Soil chemical properties (N, P, K, organic-C and soil pH) of sandy soil with various treatments after harvesting.

Treatments	N ($g\ kg^{-1}$)		P ($mg\ kg^{-1}$)		K ($cmol\ kg^{-1}$)		org-C ($g\ kg^{-1}$)		pH	
	1st	2ad	1st	2ad	1st	2ad	1st	2ad	1st	2ad
60 DAS										

F_0	1.15^{a}	0.87^{a}	16.97^{a}	16.54^{a}	0.69^{a}	0.51^{a}	12.1^{a}	24.5^{a}	6.25^{a}	6.13^{a}
F_1	1.34^{b}	1.15^{b}	23.60^{b}	23.40^{b}	0.75^{b}	0.58^{b}	17.9^{b}	26.5^{b}	6.01^{b}	6.24^{b}
F_2	1.60^{c}	1.25^{b}	34.83^{c}	36.54^{c}	0.88^{c}	0.70^{c}	22.5^{c}	33.5^{c}	5.95^{c}	6.32^{c}
F_3	1.44^{d}	1.10^{b}	26.34^{d}	27.01^{d}	0.83^{d}	0.63^{d}	21.0^{d}	29.4^{d}	6.72^{d}	6.28^{d}
F_4	1.40^{e}	0.93^{b}	20.59^{d}	30.97^{e}	0.78^{c}	0.62^{bd}	19.1^{e}	26.3^{b}	6.08^{e}	6.26^{bd}
100 DAS										
F_0	1.31^{a}	1.14^{a}	17.62^{a}	17.33^{a}	0.75^{a}	0.63^{a}	23.8^{a}	23.5^{a}	6.18^{a}	6.54^{a}
F_1	1.44^{b}	1.36^{b}	20.86^{b}	22.68^{bc}	0.77^{b}	0.65^{b}	24.5^{b}	25.3^{ab}	6.21^{b}	6.62^{ab}
F_2	1.47^{c}	1.65^{c}	28.25^{c}	26.57^{d}	0.83^{c}	0.72^{c}	26.6^{c}	32.1^{c}	6.24^{c}	6.54^{ab}
F_3	1.38^{d}	1.41^{b}	23.32^{d}	20.62^{c}	0.81^{d}	0.67^{b}	23.9^{d}	28.0^{d}	6.62^{d}	6.75^{a}
F_4	1.33^{a}	1.43^{b}	24.38^{e}	24.57^{bd}	0.82^{e}	0.70^{c}	25.7^{e}	27.5^{d}	6.23^{c}	6.44^{b}
Before exp$^{1)}$	1.1	-	13.82	-	0.57	-	12.1	-	6.25	-

Note: Means followed by the same letters within the same column are not significantly different ($p=0.05$); 1^{st} and 2^{ad} first cycles and second cyles; F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; $^{1)}$ pre-treatment data

Previous results reported by Warnock *et al.*, (2007) showed that enrichment of AM could be escalated by the addition of organic matter and the combination has a positive effect on improving soil physico-chemical properties and therefore it is beneficial for soil microorganisms activities. This signify that better sinergy of the inoculated AM-tretment in combination with CM, as previously confirmerd by Kato and Miura (2008). Higher soil extractable-P found in this study was not only due to indirect contribution of CM in improving soil fertility status but also related to a positive contribution of mycorrhiza in producing phosphatase enzyme, for mineralization of organic-P in soil (Crowley and Rengel, 2000; Joner and Johansen, 2000) and resulted in enhancing insoluble-P in soils (Orcut and Nilsen, 2000).

Nutrient uptake, growth and yield

Nutrient absorption by soybean is presented in Table 2. In general, treatments such AM inoculation alone or combined with other sources of nutrients significantly increased nutrient aupatke by soybean, compared with control. The highest increases were observed at treatment

AM inoculation amended with CM. At this treatment, absorption of N, P, K, and Ca increased as much as 214%, 185%, 342%, and 233%, respectively on the first cropping cycle, and 191%, 308%, 224%, and 413%, consecutively on the second cropping series, compared with control. Treatment of AM inoculation followed by CM amendment also caused high absorption of P and Ca as much as 29% and 126%, respectively on the second cropping cycle.

The increase of P absorption could possibly cause such a new nutrient balance in plant that induced absorption of other nutrients such as N, K, and Ca. Sufficient availability of K created a condition in which the use of water was efficient as cell turgor was maintained. This condition leads to active metabolism process, K accumulation on the tips, buds, and roots, the accumulation that lead to formation of cortex tissue and cell elongation (Schweiger *et al.*, 2007 dan Smith *et al*,, 2010).

Table 2. Nutrient uptake (N,P, K and Ca) by soybean grown grown on sandy soil with various treatments.

Treatments	Nutrient uptake (mg/plant)							
	First cycle				Second cycle			
	N	P	K	Ca	N	P	K	Ca
60 DAS								
F_0	221.59[a]	22.43[a]	121.53[a]	25,60[a]	224.52[a]	20.37[a]	79.21[a]	37.66[a]
F_1	302.83[b]	29.03[b]	210.13[b]	34,16[b]	301.13[b]	34.46[b]	121.63[ab]	59.22[b]
F_2	697.16[c]	64.06[c]	537.33[c]	85,30[c]	653.42[c]	83.22[c]	257.43[c]	193.30[c]
F_3	412.73[d]	39.55[d]	282.73[d]	48,20[d]	429.60[d]	49.33[d]	185.20[d]	12.96[d]
F_4	344.10[e]	34.46[e]	232.46[e]	39,96[d]	311.60[b]	38.14[b]	159.44[ad]	96.93[d]

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF

The inoculation of AM followed by CM also increased plant dry biomass measured by root and shoot dry weights on 60 and 100 das (Table 3). Dry weight of roots and shoots on 60 das increased as much as 164% and 136%, respectively; while on 100 das the increases were as much as 150% and 178%, respectively on the first cropping cycle. On the second cycle, on 60 das the increases were 337% and 718%, while on 100 das the increases were as much as 390% and 1102%, consecutively.

Table 3. Root and shoot dry weight of soybean grown in sandy soil with various treatments. at 60 and 100 DAS

Treatments	Biomass dry weight (mg/plant) First cycle		Second cycle	
	Roots	Shoots	Roots	Shoots
60 DAS				
F_0	1.88[a]	11.11[a]	1.74[a]	8.50[a]
F_1	2.66[b]	14.51[b]	2.27[b]	10.48[b]
F_2	4.97[c]	26.28[c]	4.38[c]	18.67[c]
F_3	3.62[d]	20.39[d]	3.75[d]	13.51[d]
F_4	3.34[d]	17.10[e]	2.51[b]	11.41[e]
100 DAS				
F_0	3.41[a]	22.43[a]	2.83[a]	19.94[a]
F_1	4.45[b]	39.63[b]	2.54[a]	19.35[a]
F_2	8.53[c]	62.45[c]	6.27[b]	32.92[b]
F_3	5.22[d]	44.50[d]	3.41[a]	26.40[c]
F_4	4.48[b]	37.85[b]	2.36[a]	19.91[c]

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF.

The improvement of biomass was probably due to AM role to influence nutrient availability, especially P. Sufficient availability of P can indirectly induce absorption of other nutrients leading to better plant growth (Carrenho *et al*., 2001). This positive effect was suggested due to suitability of AM type, plant, and soluble P (Nikolaou *et al*., 2002; Bhadalung *et al*., 2005). Table 3 also showed significant effect of AM inoculation combined with CM as indicated by the increase of root and shoot dry weight. The similar results were also reported previously (Rossiana and Supriatun, 2003; Simarmata and Herdiani, 2003; Bertham, 2006).

Inoculation of AM with CM amendment increased cobs, weight of grain, and weight of 100 grains as much as 180%, 163%, and 139% respectively on the first cropping cycle and as much as 139%, 330%, and 23%, consecutively on the second cycle. These results were in accordance with results of previous studies indicating that fertilizer package with AM inoculation and amended with CM increased nutrient absorption, plant growth and yield (Astiko, 2009; Viti *et al.*, 2010). Similar results were also reported on dry paddy (Kabirun, 2002).

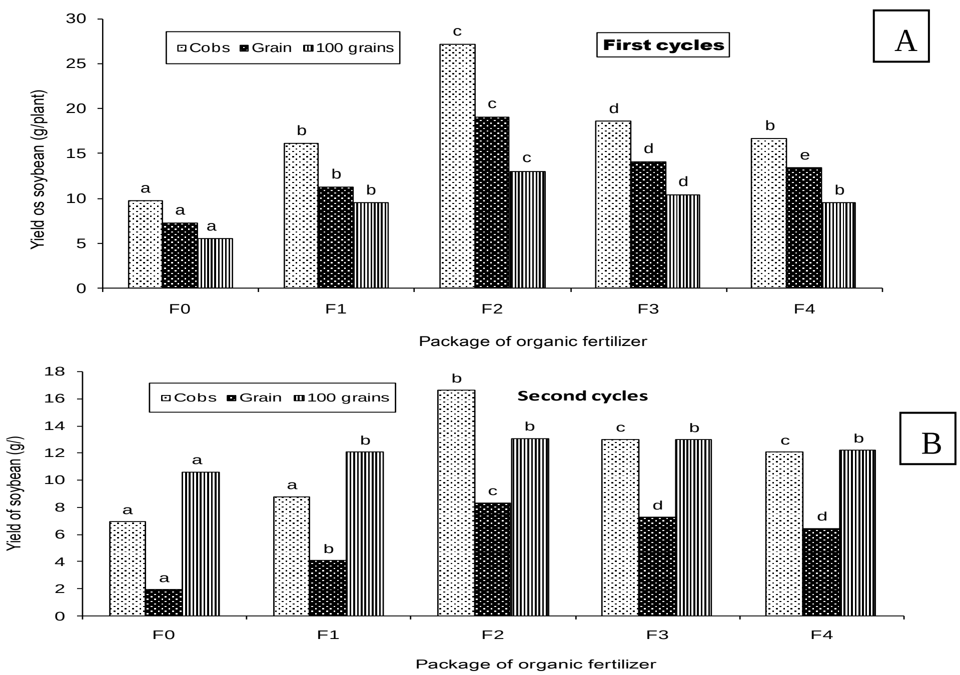

Fig 1. Yields of soybean (cobs, grain and 100 grain dry weight) grown in sandy soil with various treatments. A. the first cropping cycle, and B. the second cropping cycle. Bars with the same letters at the same category are not significantly different (p=0.05).

The increases of nutrient absorption, plant growth and yield on the tretmant of AM inoculation amended with CM were caused by the increase of AM activity in absorption of nutrient and water through its external hypha (EH). This was possibly due to EH can reach depletion zone that cannot be reached by plant roots (Zhu *et al.*, 2001). The diameter of EH which is much smaller than that of roots makes the EH possibly to penetrate soil micro pores to take nutrient and water that cannor be reached by roots

(Drew *et al*., 2003). This ability causes plants with mycorrhiza be able to absorb nutrient, growth and perform better and resistance to dought stress (Smith and Read, 2008). All above facts indicated such a suitable functional among AM, host plants, and environment that are able to increase nutrient absorbtion, plant growth and yield as also reported earlier (Burleight *et al*., 2002).

Mycorrhiza activity

The inoculation of AM followed by CM amendment could increase AM activity as shown by numbers of spores per 100 g of soil and percentage of root infection (Tabel 4). The number of spores and percentage of infection on the first cropping cycle 60 das on sandy soil inoculated by AM and amended with CM increased as much as 179% and 266%, respectively, while on the second cycle the increases were 24% and 160%, respectively compared with control and were significantly different from other treatments.

Table 4. Biological activity of mycorrhiza (number of spores and percentage of infections) in sandy soil with various treatments.

Treatments	Spores 100 g $soil^{-1}$ and root infection			
	First cycle		Second cycle	
	Spores	Infection	Spores	Infection
60 DAS				
F_0	1,060^a	21^a	3,122^a	25^a
F_1	2,159^b	41^b	3,533^b	34^b

F_2	2,960^{c}	77^{c}	3,878^{c}	65^{c}
F_3	2,343^{d}	54^{d}	3,781^{d}	51^{d}
F_4	2,215^{b}	46^{e}	3,693^{e}	41^{e}
Before exp[1)]	371	-	-	-

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; [1)] pre-treatment data

Table 4 showed that the number of spores and root infection were significantly high in all treatments when compared with control. Compared with before sowing, the number of spores and root infection 60 das increased significantly. This increases indicated that isolate M_{AA1} were able to compete with indigenuous AM present in the rhizosphere of sandy soil Northern Lombok, particularly in colony forming inside roots. The other point that can be taken from these facts was that the isolate M_{AA1} used was able to produce abundance propagules in the form of spores and such colonized roots that they were able to live on dynamic and competitive habitat (Barrios, 2007; Doud and Johnson, 2007; Gianinazzi *et al*., 2010). This fact also indicated that isolate M_{AA1} showed high effectiveness, although they were inoculated on unsterile soil. Similar research on soybean inoculated by AM and the application of organic leaf fertilizer “greenstant” also showed similar results (Wangiyana *et al.*, 2007).

CONCLUSION

Inoculation of AM amended with cattle manure improved sandy soil fertility as shown by the increasing concentrations of N, P, K, and organic-C. Soybean responded positively to the application of AM followed by cattle manure as indicated by plant improvement in nutrient absorption, plant growth and yield. The addition of cattle manure stimulated activity of AM leading to improvement of soil fertility and plant performance.

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Dikirim: Rabu, 2 Januari 2013 16:58
Judul: [AJAS] Submission Acknowledgement

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SOIL FERTILITY STATUS AND SOYBEAN [*Glycine max* (L) Merr] PERFORMANCE FOLLOWING INTRODUCTION OF INDIGENOUS MYCORRHIZA COMBINED WITH VARIOUS NUTRIENT SOURCES INTO SANDY SOIL

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ABSTRACT

A series of experiment aimed at obtaining soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil was conducted in a glasshouse. The experiment tested four treatments, namely; inoculation of soil with mycorrhiza, inoculation of soil with mycorrhiza and cattle manure, inoculation of soil with mycorrhiza and rock phosphate, and inoculation of soil with mycorrhiza and inorganic fertilizers. Soil without any inoculation was provided as control. The treatments were arranged in a Completely Randomized Design with four replications. Soil fertility status was based on the concentrations of N, P, K, and organic-C as well as soil pH. Plant performance was determined based on its ability to uptake nutrients (N, P, K, and Ca), its growth and yield. Then, the activity of mycorrhiza was measured based on total population and percentage of root infection. Overall, results of the present study showed that, compared with other treatments, inoculation of mycorrhiza into soil and amended with cattle manure significantly performed higher concentrations of N, P, K, and organic-C. This soil condition caused soybean to absorb significantly higher nutrients, grew well with higher yield compared with plant performance in other treatments. Therefore, results of this study implies that the application of mycorrhiza into soil amended with organic matter is promising to sustain soil productivity under soybean cropping system.

Key words: Arbuscular Mycorrhiza , soil fertility, soybean yield, dryland

INTRODUCTION

Shortage in availability of water, nutrients, and organic matter was a core problems in improving plant production in sandy soil of northern Lombok (Suwardji *et al*., 2007). Sandy soil with low organic matter content has low capacity in holding water and nutrients to support optimal plant growth (Suzuki and Noble, 2007; Bastida *et al*., 2010). This character of sandy soil is considered as a specific problem in managing sandy soil in northern Lombok, especially to grow soybean. To overcome this particular problem, a strategy in managing soil, in long term, to gain improvement and stability of soil organic matters which then lead to the improvement of soil characteristics especially in holding capacity of water and nutrients in the state of ready to be used by plants (Parman *et al*., 2000). In addition, soil management by improving role of indigenuous arbuscular mycorrhiza (AM) in sandy soil is one of best possible alternative solutions to improve plant productivity (Herrera *et al*., 1993 and Astiko, 2009).

Optimizing role and beneficial characteristic of AM through application of biofertilizer to improve plant productivity in dry land is prospective enough (Sastrahidayat *et al*., 2001 dan Astiko *et al*., 2005). Application of AM by introducing the organisms into soil is expected to be able to improve plant productivity significantly through role of AM in improving plant capacity to absorb N, P, K, Ca and other micro nutrients. Besides, with its external hypha, AM will improve plant resistance on drought, protect plant root from soil-born pathogen infection, stimulate activity of beneficial microorganisms, and improve soil texture and structure (Gianinazzi and Vosátka, 2004; Feldmann *et al*., 2009; Ijdo *et al*., 2011).

Results of many previous studies have proven that nutrient absorption, growth and result of plant inoculated by AM were much higher than control (Azcón-Aguilar and Barea,

1997; Nogueira *et al.*, 2007; Fisher and Jayachandran, 2008). Furtheermore, it was reported that the use of indigenous AM had advantages as the microorganism was able to establish and develop well in situ environment as well as its ability to compete with existing soil microbes compared with introduced AM (Turrini *et al.*, 2008).

Incorporation of AM fungus on soybean in sandy soil of dry-land Northern Lombok was expected to have positive impact on soil characteristic improvement, nutrient absorption, and finally plant growth and yield. This hypothesis was constructed based on results of previous research in Vertisol soil (Astiko *et al*., 2005) proving that inoculation of AM on soybean improved absorption of P and plant yield was higher tan that of plant without AM. The improvement of P absorption was due to AM activity in improving nutrient availability and root proliferation (Smith *et al.*, 2010). This increase of plant yield was suggested due to the ability of AM to increase efficiency in wáter use, nutrient absorption, and to maintain turgor of plant cells. The external hypha of AM fungus were expected to be able to absorp soil pore wáter when plant roots anable to do so. In addition, wide distribution of external hypha caused the amount of wáter taken improved on the soil with low wáter content (Drew *et al*., 2003; Smith and Read, 2008). Base don the above mentioned, the aim of this study was to assess soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil.

METHODS

Experiment Preparation and Maintainance

A series of pot experiments using sandy soil taken Northern Lombok was conducted in a glasshouse of Faculty of Agriculture, Mataram University. The experiment was conducted

in two cropping cycles, namely; the first cropping cycle in which soybean was grown and fertilized according to treatments, and the second cropping cycle in which soybean was grown on soil that had been used for the first cropping cycle without fertilization. The study tested four treatments, namely; soil was inoculated with AM (F1), soil was inoculated with AM and amended with cattle manure (F2), soil was inoculated with AM and amended with rock phosphate (F3), and soil was inoculated with AM and amended with inorganic fertilizers (F4). Soil without AM inoculation and amendment was provided as control (F0). The treatments were completely randomized designed with four replications. The AM inoculums was prepared by mixing spores, powder medium, and root residues. The AM inoculum (20 gram per pot) then was inoculated at sowing time by layering the inoculum at 10 cm depth. Soybean seeds of Kaba var. were sown (2 seeds per pot) at 3 cm depth and 14 days after sowing (das), only one seedling per pot was left to grow further. Nutrient sources applied according to treatments were cattle manure, rock phosphate, Urea, and SP36. Rock phosphate and cattle manure were applied at rate of 1.2 and 2.0 g per plant respectively, and inorganic fertilizers of Urea and SP-36 were applied at rate of 0.1 g and 0.2 g per plant, consecutively. The nutrient sources were buried 5 cm around the plant at depth of 7 cm. The plants were harvested 100 das.

Parameters Observation

Concentrations of N, P, K, and organic-C in soil, and soil pH were measured before sowing, 60 das and 100 das. Plant biomass (root and shoot dry weight) were measured at 60 das and 100 das after being oven dried at 60 ^{o}C for 48 hours. Nutrient uptake (N, P, K, and Ca) was measured at 60 das. Plant yield components recorded were weights of cobs, total

grains, and 100 grains at 100 das. Population of AM and percentage of root infection were measured at 60 das.

Soil, Plant Analysis and Mycorrhiza Observation

While analyses for N, P, and organic-C were done by using Kjeldhal method, spectrophotometer, and colorimetric method according to Walkley and Black, respectively, K and Ca were analyzed by using AAS. Mycorrhiza population was observed using wet sieving technique according to Brundrett *et al.* (1996). The supernatan caught at 38 µm-sieve was added with 60% of sucrosa solution and subsequently centrifused at 3000 rpm for 10 minute (Daniel and Skipper, 1982). The harvested spore were stored on the Whatman paper with permanent ink marked of 0,5 x 0,5 cm. Counting of mycoriza population was done using stereo mikroscope (extended 40 x). Calculation of roots percentage infections was conducted using modification of clearing and staining method (Kormanik and McGraw, 1982), counted with the *Gridline Intersect* technique (Giovenneti and Mosse, 1980) under stereo-microcope observation.

Statistical analysis

Data were statistically analyzed for analysis of variance (ANOVA) and then continued using least significance different (LSD) at p = 0.05.

RESULTS AND DISCUSSION

Soil chemical properties

In general, compared with control, all treatments significantly increased concentration of total N, available P and K, and organic-C both at 60 das and 100 das (Table 1). The highest

Commented [T1]: Simply disclosing all your results and methods in
excruciating detail is not enough. Your "value-added benefit" lies in the conclusions you draw from
your results.

Commented [T2]: Based on your title, you stated that there were an informations about soil fertility status. This discussion section could easily be made more informative by placing your results in the context of previously published studies more so than in the current paper. A more complete
comparison of your data to literature reports would help the reader to understand the newness
of the findings and how they extend the work of others. Please do not lost to the main topic.

increases of nutrient concentrations were observed in sandy soil inoculated with arbuscular mycorrhiza (AM) and amended with cattle manure (CM). This indicated that AM performed well when combined with CM compared with other combinations. Compared with control, inoculation of AM amended with CM increased N, P, K, and org-C as much as 39%, 105%, 27%, and 85%, respectively on 60 das and 12%, 60%, 10%, and 11%, consecutively on 100 das at the first cropping cycle. Interestingly, these effects were also observed at the second cropping cycle as the total N, P, K, and org-C increased as much as 43%, 120%, 37%, and 36%, respectively on 60 das, and 44%, 53%, 14%, and 36%, respectively on 100 das. Data presented in Table 1 also indicated that at the second cropping cycle the treatment of AM plus CM could improve nutrient concentration of P and N as much as 4 and 48% on 60 das, respectively, and on 100 das the treatment increased N and Org-C as much as 12 and 20%, consecutively.

Results of this study were in accordance with those of done by Jeffries *et al*. (2003) and Gianinazzi *et al*. (2010) reporting that AM inoculation with organic matter amendment can recover soil fertility on an ecosystem. Furthermore, Douds *et al*. (2006) reported that AM inoculation could increase soil nutrient content. This increase was suggested due to activity of enzyme present in external hypha of AM in the rhizosphere able to catalize and hydrolyze unavailable nutrient complex into available (Widiastuti *et al*., 2003).

Table 1. Soil chemical properties (N, P, K, organic-C and soil pH) of sandy soil with various treatments after harvesting.

Treatments	N (g kg^{-1})		P (mg kg^{-1})		K (cmol kg^{-1})		org-C (g kg^{-1})		pH	
	1st	2^{ad}	1st	2^{ad}	1st	2^{ad}	1st	2^{ad}	1st	2^{ad}
60 DAS										
F_0	1.15^a	0.87^a	16.97^a	16.54^a	0.69^a	0.51^a	12.1^a	24.5^a	6.25^a	6.13^a
F_1	1.34^b	1.15^b	23.60^b	23.40^b	0.75^b	0.58^b	17.9^b	26.5^b	6.01^b	6.24^b
F_2	1.60^c	1.25^b	34.83^c	36.54^c	0.88^c	0.70^c	22.5^c	33.5^c	5.95^c	6.32^c
F_3	1.44^d	1.10^b	26.34^d	27.01^d	0.83^d	0.63^d	21.0^d	29.4^d	6.72^d	6.28^d
F_4	1.40^e	0.93^b	20.59^d	30.97^e	0.78^c	0.62^{bd}	19.1^e	26.3^b	6.08^e	6.26^{bd}
100 DAS										
F_0	1.31^a	1.14^a	17.62^a	17.33^a	0.75^a	0.63^a	23.8^a	23.5^a	6.18^a	6.54^a
F_1	1.44^b	1.36^b	20.86^b	22.68^{bc}	0.77^b	0.65^b	24.5^b	25.3^{ab}	6.21^b	6.62^{ab}
F_2	1.47^c	1.65^c	28.25^c	26.57^d	0.83^c	0.72^c	26.6^c	32.1^c	6.24^c	6.54^{ab}
F_3	1.38^d	1.41^b	23.32^d	20.62^c	0.81^d	0.67^b	23.9^d	28.0^d	6.62^d	6.75^a
F_4	1.33^a	1.43^b	24.38^e	24.57^{bd}	0.82^e	0.70^c	25.7^e	27.5^d	6.23^c	6.44^b
Before exp[1)]	1.1	-	13.82	-	0.57	-	12.1	-	6.25	-

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); 1st and 2^{ad} first cycles and second cyles; F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; [1)] pre-treatment data

Previous results reported by Warnock *et al.*, (2007) showed that enrichment of AM could be escalated by the addition of organic matter and the combination has a positive effect on improving soil physico-chemical properties and therefore it is beneficial for soil microorganisms activities. This signify that better sinergy of the inoculated AM-tretment in combination with CM, as previously confirmerd by Kato and Miura (2008). Higher soil extractable-P found in this study was not only due to indirect contribution of CM in improving soil fertility status but also related to a positive contribution of mycorrhiza in producing phosphatase enzyme, for mineralization of organic-P in soil (Crowley and Rengel, 2000; Joner and Johansen, 2000) and resulted in enhancing insoluble-P in soils (Orcut and Nilsen, 2000).

Nutrient uptake, growth and yield

Commented [T3]: Please focus on the main topic about soybean performance. This discussion section also could easily be made more informative by placing your results in the context of previously published studies more so than in the current paper. A more complete comparison of your data to literature reports would help the reader to understand the newness of the findings and how they extend the work of others

Nutrient absorption by soybean is presented in Table 2. In general, treatments such AM inoculation alone or combined with other sources of nutrients significantly increased nutrient aupatke by soybean, compared with control. The highest increases were observed at treatment AM inoculation amended with CM. At this treatment, absorption of N, P, K, and Ca increased as much as 214%, 185%, 342%, and 233%, respectively on the first cropping cycle, and 191%, 308%, 224%, and 413%, consecutively on the second cropping series, compared with control. Treatment of AM inoculation followed by CM amendment also caused high absorption of P and Ca as much as 29% and 126%, respectively on the second cropping cycle.

The increase of P absorption could possibly cause such a new nutrient balance in plant that induced absorption of other nutrients such as N, K, and Ca. Sufficient availability of K created a condition in which the use of water was efficient as cell turgor was maintained. This condition leads to active metabolism process, K accumulation on the tips, buds, and roots, the accumulation that lead to formation of cortex tissue and cell elongation (Schweiger *et al.*, 2007 dan Smith *et al*,, 2010).

Table 2. Nutrient uptake (N,P, K and Ca) by soybean grown grown on sandy soil with various treatments.

Treatments	Nutrient uptake (mg/plant)							
	First cycle				Second cycle			
	N	P	K	Ca	N	P	K	Ca
60 DAS								
F_0	221.59[a]	22.43[a]	121.53[a]	25,60[a]	224.52[a]	20.37[a]	79.21[a]	37.66[a]
F_1	302.83[b]	29.03[b]	210.13[b]	34,16[b]	301.13[b]	34.46[b]	121.63[ab]	59.22[b]
F_2	697.16[c]	64.06[c]	537.33[c]	85,30[c]	653.42[c]	83.22[c]	257.43[c]	193.30[c]
F_3	412.73[d]	39.55[d]	282.73[d]	48,20[d]	429.60[d]	49.33[d]	185.20[d]	12.96[d]
F_4	344.10[e]	34.46[e]	232.46[e]	39,96[d]	311.60[b]	38.14[b]	159.44[ad]	96.93[d]

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF

The inoculation of AM followed by CM also increased plant dry biomass measured by root and shoot dry weights on 60 and 100 das (Table 3). Dry weight of roots and shoots on 60 das increased as much as 164% and 136%, respectively; while on 100 das the increases were as much as 150% and 178%, respectively on the first cropping cycle. On the second cycle, on 60 das the increases were 337% and 718%, while on 100 das the increases were as much as 390% and 1102%, consecutively.

Table 3. Root and shoot dry weight of soybean grown in sandy soil with various treatments. at 60 and 100 DAS

Treatments	Biomass dry weight (mg/plant)			
	First cycle		Second cycle	
	Roots	Shoots	Roots	Shoots
60 DAS				
F_0	1.88[a]	11.11[a]	1.74[a]	8.50[a]
F_1	2.66[b]	14.51[b]	2.27[b]	10.48[b]
F_2	4.97[c]	26.28[c]	4.38[c]	18.67[c]
F_3	3.62[d]	20.39[d]	3.75[d]	13.51[d]
F_4	3.34[d]	17.10[e]	2.51[b]	11.41[e]
100 DAS				
F_0	3.41[a]	22.43[a]	2.83[a]	19.94[a]
F_1	4.45[b]	39.63[b]	2.54[a]	19.35[a]
F_2	8.53[c]	62.45[c]	6.27[b]	32.92[b]
F_3	5.22[d]	44.50[d]	3.41[a]	26.40[c]
F_4	4.48[b]	37.85[b]	2.36[a]	19.91[c]

Note: Means followed by the same letters within the same column are not significantly different ($p=0.05$); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF.

The improvement of biomass was probably due to AM role to influence nutrient availability, especially P. Sufficient availability of P can indirectly induce absorption of other nutrients leading to better plant growth (Carrenho *et al*., 2001). This positive effect was suggested due to suitability of AM type, plant, and soluble P (Nikolaou *et al*., 2002; Bhadalung *et al*., 2005). Table 3 also showed significant effect of AM inoculation combined with CM as indicated by the increase of root and shoot dry weight. The similar results were also reported previously (Rossiana and Supriatun, 2003; Simarmata and Herdiani, 2003; Bertham, 2006).

Inoculation of AM with CM amendment increased cobs, weight of grain, and weight of 100 grains as much as 180%, 163%, and 139% respectively on the first cropping cycle and as much as 139%, 330%, and 23%, consecutively on the second cycle. These results were in accordance with results of previous studies indicating that fertilizer package with AM inoculation and amended with CM increased nutrient absorption, plant growth and yield (Astiko, 2009; Viti *et al.*, 2010). Similar results were also reported on dry paddy (Kabirun, 2002).

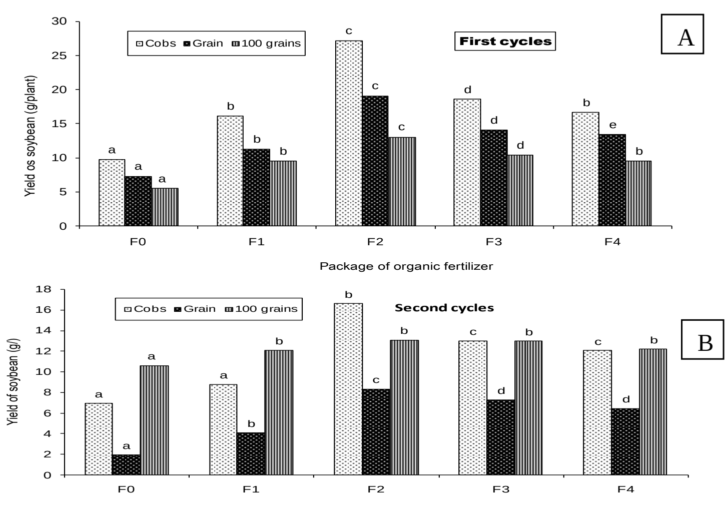

Fig 1. Yields of soybean (cobs, grain and 100 grain dry weight) grown in sandy soil with various treatments. A. the first cropping cycle, and B. the second cropping cycle. Bars with the same letters at the same category are not significantly different (p=0.05).

The increases of nutrient absorption, plant growth and yield on the tretmant of AM inoculation amended with CM were caused by the increase of AM activity in absorption of nutrient and water through its external hypha (EH). This was possibly due to EH can reach depletion zone that cannot be reached by plant roots (Zhu *et al*., 2001). The diameter of EH which is much smaller than that of roots makes the EH possibly to penetrate soil micro pores to take nutrient and water that cannor be reached by roots
(Drew *et al*., 2003). This ability causes plants with mycorrhiza be able to absorb nutrient, growth and perform better and resistance to dought stress (Smith and Read, 2008). All above facts indicated such a suitable functional among AM, host plants, and environment that are able to increase nutrient absorbtion, plant growth and yield as also reported earlier (Burleight *et al*., 2002).

Mycorrhiza activity

The inoculation of AM followed by CM amendment could increase AM activity as shown by numbers of spores per 100 g of soil and percentage of root infection (Tabel 4). The number of spores and percentage of infection on the first cropping cycle 60 das on sandy soil inoculated by AM and amended with CM increased as much as 179% and 266%, respectively, while on the second cycle the increases were 24% and 160%, respectively compared with control and were significantly different from other treatments.

Table 4. Biological activity of mycorrhiza (number of spores and percentage of infections) in sandy soil with various treatments.

Treatments	Spores 100 g soil^{-1} and root infection			
	First cycle		Second cycle	
	Spores	Infection	Spores	Infection
60 DAS				
F_0	1,060^a	21^a	3,122^a	25^a
F_1	2,159^b	41^b	3,533^b	34^b
F_2	2,960^c	77^c	3,878^c	65^c
F_3	2,343^d	54^d	3,781^d	51^d
F_4	2,215^b	46^e	3,693^e	41^e
Before exp[1)]	371	-	-	-

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; [1)] pre-treatment data

Table 4 showed that the number of spores and root infection were significantly high in all treatments when compared with control. Compared with before sowing, the number of spores and root infection 60 das increased significantly. This increases indicated that isolate M_{AA1} were able to compete with indigenuous AM present in the rhizosphere of sandy soil Northern Lombok, particularly in colony forming inside roots. The other point that can be taken from these facts was that the isolate M_{AA1} used was able to produce abundance propagules in the form of spores and such colonized roots that they were able to live on dynamic and competitive habitat (Barrios, 2007; Doud and Johnson, 2007; Gianinazzi *et al.*, 2010). This fact also indicated that isolate M_{AA1} showed high effectiveness, although they were inoculated on unsterile soil. Similar research on soybean inoculated by AM and the application of organic leaf fertilizer "greenstant" also showed similar results (Wangiyana *et al.*, 2007).

CONCLUSION

Inoculation of AM amended with cattle manure improved sandy soil fertility as shown by the increasing concentrations of N, P, K, and organic-C. Soybean responded positively to the application of AM followed by cattle manure as indicated by plant improvement in nutrient absorption, plant growth and yield. The addition of cattle manure stimulated activity of AM leading to improvement of soil fertility and plant performance.

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Reviewer's Attachments

A G R I V I T A

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MANUSCRIPT EVALUATION FORM

Manuscript Title : **SOIL FERTILITY STATUS AND SOYBEAN [*Glycine max* (L) Merr] PERFORMANCE FOLLOWING INTRODUCTION OF INDIGENOUS MYCORRHIZA COMBINED WITH VARIOUS NUTRIENT SOURCES INTO SANDY SOIL**

Manuscript no :

Evaluation Stage :

No.	Subject of Criteria	Weight (W)	Score (S)	W x S
1.	Abstract a) 200 words in maximum b) fully explaining manuscript content	10	5	
2.	Introduction a) Explaining the research significancies b) Relevancy to bibliographical coverage c) Up to dated current content	20	6	
3.	Material and Methods a) Material and methods appropriateness to research purposes and coverage b) sufficent analysis methods covering the problem discussed	25	5	
4.	Result and Discussion a) Clear result explanation and description b) Trusted analytical process and sufficient support of relevant information	25	6	
5.	Conclusion a) Explaining general results of research b) Quantitative expression is recommended	10	5	
6.	References a) Relevancy to the theme script b) Up to date (not more than 10 years) c) 60% references based on primary sources	10	6	
	Total	100		

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a. ~~No correction and dedicated to be published~~
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SOIL FERTILITY STATUS AND SOYBEAN [*Glycine max* (L) Merr] PERFORMANCE FOLLOWING INTRODUCTION OF INDIGENOUS MYCORRHIZA COMBINED WITH VARIOUS NUTRIENT SOURCES INTO SANDY SOIL

ABSTRACT

A series of experiment aimed at obtaining soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil was conducted in a glasshouse. The experiment tested four treatments, namely; inoculation of soil with mycorrhiza, inoculation of soil with mycorrhiza and cattle manure, inoculation of soil with mycorrhiza and rock phosphate, and inoculation of soil with mycorrhiza and inorganic fertilizers. Soil without any inoculation was provided as control. The treatments were arranged in a Completely Randomized Design with four replications. Soil fertility status was based on the concentrations of N, P, K, and organic-C as well as soil pH. Plant performance was determined based on its ability to uptake nutrients (N, P, K, and Ca), its growth and yield. Then, the activity of mycorrhiza was measured based on total population and percentage of root infection. Overall, results of the present study showed that, compared with other treatments, inoculation of mycorrhiza into soil and amended with cattle manure significantly performed higher concentrations of N, P, K, and organic-C. This soil condition caused soybean to absorb significantly higher nutrients, grew well with higher yield compared with plant performance in other treatments. Therefore, results of this study implies that the application of mycorrhiza into soil amended with organic matter is promising to sustain soil productivity under soybean cropping system.

Comment [T1]: Please define the general problem area. What does the author intend to discuss?
Why???

Key words: Arbuscular Mycorrhiza , soil fertility, soybean yield, dryland

INTRODUCTION

Shortage in availability of water, nutrients, and organic matter was a core problems in improving plant production in sandy soil of northern Lombok (Suwardji *et al*., 2007). Sandy soil with low organic matter content has low capacity in holding water and nutrients to support optimal plant growth (Suzuki and Noble, 2007; Bastida *et al*., 2010). This character of sandy soil is considered as a specific problem in managing sandy soil in northern Lombok, especially to grow soybean. To overcome this particular problem, a strategy in managing soil, in long term, to gain improvement and stability of soil organic matters which then lead to the improvement of soil characteristics especially in holding capacity of water and nutrients in the state of ready to be used by plants (Parman *et al*., 2000). In addition, soil management by improving role of indigenuous arbuscular mycorrhiza (AM) in sandy soil is one of best possible alternative solutions to improve plant productivity (Herrera *et al*., 1993 and Astiko, 2009).

Comment [T2]: More focus on the maun topic. Do you try to build on past research? SO, do it in these statements.

Optimizing role and beneficial characteristic of AM through application of biofertilizer to improve plant productivity in dry land is prospective enough (Sastrahidayat *et al*., 2001 dan Astiko *et al*., 2005). Application of AM by introducing the organisms into soil is expected to be able to improve plant productivity significantly through role of AM in improving plant capacity to absorb N, P, K, Ca and other micro nutrients. Besides, with its external hypha, AM will improve plant resistance on drought, protect plant root from soil-born pathogen infection, stimulate activity of beneficial microorganisms, and improve soil texture and structure (Gianinazzi and Vosátka, 2004; Feldmann *et al*., 2009; Ijdo *et al*., 2011).

Results of many previous studies have proven that nutrient absorption, growth and result of plant inoculated by AM were much higher than control (Azcón-Aguilar and Barea,

1997; Nogueira *et al.*, 2007; Fisher and Jayachandran, 2008). Furtheermore, it was reported that the use of indigenous AM had advantages as the microorganism was able to establish and develop well in situ environment as well as its ability to compete with existing soil microbes compared with introduced AM (Turrini *et al.*, 2008).

Incorporation of AM fungus on soybean in sandy soil of dry-land Northern Lombok was expected to have positive impact on soil characteristic improvement, nutrient absorption, and finally plant growth and yield. This hypothesis was constructed based on results of previous research in Vertisol soil (Astiko *et al*., 2005) proving that inoculation of AM on soybean improved absorption of P and plant yield was higher tan that of plant without AM. The improvement of P absorption was due to AM activity in improving nutrient availability and root proliferation (Smith *et al.*, 2010). This increase of plant yield was suggested due to the ability of AM to increase efficiency in wáter use, nutrient absorption, and to maintain turgor of plant cells. The external hypha of AM fungus were expected to be able to absorp soil pore wáter when plant roots anable to do so. In addition, wide distribution of external hypha caused the amount of wáter taken improved on the soil with low wáter content (Drew *et al*., 2003; Smith and Read, 2008). Base don the above mentioned, the aim of this study was to assess soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil.

METHODS

Comment [T3]: Add more for more detail information.

Experiment Preparation and Maintainance

A series of pot experiments using sandy soil taken Northern Lombok was conducted in a glasshouse of Faculty of Agriculture, Mataram University. The experiment was conducted

in two cropping cycles, namely; the first cropping cycle in which soybean was grown and fertilized according to treatments, and the second cropping cycle in which soybean was grown on soil that had been used for the first cropping cycle without fertilization. The study tested four treatments, namely; soil was inoculated with AM (F1), soil was inoculated with AM and amended with cattle manure (F2), soil was inoculated with AM and amended with rock phosphate (F3), and soil was inoculated with AM and amended with inorganic fertilizers (F4). Soil without AM inoculation and amendment was provided as control (F0). The treatments were completely randomized designed with four replications. The AM inoculums was prepared by mixing spores, powder medium, and root residues. The AM inoculum (20 gram per pot) then was inoculated at sowing time by layering the inoculum at 10 cm depth. Soybean seeds of Kaba var. were sown (2 seeds per pot) at 3 cm depth and 14 days after sowing (das), only one seedling per pot was left to grow further. Nutrient sources applied according to treatments were cattle manure, rock phosphate, Urea, and SP36. Rock phosphate and cattle manure were applied at rate of 1.2 and 2.0 g per plant respectively, and inorganic fertilizers of Urea and SP-36 were applied at rate of 0.1 g and 0.2 g per plant, consecutively. The nutrient sources were buried 5 cm around the plant at depth of 7 cm. The plants were harvested 100 das.

Parameters Observation

Comment [T4]: Is it parameters observation?? I dont think so..

Concentrations of N, P, K, and organic-C in soil, and soil pH were measured before sowing, 60 das and 100 das. Plant biomass (root and shoot dry weight) were measured at 60 das and 100 das after being oven dried at 60 ^{o}C for 48 hours. Nutrient uptake (N, P, K, and Ca) was measured at 60 das. Plant yield components recorded were weights of cobs, total

grains, and 100 grains at 100 das. Population of AM and percentage of root infection were measured at 60 das.

Soil, Plant Analysis and Mycorrhiza Observation

While analyses for N, P, and organic-C were done by using Kjeldhal method, spectrophotometer, and colorimetric method according to Walkley and Black, respectively, K and Ca were analyzed by using AAS. Mycorrhiza population was observed using wet sieving technique according to Brundrett *et al.* (1996). The supernatan caught at 38 µm-sieve was added with 60% of sucrosa solution and subsequently centrifused at 3000 rpm for 10 minute (Daniel and Skipper, 1982). The harvested spore were stored on the Whatman paper with permanent ink marked of 0,5 x 0,5 cm. Counting of mycoriza population was done using stereo mikroscope (extended 40 x). Calculation of roots percentage infections was conducted using modification of clearing and staining method (Kormanik and McGraw, 1982), counted with the *Gridline Intersect* technique (Giovenneti and Mosse, 1980) under stereo-microcope observation.

Statistical analysis

Comment [T5]: Not complete yet, it's important to be completed because your research depend on this statistical analysis.

Data were statistically analyzed for analysis of variance (ANOVA) and then continued using least significance different (LSD) at $p = 0.05$.

RESULTS AND DISCUSSION

Comment [T6]: Did you accomplish the objective? I dont think so. So, please accomplish the objective and that will give better information.

Soil chemical properties

In general, compared with control, all treatments significantly increased concentration of total N, available P and K, and organic-C both at 60 das and 100 das (Table 1). The highest

increases of nutrient concentrations were observed in sandy soil inoculated with arbuscular mycorrhiza (AM) and amended with cattle manure (CM). This indicated that AM performed well when combined with CM compared with other combinations. Compared with control, inoculation of AM amended with CM increased N, P, K, and org-C as much as 39%, 105%, 27%, and 85%, respectively on 60 das and 12%, 60%, 10%, and 11%, consecutively on 100 das at the first cropping cycle. Interestingly, these effects were also observed at the second cropping cycle as the total N, P, K, and org-C increased as much as 43%, 120%, 37%, and 36%, respectively on 60 das, and 44%, 53%, 14%, and 36%, respectively on 100 das. Data presented in Table 1 also indicated that at the second cropping cycle the treatment of AM plus CM could improve nutrient concentration of P and N as much as 4 and 48% on 60 das, respectively, and on 100 das the treatment increased N and Org-C as much as 12 and 20%, consecutively.

Results of this study were in accordance with those of done by Jeffries *et al*. (2003) and Gianinazzi *et al*. (2010) reporting that AM inoculation with organic matter amendment can recover soil fertility on an ecosystem. Furthermore, Douds *et al*. (2006) reported that AM inoculation could increase soil nutrient content. This increase was suggested due to activity of enzyme present in external hypha of AM in the rhizosphere able to catalize and hydrolyze unavailable nutrient complex into available (Widiastuti *et al.*, 2003).

Table 1. Soil chemical properties (N, P, K, organic-C and soil pH) of sandy soil with various treatments after harvesting.

Treatments	N ($g\ kg^{-1}$)		P ($mg\ kg^{-1}$)		K ($cmol\ kg^{-1}$)		org-C ($g\ kg^{-1}$)		pH	
	1st	2ad	1st	2ad	1st	2ad	1st	2ad	1st	2ad
60 DAS										

F_0	1.15[a]	0.87[a]	16.97[a]	16.54[a]	0.69[a]	0.51[a]	12.1[a]	24.5[a]	6.25[a]	6.13[a]
F_1	1.34[b]	1.15[b]	23.60[b]	23.40[b]	0.75[b]	0.58[b]	17.9[b]	26.5[b]	6.01[b]	6.24[b]
F_2	1.60[c]	1.25[b]	34.83[c]	36.54[c]	0.88[c]	0.70[c]	22.5[c]	33.5[c]	5.95[c]	6.32[c]
F_3	1.44[d]	1.10[b]	26.34[d]	27.01[d]	0.83[d]	0.63[d]	21.0[d]	29.4[d]	6.72[d]	6.28[d]
F_4	1.40[e]	0.93[b]	20.59[d]	30.97[e]	0.78[c]	0.62[bd]	19.1[e]	26.3[b]	6.08[e]	6.26[bd]
100 DAS										
F_0	1.31[a]	1.14[a]	17.62[a]	17.33[a]	0.75[a]	0.63[a]	23.8[a]	23.5[a]	6.18[a]	6.54[a]
F_1	1.44[b]	1.36[b]	20.86[b]	22.68[bc]	0.77[b]	0.65[b]	24.5[b]	25.3[ab]	6.21[b]	6.62[ab]
F_2	1.47[c]	1.65[c]	28.25[c]	26.57[d]	0.83[c]	0.72[c]	26.6[c]	32.1[c]	6.24[c]	6.54[ab]
F_3	1.38[d]	1.41[b]	23.32[d]	20.62[c]	0.81[d]	0.67[b]	23.9[d]	28.0[d]	6.62[d]	6.75[a]
F_4	1.33[a]	1.43[b]	24.38[e]	24.57[bd]	0.82[e]	0.70[c]	25.7[e]	27.5[d]	6.23[c]	6.44[b]
Before exp[1)]	1.1	-	13.82	-	0.57	-	12.1	-	6.25	-

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); 1st and 2ad first cycles and second cyles; F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; [1)] pre-treatment data

Previous results reported by Warnock *et al.*, (2007) showed that enrichment of AM could be escalated by the addition of organic matter and the combination has a positive effect on improving soil physico-chemical properties and therefore it is beneficial for soil microorganisms activities. This signify that better sinergy of the inoculated AM-tretment in combination with CM, as previously confirmerd by Kato and Miura (2008). Higher soil extractable-P found in this study was not only due to indirect contribution of CM in improving soil fertility status but also related to a positive contribution of mycorrhiza in producing phosphatase enzyme, for mineralization of organic-P in soil (Crowley and Rengel, 2000; Joner and Johansen, 2000) and resulted in enhancing insoluble-P in soils (Orcut and Nilsen, 2000).

Nutrient uptake, growth and yield

Nutrient absorption by soybean is presented in Table 2. In general, treatments such AM inoculation alone or combined with other sources of nutrients significantly increased nutrient aupatke by soybean, compared with control. The highest increases were observed at treatment

Comment [T7]: Please focus on the main topic about soybean performance. This discussion section also could easily be made more informative by placing your results in the context of previously published studies more so than in the current paper. A more complete comparison of your data to literature reports would help the reader to understand the newness of the findings and how they extend the work of others

AM inoculation amended with CM. At this treatment, absorption of N, P, K, and Ca increased as much as 214%, 185%, 342%, and 233%, respectively on the first cropping cycle, and 191%, 308%, 224%, and 413%, consecutively on the second cropping series, compared with control. Treatment of AM inoculation followed by CM amendment also caused high absorption of P and Ca as much as 29% and 126%, respectively on the second cropping cycle.

The increase of P absorption could possibly cause such a new nutrient balance in plant that induced absorption of other nutrients such as N, K, and Ca. Sufficient availability of K created a condition in which the use of water was efficient as cell turgor was maintained. This condition leads to active metabolism process, K accumulation on the tips, buds, and roots, the accumulation that lead to formation of cortex tissue and cell elongation (Schweiger *et al.*, 2007 dan Smith *et al,*, 2010).

Table 2. Nutrient uptake (N,P, K and Ca) by soybean grown grown on sandy soil with various treatments.

Treatments	Nutrient uptake (mg/plant)							
	First cycle				Second cycle			
	N	P	K	Ca	N	P	K	Ca
60 DAS								
F_0	221.59[a]	22.43[a]	121.53[a]	25,60[a]	224.52[a]	20.37[a]	79.21[a]	37.66[a]
F_1	302.83[b]	29.03[b]	210.13[b]	34,16[b]	301.13[b]	34.46[b]	121.63[ab]	59.22[b]
F_2	697.16[c]	64.06[c]	537.33[c]	85,30[c]	653.42[c]	83.22[c]	257.43[c]	193.30[c]
F_3	412.73[d]	39.55[d]	282.73[d]	48,20[d]	429.60[d]	49.33[d]	185.20[d]	12.96[d]
F_4	344.10[e]	34.46[e]	232.46[e]	39,96[d]	311.60[b]	38.14[b]	159.44[ad]	96.93[d]

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF

The inoculation of AM followed by CM also increased plant dry biomass measured by root and shoot dry weights on 60 and 100 das (Table 3). Dry weight of roots and shoots on 60 das increased as much as 164% and 136%, respectively; while on 100 das the increases were as much as 150% and 178%, respectively on the first cropping cycle. On the second cycle, on 60 das the increases were 337% and 718%, while on 100 das the increases were as much as 390% and 1102%, consecutively.

Table 3. Root and shoot dry weight of soybean grown in sandy soil with various treatments. at 60 and 100 DAS

Treatments	Biomass dry weight (mg/plant)			
	First cycle		Second cycle	
	Roots	Shoots	Roots	Shoots
60 DAS				
F_0	1.88[a]	11.11[a]	1.74[a]	8.50[a]
F_1	2.66[b]	14.51[b]	2.27[b]	10.48[b]
F_2	4.97[c]	26.28[c]	4.38[c]	18.67[c]
F_3	3.62[d]	20.39[d]	3.75[d]	13.51[d]
F_4	3.34[d]	17.10[e]	2.51[b]	11.41[e]
100 DAS				
F_0	3.41[a]	22.43[a]	2.83[a]	19.94[a]
F_1	4.45[b]	39.63[b]	2.54[a]	19.35[a]
F_2	8.53[c]	62.45[c]	6.27[b]	32.92[b]
F_3	5.22[d]	44.50[d]	3.41[a]	26.40[c]
F_4	4.48[b]	37.85[b]	2.36[a]	19.91[c]

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF.

The improvement of biomass was probably due to AM role to influence nutrient availability, especially P. Sufficient availability of P can indirectly induce absorption of other nutrients leading to better plant growth (Carrenho *et al*., 2001). This positive effect was suggested due to suitability of AM type, plant, and soluble P (Nikolaou *et al*., 2002; Bhadalung *et al*., 2005). Table 3 also showed significant effect of AM inoculation combined with CM as indicated by the increase of root and shoot dry weight. The similar results were also reported previously (Rossiana and Supriatun, 2003; Simarmata and Herdiani, 2003; Bertham, 2006).

Inoculation of AM with CM amendment increased cobs, weight of grain, and weight of 100 grains as much as 180%, 163%, and 139% respectively on the first cropping cycle and as much as 139%, 330%, and 23%, consecutively on the second cycle. These results were in accordance with results of previous studies indicating that fertilizer package with AM inoculation and amended with CM increased nutrient absorption, plant growth and yield (Astiko, 2009; Viti *et al.*, 2010). Similar results were also reported on dry paddy (Kabirun, 2002).

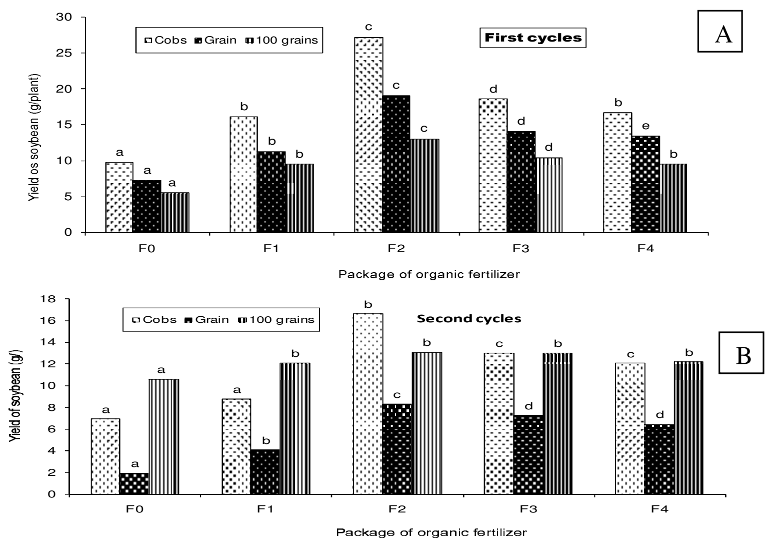

Fig 1. Yields of soybean (cobs, grain and 100 grain dry weight) grown in sandy soil with various treatments. A. the first cropping cycle, and B. the second cropping cycle. Bars with the same letters at the same category are not significantly different (p=0.05).

The increases of nutrient absorption, plant growth and yield on the tretmant of AM inoculation amended with CM were caused by the increase of AM activity in absorption of nutrient and water through its external hypha (EH). This was possibly due to EH can reach depletion zone that cannot be reached by plant roots (Zhu *et al.*, 2001). The diameter of EH

which is much smaller than that of roots makes the EH possibly to penetrate soil micro pores to take nutrient and water that cannor be reached by roots
(Drew *et al*., 2003). This ability causes plants with mycorrhiza be able to absorb nutrient, growth and perform better and resistance to dought stress (Smith and Read, 2008). All above facts indicated such a suitable functional among AM, host plants, and environment that are able to increase nutrient absorbtion, plant growth and yield as also reported earlier (Burleight *et al*., 2002).

Mycorrhiza activity

The inoculation of AM followed by CM amendment could increase AM activity as shown by numbers of spores per 100 g of soil and percentage of root infection (Tabel 4). The number of spores and percentage of infection on the first cropping cycle 60 das on sandy soil inoculated by AM and amended with CM increased as much as 179% and 266%, respectively, while on the second cycle the increases were 24% and 160%, respectively compared with control and were significantly different from other treatments.

Table 4. Biological activity of mycorrhiza (number of spores and percentage of infections) in sandy soil with various treatments.

Treatments	Spores 100 g soil^{-1} and root infection			
	First cycle		Second cycle	
	Spores	Infection	Spores	Infection
60 DAS				
F_0	1,060^{a}	21^{a}	3,122^{a}	25^{a}
F_1	2,159^{b}	41^{b}	3,533^{b}	34^{b}

F_2	2,960^c	77^c	3,878^c	65^c
F_3	2,343^d	54^d	3,781^d	51^d
F_4	2,215^b	46^e	3,693^e	41^e
Before exp[1)]	371	-	-	-

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; [1)] pre-treatment data

Table 4 showed that the number of spores and root infection were significantly high in all treatments when compared with control. Compared with before sowing, the number of spores and root infection 60 das increased significantly. This increases indicated that isolate M_{AA1} were able to compete with indigenuous AM present in the rhizosphere of sandy soil Northern Lombok, particularly in colony forming inside roots. The other point that can be taken from these facts was that the isolate M_{AA1} used was able to produce abundance propagules in the form of spores and such colonized roots that they were able to live on dynamic and competitive habitat (Barrios, 2007; Doud and Johnson, 2007; Gianinazzi *et al*., 2010). This fact also indicated that isolate M_{AA1} showed high effectiveness, although they were inoculated on unsterile soil. Similar research on soybean inoculated by AM and the application of organic leaf fertilizer “greenstant” also showed similar results (Wangiyana *et al.*, 2007).

CONCLUSION

Inoculation of AM amended with cattle manure improved sandy soil fertility as shown by the increasing concentrations of N, P, K, and organic-C. Soybean responded positively to the application of AM followed by cattle manure as indicated by plant improvement in nutrient absorption, plant growth and yield. The addition of cattle manure stimulated activity of AM leading to improvement of soil fertility and plant performance.

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SOIL FERTILITY STATUS AND SOYBEAN [*Glycine max* (L) Merr] PERFORMANCE FOLLOWING INTRODUCTION OF INDIGENOUS MYCORRHIZA COMBINED WITH VARIOUS NUTRIENT SOURCES INTO SANDY SOIL

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ABSTRACT

The experiment tested four treatments, namely; inoculation of soil with mycorrhiza, inoculation of soil with mycorrhiza and cattle manure, inoculation of soil with mycorrhiza and rock phosphate, and inoculation of soil with mycorrhiza and inorganic fertilizers. Soil without any inoculation was provided as control. The treatments were arranged in a Completely Randomized Design with four replications. Soil fertility status was based on the concentrations of N, P, K, and organic-C as well as soil pH. Plant performance was determined based on its ability to uptake nutrients (N, P, K, and Ca), its growth and yield. Then, the activity of mycorrhiza was measured based on total population and percentage of root infection. Overall, results of the present study showed that, compared with other treatments, inoculation of mycorrhiza into soil and amended with cattle manure significantly performed higher concentrations of N, P, K, and organic-C. This soil condition caused soybean to absorb significantly higher nutrients, grew well with higher yield compared with plant performance in other treatments. Therefore, results of this study implies that the application of mycorrhiza into soil amended with organic matter is promising to sustain soil productivity under soybean cropping system.
Keywords: *Arbuscular Mycorrhiza ,* soil fertility, soybean yield, dryland

INTRODUCTION

Shortage in availability of water, nutrients, and organic matter was a core problems in improving plant production in sandy soil of northern Lombok (Suwardji *et al*., 2007). Sandy soil with low organic matter content has low capacity in holding water and nutrients to support optimal soybean performance (Suzuki and Noble, 2007; Bastida *et al*., 2010). This character of sandy soil is considered as a specific problem in managing sandy soil in northern Lombok, especially to grow soybean. To overcome this particular problem, a strategy in managing soil, in long term, to gain improvement and stability of soil organic matters which then lead to the improvement of soil characteristics especially in holding capacity of water, plant performance and nutrients in the state of ready to be used by plants (Astiko *et al*., 2013). In addition, soil management by improving role of indigenuous arbuscular mycorrhiza (AM) in sandy soil is one of best possible alternative solutions to improve plant productivity (Herrera *et al*., 1993 and Astiko, 2009). This was due to the role of AM in improving soil quality through the improvement of agregate and colloid of the soil (Ijdo *et al*., 2011). This role bécame better in soil with adequate content of organic matter supplying carbón and micro nutrients required by AM to grow (Öpik *et al*., 2008; Smith and Read, 2008).

Optimizing role and beneficial characteristic of AM through application of biofertilizer to improve plant productivity in dry land is prospective enough (Sastrahidayat *et al.*, 2001 and Astiko *et al*., 2005). Application of AM by introducing the organisms into soil is expected to be able to improve plant productivity significantly through role of AM in improving plant capacity to absorb N, P, K, Ca and other micro nutrients. Besides, with its external hypha, AM will improve plant resistance on drought, protect plant root from soil-born pathogen infection, stimulate activity of beneficial microorganisms, and improve soil texture and structure (Gianinazzi and Vosátka, 2004; Feldmann *et al*., 2009; Ijdo *et al*., 2011).

Results of many previous studies have proven that nutrient absorption, growth and result of plant inoculated by AM were much higher than control (Azcón-Aguilar and Barea, 1997; Nogueira *et* *al.*, 2007; Fisher and Jayachandran, 2008). Furtheermore, it was reported that the use of indigenous AM had advantages as the microorganism was able to establish and develop well in situ environment as well as its ability to compete with existing soil microbes compared with introduced AM (Turrini *et al.*, 2008).

Incorporation of AM fungus on soybean in sandy soil of dry-land Northern Lombok was expected to have positive impact on soil characteristic improvement, nutrient absorption, and finally plant growth and yield. This hypothesis was constructed based on results of previous research in Vertisol soil (Astiko *et al*., 2005) proving that inoculation of AM on soybean improved absorption of P and plant yield was higher tan that of plant without AM. The improvement of P absorption was due to AM activity in improving nutrient availability and root proliferation (Smith *et al.*, 2010). This increase of plant yield was suggested due to the ability of AM to increase efficiency in wáter use, nutrient absorption, and to maintain turgor of plant cells. The external hypha of AM fungus were expected to be able to absorp soil pore wáter when plant roots anable to do so. In addition, wide distribution of external hypha caused the amount of wáter taken improved on the soil with low wáter content (Drew *et al*., 2003; Smith and Read, 2008). Base don the above mentioned, the aim of this study was to assess soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil.

MATERIALS AND METHODS

Experiment Preparation and Maintainance

A series of pot experiments using sandy soil taken Northern Lombok was conducted in a glasshouse of Faculty of Agriculture, Mataram University using 10 kg polybag as experimental unit. Taken from upper layer up to 30 cm depth, soil was passed through 2-mm holes sieve, air dried, then filled into the polybags. The experiment was conducted in two cropping cycles, namely; the first cropping cycle in which soybean was grown and fertilized according to treatments, and the second cropping cycle in which soybean was grown on soil that had been used for the first cropping cycle without fertilization. The study tested four treatments, namely; soil was inoculated with AM (F1), soil was inoculated with AM and amended with cattle manure (F2), soil was inoculated with AM and amended with rock phosphate (F3), and soil was inoculated with AM and amended with inorganic fertilizers (F4). Soil without AM inoculation and amendment was provided as control (F0). The treatments were completely randomized designed with four replications. AM inoculum was mass produced in corn plants grown in pots. Pots were filled with mixture of sandy soil and sterile cattle manure (ratio 1:1). The soil was watered aboued capacity and the plants were maintained in glass house for three months. Then the plants were harvested, dried, blundered, and passed through 50-mesh sieve. The final form of the proliferation was powder. The AM inoculums was prepared by mixing spores, powder medium, and root residues. The AM inoculum (20 gram per pot) then was inoculated at sowing time by layering the inoculum at 10 cm depth. Soybean seeds of Kaba var. were sown (2 seeds per pot) at 3 cm depth and 14 days after sowing (das), only one seedling per pot was left to grow further. Nutrient sources applied according to treatments were cattle manure, rock phosphate, Urea, and SP36. Rock phosphate and cattle manure were applied at rate of 1.2 and 2.0 g per plant respectively, and inorganic fertilizers of Urea and SP-36 were applied at rate of 0.1 g and 0.2 g per plant, consecutively. The nutrient sources were buried 5 cm around the plant at depth of 7 cm. Weeding was done manually while watering plants in the afternoon. Plnat protection was done by applying organic pesticide 0.5% Azadirachtin (OrgaNeem^{TM}) every three days. The plants were harvested 100 das.

Parameters Observation

Parameters dealing with soil fertility status (N, P, K, organic-C and soil pH) were measured before sowing and at 60 and 100 das. The agronomic parameters such as: top and roots dry weight biomass (60 das and 100 das), nutrients uptake (N, P, K, and Ca) at 60 das and component yield of soybean (cops, grain and 100 grain weight). The dry weight of the agronomic parameters were measured after being oven dried at 60 ºC for 48 hours. Parameters related to AM activities including fungi population, roots percentage infections at 60 das were also measured. Plant analysis for N was determined using Kjeldhal method, P using spectrophotometer, C-organic with colorimetric method according to Walkley and Black, K and Ca was recorded using Automatic Absorbtion Spectrophotometer (AAS).

Soil, Plant Analysis and Mycorrhiza Observation

While analyses for N, P, and organic-C were done by using Kjeldhal method, spectrophotometer, and colorimetric method according to Walkley and Black, respectively, K and Ca were analyzed by using AAS. Mycorrhiza population was observed using wet sieving technique according to Brundrett *et al.* (1996). The supernatan caught at 38 µm-sieve was added with 60% of sucrosa solution and subsequently centrifused at 3000 rpm for 10 minute (Daniel and Skipper, 1982). The harvested spore were stored on the Whatman paper with permanent ink marked of 0,5 x 0,5 cm. Counting of mycoriza population was done using stereo mikroscope (extended 40 x). Calculation of roots percentage infections was conducted using modification of clearing and staining method (Kormanik and McGraw, 1982), counted with the *Gridline Intersect* technique (Giovenneti and Mosse, 1980) under stereo-microcope observation.

Statistical analysis

To know if there is any difference among treatments, a mathematic model below was applied:

$X_{ijk} = \mu + \rho_i + \beta_j + \varepsilon_{ij}$

Remarks:

μ = general average

ρ_i = effect of replication-i

β_j = effect of treatment-j

ε_{ij} = galat

The model was applied when $X_{ijk} = \mu + \rho_i + \beta_j$ linear and additive to $\Sigma\rho_i = \Sigma\beta_j = 0$, and ε_{ijk} was free and normally distributed with average and variance = $(0,\sigma^2)$. Data were analysed by analysis of variance (ANOVA-MStat) and the effect of treatments was determined. When the variance ratio (F) was significant, means for each treatment were separated using a least significant difference test at 5% level.

RESULTS AND DISCUSSION

Soil Chemical Properties

In general, compared with control, all treatments significantly increased soil fertility status as indicated by concentration of total N, available P and K, and organic-C both at 60 das and 100 das (Table 1). The highest increases of soil fertility status were observed in sandy soil inoculated with arbuscular mycorrhiza (AM) and amended with cattle manure (CM). This indicated that AM performed well when combined with CM compared with other combinations. Compared with control, inoculation of AM amended with CM increased N, P, K, and org-C as much as 39%, 105%, 27%, and 85%, respectively on 60 das and 12%, 60%, 10%, and 11%, consecutively on 100 das at the first cropping cycle. Interestingly, these effects were also observed at the second cropping cycle as the total N, P, K, and org-C increased as much as 43%, 120%, 37%, and 36%, respectively on 60 das, and 44%, 53%, 14%, and 36%, respectively on 100 das. Data presented in Table 1 also indicated that at the second cropping cycle soybean performance on treatment of AM plus CM could improve soil fertility status as shown by nutrient concentration of P and N as much as 4 and 48% on 60 das, respectively, and on 100 das the treatment increased N and Org-C as much as 12 and 20%, consecutively.

Results of this study were in accordance with those of done by Jeffries *et al*. (2003) and Gianinazzi *et al*. (2010) reporting that AM inoculation with organic matter amendment can recover soil fertility status on an ecosystem. Furthermore, Douds *et al*. (2006) reported that introduction AM inoculation combined with various nutrient sorces into sandy soil could increase soil nutrient content. The same result was also reported by Astiko *et al*. (2013) evaluating contribution of indigenous AM combined with cattle manure to increase corn performance in snady soil of northern Lombok.

Combination of AM and cattle manure resulted in significant improvement on soil fertility status especially N, P, K, and organic-C. This increase was suggested due to activity of enzyme present in external hypha of AM in the rhizosphere able to catalize and hydrolyze unavailable nutrient complex into available (Widiastuti *et al.*, 2003).

Table 1. Soil fertility status (N, P, K, organic-C and soil pH) of sandy soil with various treatments after harvesting

Treatments	N ($g\ kg^{-1}$)		P ($mg\ kg^{-1}$)		K ($cmol\ kg^{-1}$)		org-C ($g\ kg^{-1}$)		pH	
	1^{st}	2^{ad}	1^{st}	2^{ad}	1^{st}	2^{ad}	1^{st}	2^{ad}	1^{st}	2^{ad}
60 DAS										
F_0	1.15^{a}	0.87^{a}	16.97^{a}	16.54^{a}	0.69^{a}	0.51^{a}	12.1^{a}	24.5^{a}	6.25^{a}	6.13^{a}
F_1	1.34^{b}	1.15^{b}	23.60^{b}	23.40^{b}	0.75^{b}	0.58^{b}	17.9^{b}	26.5^{b}	6.01^{b}	6.24^{b}
F_2	1.60^{c}	1.25^{b}	34.83^{c}	36.54^{c}	0.88^{c}	0.70^{c}	22.5^{c}	33.5^{c}	5.95^{c}	6.32^{c}
F_3	1.44^{d}	1.10^{b}	26.34^{d}	27.01^{d}	0.83^{d}	0.63^{d}	21.0^{d}	29.4^{d}	6.72^{d}	6.28^{d}
F_4	1.40^{e}	0.93^{b}	20.59^{d}	30.97^{e}	0.78^{c}	0.62^{bd}	19.1^{e}	26.3^{b}	6.08^{e}	6.26^{bd}
100 DAS										
F_0	1.31^{a}	1.14^{a}	17.62^{a}	17.33^{a}	0.75^{a}	0.63^{a}	23.8^{a}	23.5^{a}	6.18^{a}	6.54^{a}
F_1	1.44^{b}	1.36^{b}	20.86^{b}	22.68^{bc}	0.77^{b}	0.65^{b}	24.5^{b}	25.3^{ab}	6.21^{b}	6.62^{ab}
F_2	1.47^{c}	1.65^{c}	28.25^{c}	26.57^{d}	0.83^{c}	0.72^{c}	26.6^{c}	32.1^{c}	6.24^{c}	6.54^{ab}
F_3	1.38^{d}	1.41^{b}	23.32^{d}	20.62^{c}	0.81^{d}	0.67^{b}	23.9^{d}	28.0^{d}	6.62^{d}	6.75^{a}
F_4	1.33^{a}	1.43^{b}	24.38^{e}	24.57^{bd}	0.82^{e}	0.70^{c}	25.7^{e}	27.5^{d}	6.23^{c}	6.44^{b}
Before exp$^{1)}$	1.1	-	13.82	-	0.57	-	12.1	-	6.25	-

Remarks: Means followed by the same letters within the same column are not significantly different (p=0.05); 1^{st} and 2^{ad} first cycles and second cyles; F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; $^{1)}$ pre-treatment data

Previous results reported by Warnock *et al.*, (2007) showed that enrichment of AM could be escalated by the addition of organic matter and the combination has a positive effect on improving soil fertility status and therefore it is beneficial for soybean performance. This signify that better sinergy of the inoculated AM-tretment in combination with CM, as previously confirmerd by Kato and Miura (2008). Higher soil extractable-P found in this study was not only due to indirect contribution of CM in improving soil fertility status but also related to a positive contribution of mycorrhiza in producing phosphatase enzyme, for mineralization of organic-P in soil (Crowley and Rengel, 2000; Joner and Johansen, 2000) and resulted in enhancing insoluble-P in soils (Orcut and Nilsen, 2000).

Nutrient Uptake, Growth and Yield

Nutrient absorption by soybean is presented in Table 2. In general, treatments such AM inoculation alone or combined with other sources of nutrients significantly increased nutrient aupatke by soybean, compared with control. The highest increases were observed at treatment AM inoculation amended with CM. At this treatment, absorption of N, P, K, and Ca increased as much as 214%, 185%, 342%, and 233%, respectively on the first cropping cycle, and 191%, 308%, 224%, and 413%, consecutively on the second cropping series, compared with control. Treatment of AM inoculation followed by CM amendment also caused high absorption of P and Ca as much as 29% and 126%, respectively on the second cropping cycle. This indicated an increase in soybean performance when grown in sandy soil amended with various nutrient sources.

The increase of P absorption could possibly cause such a new nutrient balance in plant that induced absorption of other nutrients such as N, K, and Ca. Sufficient availability of K created a condition in which the use of water was efficient as cell turgor was maintained. This condition leads to active metabolism process, K accumulation on the tips, buds, and roots, the accumulation that lead to formation of cortex tissue and cell elongation leading to improvement of soybean performance (Schweiger *et al.*, 2007 and Smith *et al,*, 2010). This result was in accordance with that of Kaschuk *et al*. (2010) stating that the increase of nutrient abrorption by host plant lead to plant vitality to supply Carbon to rhizosphere to form AM external hypha. Mathur and Vyas (2000) stated that AM inoculation was also resulted in the increase of accumulation of amino acids, protein, chlorophyll, and sugar contents compared with non-AM plants. N status of shoot tip of plant with mycorrhiza at extreme condition was higher than that of plant ithout mycorrhiza (Subramania and Charest, 1999).The same trends were also recorded for N, P, K, and Ca (Liu *et al*., 2000).

Table 2. Nutrient uptake (N,P, K and Ca) by soybean grown in sandy soil with various treatments.

Treatments	Nutrient uptake (mg/plant)							
	First cycle				Second cycle			
	N	P	K	Ca	N	P	K	Ca
60 DAS								
F_0	221.59^{a}	22.43^{a}	121.53^{a}	25,60^{a}	224.52^{a}	20.37^{a}	79.21^{a}	37.66^{a}
F_1	302.83^{b}	29.03^{b}	210.13^{b}	34,16^{b}	301.13^{b}	34.46^{b}	121.63^{ab}	59.22^{b}
F_2	697.16^{c}	64.06^{c}	537.33^{c}	85,30^{c}	653.42^{c}	83.22^{c}	257.43^{c}	193.30^{c}
F_3	412.73^{d}	39.55^{d}	282.73^{d}	48,20^{d}	429.60^{d}	49.33^{d}	185.20^{d}	12.96^{d}
F_4	344.10^{e}	34.46^{e}	232.46^{e}	39,96^{d}	311.60^{b}	38.14^{b}	159.44^{ad}	96.93^{d}

Remarks: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF

The inoculation of AM followed by CM also increased soybean performance, as observed on plant dry biomass measured by root and shoot dry weights on 60 and 100 das (Table 3). Dry weight of roots and shoots on 60 das increased as much as 164% and 136%, respectively; while on 100 das the increases were as much as 150% and 178%, respectively on the first cropping cycle. On the second cycle, on 60 das the increases were 337% and 718%, while on 100 das the increases were as much as 390% and 1102%, consecutively.

Table 3. Root and shoot dry weight of soybean grown in sandy soil with various treatments at 60 and 100 das

Treatments	Biomass dry weight (g/plant)			
	First cycle		Second cycle	
	Roots	Shoots	Roots	Shoots
60 DAS				
F_0	1.88^{a}	11.11^{a}	1.74^{a}	8.50^{a}
F_1	2.66^{b}	14.51^{b}	2.27^{b}	10.48^{b}
F_2	4.97^{c}	26.28^{c}	4.38^{c}	18.67^{c}
F_3	3.62^{d}	20.39^{d}	3.75^{d}	13.51^{d}
F_4	3.34^{d}	17.10^{e}	2.51^{b}	11.41^{e}
100 DAS				
F_0	3.41^{a}	22.43^{a}	2.83^{a}	19.94^{a}
F_1	4.45^{b}	39.63^{b}	2.54^{a}	19.35^{a}
F_2	8.53^{c}	62.45^{c}	6.27^{b}	32.92^{b}
F_3	5.22^{d}	44.50^{d}	3.41^{a}	26.40^{c}
F_4	4.48^{b}	37.85^{b}	2.36^{a}	19.91^{c}

Remarks: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF

The improvement of biomass was probably due to AM role to influence soil fertility status, especially P. Sufficient availability of P can indirectly induce absorption of other nutrients leading to better plant growth (Carrenho *et al*., 2001). This positive effect was suggested due to suitability of AM type, plant, and soluble P (Nikolaou *et al*., 2002; Bhadalung *et al*., 2005). This condition was due to good association between plant and AM to perform maximal actrivity. While AM received carbon from the plant, and the latter got P from the first. It was reported that each particular combination of AM and plant showed such a specific carbon translocation pattern that production of plant biomass with AM was influenced (Smith and Read, 2008; Smith *et al*., 2009). Inefficient symbiosis in use of carbon could decrease plant biomass. AM colonization could result in positive, neutral, or negative impact depending on AM types, plants, and growth environment (Johnson *et al*., 1997; Hoeksema *et al*., 2010). Negative impact of mycorrhiza colonization on initial growth of plant was reported previously (Bethlenfalvay *et al*., 1982; Koide 1985; Johnson *et al*., 1997). Such sort of impact may be due to various factors, such as; high availability of P in soil (Mosse *et al*., 1973), competition for carbon between plant and AM in low light intensity condition (Buwalda and Goh 1982), and difference in biomass allocation pattern between plant with and without mycorrhiza (Smith and Read 2008). Therefore, the use of low solubility of phosphate, like phosphate stone, although with high dosage, in fact, was still effective to support AM and to increase plant performance (Nikolaou *et al*., 2002). In addition, quite high dependency of plant on AM indicated that in early stage of its growth, plant needs to associate with AM. Table 3 also showed significant effect of AM inoculation combined with CM as indicated by the increase plant performance as observed on root and shoot dry weight. The similar results were also reported previously (Rochdjatun *et al*., 2011; Astiko *et al*., 2012).

Inoculation of AM with CM amendment increased plant performance as observed on cobs weight, weight of grain, and weight of 100 grains as much as 180%, 163%, and 139% respectively on the first cropping cycle and as much as 139%, 330%, and 23%, consecutively on the second cycle. These results were in accordance with results of previous studies indicating that fertilizer package with AM inoculation and amended with CM increased soil fertility status, plant performance and yield (Astiko, 2009; Viti *et al.*, 2010). Similar results were also reported on dry paddy (Kabirun, 2002).

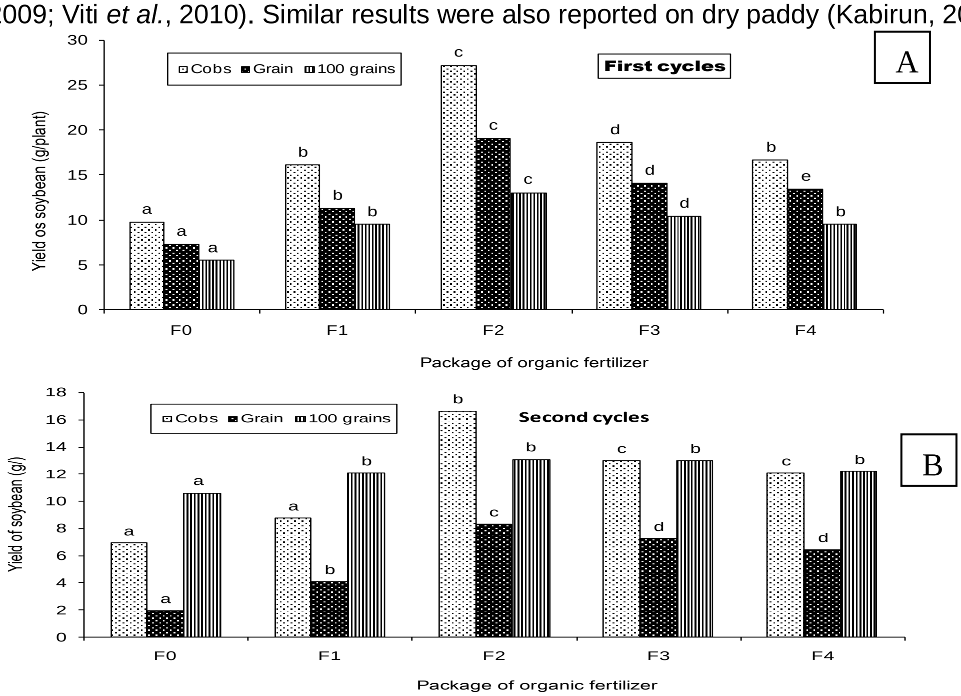

Figure 1. Yields of soybean (cobs, grain and 100 grain dry weight) grown in sandy soil with various treatments. A. the first cropping cycle, and B. the second cropping cycle. Bars with the same letters at the same category are not significantly different (p=0.05).

The increases of soil fertility status, plant performance and yield on the tretmant of AM inoculation amended with CM were caused by the increase of AM activity in absorption of nutrient and water through its external hypha (EH). This was possibly due to EH can reach depletion zone that cannot be reached by plant roots (Zhu *et al*., 2001). The diameter of EH which is much smaller than that of roots makes the EH possibly to penetrate soil micro pores to take nutrient and water that cannor be reached by roots (Drew *et al*., 2003). This ability causes plants with mycorrhiza be able to absorb nutrient, growth and perform better and resistance to dought stress (Smith and Read, 2008). In addition, AM is able to dilute phosphate tied in soil and fertilizer, to improve absorption of N, P, and K, to improve plant tolerance to drought, to improve plant ability to produce growth regulator, to stimulate activity of beneficial microbes, to improve soil structure and aggregation, and to enhance mineral cyclic (Cruz, 1990). Decomposition and mineralization of organic matter were better with the presence of AM which positively affect physic, chemistry, and biology factors of the soil which in turn play key role in improving plant yield (Smith and Read, 2008). All above facts indicated such a suitable functional among AM, host plants, and environment that are able to increase nutrient absorbtion, plant growth and yield as also reported earlier (Burleight *et al*., 2002).

Mycorrhiza activity

The inoculation of AM followed by CM amendment could increase AM activity as shown by numbers of spores per 100 g of soil and percentage of root infection (Tabel 4). The number of spores and percentage of infection on the first cropping cycle 60 das on sandy soil inoculated by AM and amended with CM increased as much as 179% and 266%, respectively, while on the second cycle the increases were 24% and 160%, respectively compared with control and were significantly different from other treatments.

Table 4. Biological activity of mycorrhiza (number of spores and percentage of infections) in sandy soil with various treatments.

Treatments	Spores 100 g soil^{-1} and root infection			
	First cycle		Second cycle	
	Spores	Infection	Spores	Infection
60 DAS				
F_0	1,060^a	21^a	3,122^a	25^a
F_1	2,159^b	41^b	3,533^b	34^b
F_2	2,960^c	77^c	3,878^c	65^c
F_3	2,343^d	54^d	3,781^d	51^d
F_4	2,215^b	46^e	3,693^e	41^e
Before exp[1)]	371	-	-	-

Note: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; [1)] pre-treatment data

Table 4 showed that the number of spores and root infection were significantly high in all treatments when compared with control. Compared with before sowing, the number of spores and root infection 60 das increased significantly. This increases indicated that isolate M_{AA1} were able to compete with indigenuous AM present in the rhizosphere of sandy soil Northern Lombok, particularly in colony forming inside roots. The other point that can be taken from these facts was that the isolate M_{AA1} used was able to produce abundance propagules in the form of spores and such colonized roots that they were able to live on dynamic and competitive habitat (Barrios, 2007; Doud and Johnson, 2007; Gianinazzi *et al*., 2010). Result of a research on the role of indigenous mycorrhiza combined with cattle manure in improving yield of maize (*Zea mays* L.) on sandy loam of Norhern Lombok showed that inoculation of AM combined with cattle manure resulted in higher number of spores and infected roots both in the first and the second growth season (Astiko *et al*., 2013). Similar results were also shown by Prasetya and Anderson (2011) on the assessment of the effect of long term tillage on the arbuscular mycorrhiza colonitation of vegetable crops grown in andisols. This fact also indicated that isolate M_{AA1} showed high effectiveness, although they were inoculated on unsterile soil. Similar research on soybean inoculated by AM and the application of organic leaf fertilizer “greenstant” also showed similar results (Wangiyana *et al.*, 2007).

CONCLUSION

Inoculation of AM amended with cattle manure improved sandy soil fertility as shown by the increasing soil fertility status as observed on concentrations of N, P, K, and organic-C. Soybean performance responded positively to the application of AM followed by cattle manure as indicated by plant improvement in nutrient absorption, plant growth and yield. The addition of cattle manure stimulated activity of AM leading to improvement of soil fertility and plant performance.

ACKNOWLEDGEMENTS

This manuscript is a part of the first authors dissertation for the Doctoral Program funded by Post Graduate Scholarships at Agriculural Faculty University of Brawijaya Malang. Thank to the institution of Directorate General of Higher Education (DGHE) as the funders of Ph.D research for 2012 Grant Program by DIPA Number: 023.04.2.415278/2013, December 5, 2012, Letter of Agreement, Impelemtation Research Number: 295.G/SPP-APDD/H18.12/PL/2013, May 2, 2013.

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Manuscript Title SOIL FERTILITY STATUS AND SOYBEAN [*Glycine max* (L) Merr] PERFORMANCE FOLLOWING INTRODUCTION OF INDIGENOUS MYCORRHIZA COMBINED WITH VARIOUS NUTRIENT SOURCES INTO SANDY SOIL

Declare that the author (s) have read and make some correction in the proof of manuscriptand no more error (s) in the manuscript.

Malang, October 26. 2013

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AGRIVITA VOLUME 35 No. 2 JUNE - 2013 ISSN : 0126-0537

SOIL FERTILITY STATUS AND SOYBEAN [*Glycine max* (L) Merr] PERFORMANCE FOLLOWING INTRODUCTION OF INDIGENOUS MYCORRHIZA COMBINED WITH VARIOUS NUTRIENT SOURCES INTO SANDY SOIL

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Received: January 2, 2013/ Accepted: September, 2013

ABSTRACT

The experiment tested four treatments, namely inoculation of soil with mycorrhiza, inoculation of soil with mycorrhiza and cattle manure, inoculation of soil with mycorrhiza and rock phosphate, and inoculation of soil with mycorrhiza and inorganic fertilizers. Soil without any inoculation was provided as control. The treatments were arranged in a Completely Randomized Design with four replications. Soil fertility status was based on the concentrations of N, P, K, and organic-C as well as soil pH. Plant performance was determined based on its ability to uptake nutrients (N, P, K, and Ca), its growth and yield. Then, the activity of mycorrhiza was measured based on total population and percentage of root infection. Overall, results of the present study showed that, compared with other treatments, inoculation of mycorrhiza into soil and amended with cattle manure significantly performed higher concentrations of N, P, K, and organic-C. This soil condition caused soybean to absorb significantly higher nutrients, grew well with higher yield compared with plant performance in other treatments. Therefore, results of this study implies that the application of mycorrhiza into soil amended with organic matter is promising to sustain soil productivity under soybean cropping system.

Keywords: *Arbuscular Mycorrhiza,* soil fertility, soybean yield, dryland

INTRODUCTION

Shortage in availability of water, nutrients, and organic matter was a core problems in improving plant production in sandy soil of northern Lombok (Suwardji *et al*., 2007). Sandy soil with low organic matter content has low capacity in holding water and nutrients to support optimal soybean performance (Suzuki and Noble, 2007; Bastida *et al*., 2010). This character of sandy soil is considered as a specific problem in managing sandy soil in northern Lombok, especially to grow soybean. To overcome this particular problem, a strategy in managing soil, in long term, to gain improvement and stability of soil organic matters which then lead to the improvement of soil characteristics especially in holding capacity of water, plant performance and nutrients in the state of ready to be used by plants (Astiko *et al*., 2013). In addition, soil management by improving role of indigenuous arbuscular mycorrhiza (AM) in sandy soil is one of best possible alternative solutions to improve plant productivity (Herrera *et al*., 1993 and Astiko, 2009). This was due to the role of AM in improving soil quality through the improvement of agregate and colloid of the soil (Ijdo *et al*., 2011). This role bécame better in soil with adequate content of organic matter supplying carbón and micro nutrients required by AM to grow (Öpik *et al*., 2008; Smith and Read, 2008).

Optimizing role and beneficial characteristic of AM through application of biofertilizer to improve plant productivity in dry land is prospective enough (Sastrahidayat *et al.*, 2001 and Astiko *et al*., 2005). Application of AM by introducing the organisms into soil is expected to be able to improve plant productivity significantly through role of AM in improving plant capacity to absorb N, P, K, Ca and other micro nutrients. Besides, with its external hypha, AM will improve plant resistance on drought, protect plant root from soil-born pathogen infection, stimulate activity of beneficial microorganisms, and improve soil texture and structure (Gianinazzi and Vosátka, 2004; Feldmann *et al*., 2009; Ijdo *et al*., 2011).

Results of many previous studies have proven that nutrient absorption, growth and result of plant inoculated by AM were much higher than control (Azcón-Aguilar and Barea, 1997; Nogueira *et al*., 2007; Fisher and Jayachandran, 2008). Furthermore, it was reported that the use of indigenous AM had advantages as the microorganism was able to establish and develop well in situ environment as well as its better ability to compete with existing soil microbes compared with introduced AM (Turrini *et al*., 2008).

Incorporation of AM fungus on soybean in sandy soil of dry-land Northern Lombok was expected to have positive impact on soil characteristic improvement, nutrient absorption, and finally plant growth and yield. This hypothesis was constructed based on results of previous research in Vertisol soil (Astiko *et al*., 2005) proving that inoculation of AM on soybean improved absorption of P and plant yield was higher tan that of plant without AM. The improvement of P absorption was due to AM activity in improving nutrient availability and root proliferation (Smith *et al*., 2010). This increase of plant yield was suggested due to the ability of AM to increase efficiency in wáter use, nutrient absorption, and to maintain turgor of plant cells. The external hypha of AM fungus were expected to be able to absorp soil pore wáter when plant roots enable to do so. In addition, wide distribution of external hypha caused the amount of wáter taken improved on the soil with low wáter content (Drew *et al*., 2003; Smith and Read, 2008). Base on the above mentioned, the aim of this study was to assess soil fertility status and soybean performance following introduction of indigenous mycorrhiza combined with various nutrient sources into sandy soil.

MATERIALS AND METHODS

Experiment Preparation and Maintainance

A series of pot experiments using sandy soil taken Northern Lombok was conducted in a glasshouse of Faculty of Agriculture, Mataram University using 10 kg polybag as experimental unit. Taken from upper layer up to 30 cm depth, soil was passed through 2-mm holes sieve, air dried, then filled into the polybags. The experiment was conducted in two cropping cycles, namely; the first cropping cycle in which soybean was grown and fertilized according to treatments, and the second cropping cycle in which soybean was grown on soil that had been used for the first cropping cycle without fertilization. The study tested four treatments, namely; soil was inoculated with AM (F1), soil was inoculated with AM and amended with cattle manure (F2), soil was inoculated with AM and amended with rock phosphate (F3), and soil was inoculated with AM and amended with inorganic fertilizers (F4). Soil without AM inoculation and amendment was provided as control (F0). The treatments were completely randomized designed with four replications. AM inoculum was mass produced in corn plants grown in pots. Pots were filled with mixture of sandy soil and sterile cattle manure (ratio 1:1). The soil was watered about field capacity and the plants were maintained in glass house for three months. Then the plants were harvested, dried, blended**,** and passed through 50-mesh sieve. The final form of the proliferation was powder. The AM inoculums was prepared by mixing spores, powder medium, and root residues. The AM inoculum (20 gram per pot) then was inoculated at sowing time by layering the inoculum at 10 cm depth. Soybean seeds of Kaba var. were sown (2 seeds per pot) at 3 cm depth and 14 days after sowing (das), only one seedling per pot was left to grow further. Nutrient sources applied according to treatments were cattle manure, rock phosphate, Urea, and SP36. Rock phosphate and cattle manure were applied at rate of 1.2 and 2.0 g per plant respectively, and inorganic fertilizers of Urea and SP-36 were applied at rate of 0.1 g and 0.2 g per plant, consecutively. The nutrient sources were buried 5 cm around the plant at depth of 7 cm. Weeding was done manually while watering plants in the afternoon. Plnat protection was done by applying organic pesticide 0.5% Azadirachtin (OrgaNeem^{TM}) every three days. The plants were harvested 100 das.

Parameters Observation

Parameters dealing with soil fertility status (N, P, K, organic-C and soil pH) were measured before sowing and at 60 and 100 das. The agronomic parameters such as: top and roots dry weight biomass (60 das and 100 das), nutrients uptake (N, P, K, and Ca) at 60 das and component yield of soybean (cops, grain and 100 grain weight). The dry weight of the agronomic parameters were measured after being oven dried at 60 ^{o}C for 48 hours. Parameters related to AM activities including fungi population, roots percentage infections at 60 das were also measured. Plant analysis for N was determined using Kjeldhal method, P using spectro-photometer, C-organic with colorimetric method according to Walkley and Black, K and Ca was recorded using Automatic Absorbtion Spectro-photometer (AAS).

Soil, Plant Analysis and Mycorrhiza Observation

Analyses for N, P, and organic-C were done by using Kjeldhal method, spectrophoto-meter, and colorimetric method according to Walkley and Black, respectively. K and Ca were analyzed by using AAS. Mycorrhiza population was observed using wet sieving technique according to Brundrett *et al.* (1996). The supernatan caught at 38 µm-sieve was added with 60% of sucrosa solution and subsequently centrifused at 3000 rpm for 10 minute (Daniel and Skipper, 1982). The harvested spore were stored on the Whatman paper with permanent ink marked of 0.5 x 0.5 cm. Counting of mycoriza population was done using stereo mikroscope (extended 40 x). Calculation of roots percentage infections was conducted using modification of clearing and staining method (Kormanik and McGraw, 1982), counted with the *Gridline Intersect* technique (Giovenneti and Mosse, 1980) under stereo-microcope observation.

Statistical analysis

To know if there is any difference among treatments, a mathematic model below was applied:

$$X_{ijk} = \mu + \rho_i + \beta_j + \varepsilon_{ij}$$

Remarks:

μ = general average
ρ_i = effect of replication-i
β_j = effect of treatment-j
ε_{ij} = error

The model was applied when $X_{ijk} = \mu + \rho_i + \beta_j$ linear and additive to $\Sigma\rho_i = \Sigma\beta_j = 0$, and ε_{ijk} was free and normally distributed with average and variance = $(0,\sigma^2)$. Data were analysed by analysis of variance (ANOVA-MStat) and the effect of treatments was determined. When the variance ratio (F) was significant, means for each treatment were separated using a least significant difference test at 5% level.

RESULTS AND DISCUSSION

Soil Chemical Properties

In general, compared with control, all treatments significantly increased soil fertility status as indicated by concentration of total N, available P and K, and organic-C both at 60 das and 100 das (Table 1). The highest increases of soil fertility status were observed in sandy soil inoculated with arbuscular mycorrhiza (AM) and amended with cattle manure (CM). This indicated that AM performed well when combined with CM compared with other combinations. Compared with control, inoculation of AM amended with CM increased N, P, K, and org-C as much as 39%, 105%, 27%, and 85%, respectively on 60 das and 12%, 60%, 10%, and 11%, consecutively on 100 das at the first cropping cycle. Interestingly, these effects were also observed at the second cropping cycle as the total N, P, K, and org-C increased as much as 43%, 120%, 37%, and 36%, respectively on 60 das, and 44%, 53%, 14%, and 36%, respectively on 100 das. Data presented in Table 1 also indicated that at the second cropping cycle soybean performance on treatment of AM plus CM could improve soil fertility status as shown by nutrient concentration of P and N as much as 4 and 48% on 60 das, respectively, and on 100 das the treatment increased N and Org-C as much as 12 and 20%, consecutively.

Table 1. Soil fertility status (N, P, K, organic-C and soil pH) of sandy soil with various treatments after harvesting

Treatments	N (g kg^{-1})		P (mg kg^{-1})		K (cmol kg^{-1})		org-C (g kg^{-1})		pH	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
60 DAS										
F_0	1.15 ^{a}	0.87 ^{a}	16.97 ^{a}	16.54 ^{a}	0.69 ^{a}	0.51 ^{a}	12.1 ^{a}	24.5 ^{a}	6.25 ^{a}	6.13 ^{a}
F_1	1.34 ^{b}	1.15 ^{b}	23.60 ^{b}	23.40 ^{b}	0.75 ^{b}	0.58 ^{b}	17.9 ^{b}	26.5 ^{b}	6.01 ^{b}	6.24 ^{b}
F_2	1.60 ^{c}	1.25 ^{b}	34.83 ^{c}	36.54 ^{c}	0.88 ^{c}	0.70 ^{c}	22.5 ^{c}	33.5 ^{c}	5.95 ^{c}	6.32 ^{c}
F_3	1.44 ^{d}	1.10 ^{b}	26.34 ^{d}	27.01 ^{d}	0.83 ^{d}	0.63 ^{d}	21.0 ^{d}	29.4 ^{d}	6.72 ^{d}	6.28 ^{d}
F_4	1.40 ^{e}	0.93 ^{b}	20.59 ^{d}	30.97 ^{e}	0.78 ^{c}	0.62 ^{bd}	19.1 ^{e}	26.3 ^{b}	6.08 ^{e}	6.26 ^{bd}
100 DAS										
F_0	1.31 ^{a}	1.14 ^{a}	17.62 ^{a}	17.33 ^{a}	0.75 ^{a}	0.63 ^{a}	23.8 ^{a}	23.5 ^{a}	6.18 ^{a}	6.54 ^{a}
F_1	1.44 ^{b}	1.36 ^{b}	20.86 ^{b}	22.68 ^{bc}	0.77 ^{b}	0.65 ^{b}	24.5 ^{b}	25.3 ^{ab}	6.21 ^{b}	6.62 ^{ab}
F_2	1.47 ^{c}	1.65 ^{c}	28.25 ^{c}	26.57 ^{d}	0.83 ^{c}	0.72 ^{c}	26.6 ^{c}	32.1 ^{c}	6.24 ^{c}	6.54 ^{ab}
F_3	1.38 ^{d}	1.41 ^{b}	23.32 ^{d}	20.62 ^{c}	0.81 ^{d}	0.67 ^{b}	23.9 ^{d}	28.0 ^{d}	6.62 ^{d}	6.75 ^{a}
F_4	1.33 ^{a}	1.43 ^{b}	24.38 ^{e}	24.57 ^{bd}	0.82 ^{e}	0.70 ^{c}	25.7 ^{e}	27.5 ^{d}	6.23 ^{c}	6.44 ^{b}
Before exp[1)]	1.10	-	13.82	-	0.57	-	12.1	-	6.25	-

Remarks: Means followed by the same letters within the same column are not significantly different (p=0.05); 1st and 2nd (first cycles and second cyles); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF; [1)] pre-treatment data

Results of this study were in accordance with those of done by Jeffries *et al*. (2003) and Gianinazzi *et al*. (2010) reporting that AM inoculation with organic matter amendment can recover soil fertility status on an ecosystem. Furthermore, Douds *et al*. (2006) reported that introduction AM inoculation combined with various nutrient sorces into sandy soil could increase soil nutrient content. The same result was also reported by Astiko *et al*. (2013) evaluating contribution of indigenous AM combined with cattle manure to increase corn performance in sandy soil of northern Lombok. Combination of AM and cattle manure resulted in significant improvement on soil fertility status especially N, P, K, and organic-C. This increase was suggested due to activity of enzyme present in external hypha of AM in the rhizosphere able to catalize and hydrolyze unavailable nutrient complex into available nutrients (Widiastuti *et al.*, 2003).

Previous results reported by Warnock *et al.*, (2007) showed that enrichment of AM could be escalated by the addition of organic matter and the combination has a positive effect on improving soil fertility status and therefore it is beneficial for soybean performance. This signify that better sinergy of the inoculated AM-tretment in combination with CM, as previously confirmerd by Kato and Miura (2008). Higher soil extractable-P found in this study was not only due to indirect contribution of CM in improving soil fertility status but also related to a positive contribution of mycorrhiza in producing phosphatase enzyme, for mineralization of organic-P in soil (Crowley and Rengel, 2000; Joner and Johansen, 2000) and resulted in enhancing insoluble-P in soils (Orcut and Nilsen, 2000).

Nutrient Uptake, Growth and Yield

Nutrient absorption by soybean is presented in Table 2. In general, treatments such AM inoculation alone or combined with other sources of nutrients significantly increased nutrient aupatke by soybean, compared with control. The highest increases were observed at treatment AM inoculation amended with CM. At this treatment, absorption of N, P, K, and Ca increased as much as 214%, 185%, 342%, and 233%, respectively on the first cropping cycle, and 191%, 308%, 224%, and 413%, consecutively on the second cropping series, compared with control. Treatment of AM inoculation followed by CM amendment also caused high absorption of P and Ca as much as 29% and 126%, respectively on the second cropping cycle. This indicated an increase in soybean performance when grown in sandy soil amended with various nutrient sources.

Table 2. Nutrient uptake (N,P, K and Ca) by soybean grown in sandy soil with various treatments.

Treatments	Nutrient uptake (mg.plant^{-1})							
	First cycle				Second cycle			
	N	P	K	Ca	N	P	K	Ca
60 DAS								
F_0	221.59 ^{a}	22.43 ^{a}	121.53 ^{a}	25,60 ^{a}	224.52 ^{a}	20.37 ^{a}	79.21 ^{a}	37.66 ^{a}
F_1	302.83 ^{b}	29.03 ^{b}	210.13 ^{b}	34,16 ^{b}	301.13 ^{b}	34.46 ^{b}	121.63 ^{ab}	59.22 ^{b}
F_2	697.16 ^{c}	64.06 ^{c}	537.33 ^{c}	85,30 ^{c}	653.42 ^{c}	83.22 ^{c}	257.43 ^{c}	.93.30 ^{c}
F_3	412.73 ^{d}	39.55 ^{d}	282.73 ^{d}	48,20 ^{d}	429.60 ^{d}	49.33 ^{d}	185.20 ^{d}	12.96 ^{d}
F_4	344.10 ^{e}	34.46 ^{e}	232.46 ^{e}	39,96 ^{d}	311.60 ^{b}	38.14 ^{b}	159.44 ^{ad}	96.93 ^{d}

Remarks: Means followed by the same letters within the same column are not significantly different ($p=0.05$); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF

The increase of P absorption could possibly cause such a new nutrient balance in plant that induced absorption of other nutrients such as N, K, and Ca. Sufficient availability of K created a condition in which the use of water was efficient as cell turgor was maintained. This condition leads to active metabolism process, K accumulation on the tips, buds, and roots, the accumulation that lead to formation of cortex tissue and cell elongation leading to improvement of soybean performance (Schweiger *et al.*, 2007 and Smith *et al.*, 2010). This result was in accordance with that of Kaschuk *et al*. (2010) stating that the increase of nutrient abrorption by host plant lead to plant vitality to supply Carbon to rhizosphere to form AM external hypha. Mathur and Vyas (2000) stated that AM inoculation was also resulted in the increase of accumulation of amino acids, protein, chlorophyll, and sugar contents compared with non-AM plants. N status of shoot tip of plant with mycorrhiza at extreme condition was higher than that of plant ithout mycorrhiza (Subramanian and Charest, 1999).The same trends were also recorded for N, P, K, and Ca (Liu *et al*., 2000).

The inoculation of AM followed by CM also increased soybean performance, as observed on plant dry biomass measured by root and shoot dry weights on 60 and 100 das (Table 3). Dry weight of roots and shoots on 60 das increased as much as 164% and 136%, respectively; while on 100 das the increases were as much as 150% and 178%, respectively on the first cropping cycle. On the second cycle, on 60 das the increases were 337% and 718%, while on 100 das the increases were as much as 390% and 1102%, consecutively.

The improvement of biomass was probably due to AM role to influence soil fertility status, especially P. Sufficient availability of P can indirectly induce absorption of other nutrients leading to better plant growth (Carrenho *et al*., 2001). This positive effect was suggested due to suitability of AM type, plant, and soluble P (Nikolaou *et al*., 2002; Bhadalung *et al*., 2005).

This condition was due to good association between plant and AM to perform maximal acrtivity. While AM received carbon from the plant, and the latter got P from the first. It was reported that each particular combination of AM and plant showed such a specific carbon translocation pattern that influenced production of plant biomass (Smith and Read, 2008; Smith *et al*., 2009). Inefficient symbiosis in use of carbon could decrease plant biomass. AM colonization could result in positive, neutral, or negative impact depending on AM types, plants, and growth environment (Johnson *et al*., 1997; Hoeksema *et al*., 2010).

Negative impact of mycorrhiza colonization on initial growth of plant was reported previously (Bethlenfalvay *et al*., 1982; Koide 1985; Johnson *et al*., 1997). Such sort of impact may be due to various factors, such as; high availability of P in soil (Mosse *et al*., 1973), competition for carbon between plant and AM in low light intensity condition (Buwalda and Goh 1982), and difference in biomass allocation pattern between plant with and without mycorrhiza (Smith and Read, 2008).

Table 3. Root and shoot dry weight of soybean grown in sandy soil with various treatments at 60 and 100 das

Treatments	Biomass dry weight (g.plant^{-1})			
	First cycle		Second cycle	
	Roots	Shoots	Roots	Shoots
60 DAS				
F_0	1.88 ^{a}	11.11^{a}	1.74 ^{a}	8.50 ^{a}
F_1	2.66 ^{b}	14.51 ^{b}	2.27 ^{b}	10.48 ^{b}
F_2	4.97 ^{c}	26.28 ^{c}	4.38 ^{c}	18.67 ^{c}
F_3	3.62 ^{d}	20.39 ^{d}	3.75 ^{d}	13.51 ^{d}
F_4	3.34 ^{d}	17.10 ^{e}	2.51 ^{b}	11.41 ^{e}
100 DAS				
F_0	3.41 ^{a}	22.43 ^{a}	2.83 ^{a}	19.94 ^{a}
F_1	4.45 ^{b}	39.63 ^{b}	2.54 ^{a}	19.35 ^{a}
F_2	8.53 ^{c}	62.45 ^{c}	6.27 ^{b}	32.92 ^{b}
F_3	5.22 ^{d}	44.50 ^{d}	3.41 ^{a}	26.40 ^{c}
F_4	4.48 ^{b}	37.85 ^{b}	2.36 ^{a}	19.91 ^{c}

Remarks: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control, F_1= AM inoculation, F_2= AM inoculation plus CM, F_3= AM inoculation plus RP, and F_4= AM inoculation plus CF

The use of low solubility of phosphate, like phosphate stone, although with high dosage, in fact, was still effective to support AM and to increase plant performance (Nikolaou *et al*., 2002). In addition, quite high dependency of plant on AM indicated that in early stage of its growth, plant needs to associate with AM. Table 3 also showed significant effect of AM inoculation combined with CM as indicated by the increase plant performance as observed on root and shoot dry weight. The similar results were also reported previously (Rochdjatun *et al*., 2011; Astiko *et al*., 2012).

Inoculation of AM with CM amendment increased plant performance as observed on cobs weight, weight of grain, and weight of 100 grains as much as 180%, 163%, and 139% respectively on the first cropping cycle and as much as 139%, 330%, and 23%, consecutively on the second cycle (Figure 1). These results were in accordance with results of previous studies indicating that fertilizer package with AM inoculation and amended with CM increased soil fertility status, plant performance and yield (Astiko, 2009; Viti *et al.*, 2010). Similar results were also reported on dry paddy (Kabirun, 2002).

The increases of soil fertility status, plant performance and yield on the tretmant of AM inoculation amended with CM were caused by the increase of AM activity in absorption of nutrient and water through its external hypha (EH). This was possibly due to EH can reach depletion zone that cannot be reached by plant roots (Zhu *et al*., 2001). The diameter of EH which is much smaller than that of roots makes the EH possibly to penetrate soil micro pores to take nutrient and water that cannor be reached by roots (Drew *et al*., 2003).

This ability causes plants with mycorrhiza be able to absorb nutrient, growth and perform better and resistance to dought stress (Smith and Read, 2008). In addition, AM is able to dilute phosphate tied in soil and fertilizer, to improve absorption of N, P, and K, to improve plant tolerance to drought, to improve plant ability to produce growth regulator, to stimulate activity of beneficial microbes, to improve soil structure and aggregation, and to enhance mineral cyclic (Cruz, 1990).

Decomposition and mineralization of organic matter were better with the presence of AM which positively affect physic, chemistry, and biology factors of the soil which in turn play key role in improving plant yield (Smith and Read, 2008). All above facts indicated such a suitable functional among AM, host plants, and environment that are able to increase nutrient absorbtion, plant growth and yield as also reported earlier (Burleight *et al*., 2002).

Mycorrhiza Activity

The inoculation of AM followed by CM amendment could increase AM activity as shown by numbers of spores per 100 g of soil and percentage of root infection (Tabel 4). The number of spores and percentage of infection on the first cropping cycle 60 das on sandy soil inoculated by AM and amended with CM increased as much as 179% and 266%, respectively, while on the second cycle the increases were 24% and 160%, respectively compared with control and were significantly different from other treatments.

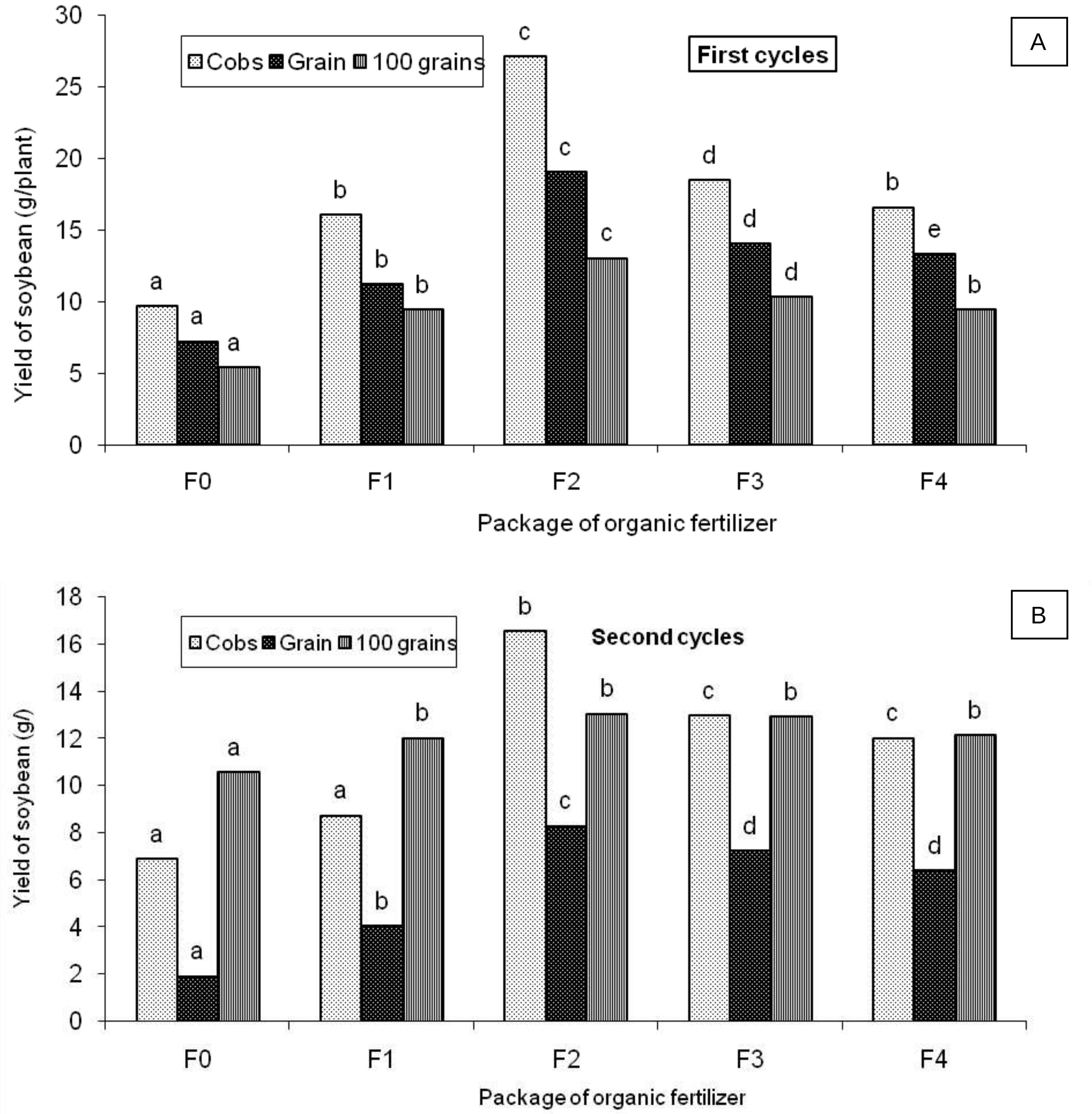

Figure 1. Yields of soybean (cobs, grain and 100 grain dry weight) grown in sandy soil with various treatments. A. the first cropping cycle, and B. the second cropping cycle. Bars with the same letters at the same category are not significantly different (p=0.05).

Table 4 showed that the number of spores and root infection were significantly high in all treatments when compared with control. Compared with before sowing. the number of spores and root infection 60 das increased significantly. This increases indicated that isolate M_{AA1} were able to compete with indigenuous AM present in the rhizosphere of sandy soil Northern Lombok. particularly in colony forming inside roots. The other point that can be taken from these facts was that the isolate M_{AA1} used was able to produce abundance propagules in the form of spores and such colonized roots that they were able to live on dynamic and competitive habitat (Barrios, 2007; Doud and Johnson, 2007; Gianinazzi *et al*., 2010). Result of a research on the role of indigenous mycorrhiza combined with cattle manure in improving yield of maize (*Zea mays* L.) on sandy loam of Norhern Lombok showed that inoculation of AM combined with cattle manure resulted in higher number of spores and infected roots both in the first and the second growth season (Astiko *et al*., 2013). Similar results were also shown by Prasetya and Anderson (2011) on the assessment of the effect of long term tillage on the arbuscular mycorrhiza colonitation of vegetable crops grown in andisols. This fact also indicated that isolate M_{AA1} showed high effectiveness. although they were inoculated on unsterile soil. Similar research on soybean inoculated by AM and the application of organic leaf fertilizer “greenstant” also showed similar results (Wangiyana *et al.*. 2007).

Table 4. Biological activity of mycorrhiza (number of spores and percentage of infections) in sandy soil with various treatments.

Treatments	Spores 100 g soil^{-1} and root infection			
	First cycle		Second cycle	
	Spores	Infection	Spores	Infection
60 DAS				
F_0	1.060^a	21^a	3.122^a	25^a
F_1	2.159^b	41^b	3.533^b	34^b
F_2	2.960^c	77^c	3.878^c	65^c
F_3	2.343^d	54^d	3.781^d	51^d
F_4	2.215^b	46^e	3.693^e	41^e
Before exp$^{1)}$	371	-	-	-

Remarks: Means followed by the same letters within the same column are not significantly different (p=0.05); F_0= Control. F_1= AM inoculation. F_2= AM inoculation plus CM. F_3= AM inoculation plus RP. and F_4= AM inoculation plus CF; $^{1)}$ pre-treatment data

CONCLUSION

Inoculation of AM amended with cattle manure improved sandy soil fertility as shown by the increasing soil fertility status as observed on concentrations of N. P. K. and organic-C.

Soybean performance responded positively to the application of AM followed by cattle manure as indicated by plant improvement in nutrient absorption. plant growth and yield. The addition of cattle manure stimulated activity of AM leading to improvement of soil fertility and plant performance.

ACKNOWLEDGEMENTS

This manuscript is a part of the first authors dissertation for the Doctoral Program funded by Post Graduate Scholarships at Agriculural Faculty University of Brawijaya Malang. Thank to the institution of Directorate General of Higher Education (DGHE) as the funders of Ph.D research for 2012 Grant Program by DIPA Number: 023.04.2.415278/2013. December 5. 2012. Letter of Agreement. Impelemtation Research Number: 295.G/SPP-APDD/H18.12/PL/ 2013. May 2. 2013.

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