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Soil Chemical and Biological Characteristics for Diagnostic the Potency of Acid Dry Land for Soybean Extensification

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ABSTRACT

The descriptive explored study was conducted in districts Bumi Nabung, Sari Bakti, Seputih Banyak and Rumbia, Central Lampung, Indonesia. The parameters observed consisted of chemical and biological aspects of soil, which directly affected plant growth. This activity was carried out as preliminary studies for the diagnosis of soybean extensification on acid dry land. The results showed that all locations observed were less suitable for soybean development, indicated by low pH values (4.35 – 6.00), nutrient contents (N < 0.1% and C-organic < 2.0%), and low soil microbial populations. Population of bacteria was $17 \times 10^3 – 29 \times 10^4$ CFU g^{-1} soil and fungi was $21 \times 10^1 – 63 \times 10^2$ CFU g^{-1} soil of soils. Beneficial microbe types included non-symbiotic nitrogen-fixing bacteria (with the capability to fix the Nitrogen around 0.16 – 1.53 mM 100 ml^{-1} medium h^{-1}), phosphate solubilizing bacteria (with the value index 1.22 – 6.25) and arbuscular mycorrhizal fungi (with root colonization by 70.50 – 90.33% and the number of spores were 49 – 175 spores g^{-1} soil). This less suitable land can be improved to become suitable for developing soybean by using innovative technology. Soil biological and chemical improvement technology through liming and amelioration as well as organic and bio-fertilizers applications were required for soybeans extensification on acid dry land.

Keywords: Soil biology, soil chemsitry, soybean, Ultisols

INTRODUCTION

The acid dry land has a great potential to be expanded as the effort for planting soybeans. Sudaryono *et al*. (2007) reported that an identification of acid dry land issues included: limited water availability, acidic pH, sensitively high erosion, fertilization and amelioration not appropriate, nutrient leaching and toxicity of Al, low levels of soil organic matter, low fertilization efficiency, and degradation of productivity. Therefore, the obstacles encountered in the alternative land use and management will be different.

Management of acid dry land is required to improve crop productivity optimally. Acid dry land management strategies can be done through a high input by liming and fertilizing P or low input by addition of organic manure, crop rotation, legumes and alley cropping, integrated management, increasing nutrient efficiency, organic-inorganic fertilizer combination (Vestberg *et al*. 2002). The biological management of acid dry land is surely being interesting to achieve sustainable agricultural systems. Soil microbes play an important role in nutrient cycling in dry land agro-ecosystems (Sugihara *et al*. 2010). Generally, land management affects soil microbial dynamics (Chikowo *et al*. 2006; Spedding *et al*. 2004). Technique to improve biologically status of soil fertility can be approached by inventory of the types and dominancy of soil microbial. The early study of this problem was approached by conducting a diagnostic study of acid dry land biologically. The microbiologically interaction of soil and plant root systems affect the success of plant growth.

The objectives of this research were to study soil biology and biochemistry in relation to assess beneficial of soil microorganism and soil chemical properties of acid dry land for soybean crop.

MATERIALS AND METHODS

Study Sites

This research was descriptive exploratory emphasis on biological and chemical soil properties of acid dry land. Research was conducted from March to September 2010 in the district of Bumi Nabung, Sari Bakti, Seputih Banyak, and Rumbia Distric in Central Lampung Regency (104°35’ – 105°50’E, 0.4°30’ –0.5°15’S).

Soil Sampling and Analysis

The biological components observed in this study included productivity cultivated crops and the soil microbial population. Characterization of soil microbial was done by soil sampling at a depth of 0 – 30 cm from the soil surface with five replications. Soil microbial population was counted by a total plate count method, and further by the microbial screening on difference in shape, color and size of the colony. Isolation of microbial was carried by Nitrogen Fixation Bacterial (NFB) selective medium for symbiotic nitrogen-fixing bacterial presented by Vincent (1970) with compositions of KH_2PO_4 0.4 g; MgSO4.7H_2O 0.2 g; NaCl 0.1 g; $CaCl_2$.2H_2O 0.026 g; $FeCl_3$.6H_2O 0.017 g, Na_2MoO_4.2H_2O 2 mg; DL-malic acid 3.58 g; bromothymolblue 0.025 g; agar 1.75 g, and distilled water to 1 L. Phosphate solubilizing microbial was isolated by Pikovskaya medium with compositions of glucose 10.0 g, $Ca_3(PO_4)_2$ 5.0 g, MgSO4.7H_2O 0.1 g, KCl 0.2 g, $FeSO_4$ 0.01 g, $MnSO_4$ 0.01 g, (NH_4).2 SO_2 0.5 g, yeast extract 0.5 g, agar 2% and distilled water to 1 L (Nautiyal 1999).

The ability of nitrogen fixation was calculated by determining nitrogen activity using gas chromatography (Welsh *et al*. 2007), while the ability of phosphate solubilizing was determined by measuring the clear zone area formed at Pikovskaya medium enriched with rock phosphate, $Ca_3(PO_4)_2$ or $AlPO_4$ (Mikanova 2002). Determination the level of root colonization by vesicular arbuscular mycorrhizal was determined by clearing and staining method (Phillips and Hayman 1970), while the observations and calculations of mycorrhizal spores were determined by wet sieving and decanting method (Gardermann 1975). Analysis of chemical soil properties included pH, moisture content, N, C, P, K, Al, K, Na, Ca, Mg, Al, and CEC.

RESULTS AND DISCUSSION

Chemically Diagnostic of Acid Dry Land Soil

Soil is a medium for plants and soil biota growth. Plant growth or soil microbial population dynamics were determined by soil properties in which as growth environment (Cao *et al* 2008). Soil chemical properties on acid dryland were pH value 4.35 – 6.0, N and organic-C contents were low < 0.1 g kg^{-1} and < 2.0 g kg^{-1}, respectively. Soil CEC values were ranged from 8.25 to 29.1 cmol kg^{-1} and P_2O_5 contents of the soil were varied from 4.28 to 172 mg kg^{-1} (Table 1).

The acid dry land used in this study was classified as less fertile soil in order to support the soybean growth (Table 1). Therefore, expanding soybean in this land needed improvement technology in soil biology and biochemistry. Nutrient contents in acid dry land were generally low by intensively leaching, low organic matter which were caused by the decomposition process was running fast and partially carried by erosion. Argillic horizon generally contained a high Al, in which it was sensitive to plant roots development and it could not penetrate this horizon and only grew in the upper of the argillic horizon (West *et al*. 1997).

Theoretically, soybean crop had grown widely adaptable to soil, especially in fertile soil with the optimum pH 6.2 – 7.0, Al saturation < 20% and for every 1,000 kg weight of soybean seed, the nutrient transport was more or less than 66 kg N, 15.5 kg P, 39.7 kg of K, 7.5 kg of Mg and 7.0 kg of S (Halliday and Trenkel 1992). Criterias of land suitability for

Table 1. Soil chemical properties in the four locations at Central Lampung.

Parameters	District			
	Bumi Nabung	Sari Bakti	Seputih Banyak	Rumbia
pH H_2O	6.00	4.80	5.15	4.35
Total-N (g kg^{-1})	0.08	0.07	0.05	0.10
Organic-C (g kg^{-1})	0.12	0.20	0.11	0.18
P_2O_5 (mg kg^{-1})	172	9.52	4.28	13.6
SO_4 (mg kg^{-1})	18.0	14.5	150	116
K (cmol kg^{-1})	0.06	0.05	0.03	0.12
Na (cmol kg^{-1})	0.09	0.05	0.05	0.17
Ca (cmol kg^{-1})	1.93	0.67	0.77	0.44
Mg (cmol kg^{-1})	0.57	0.49	0.54	0.37
CEC (cmol kg^{-1})	13.7	8.25	19.2	13.7
Al-exch (cmol kg^{-1})	0	1.66	2.17	2.43
H-exch (cmol kg^{-1})	0.10	0.20	0.11	0.06

Table 2. Criteria of land suitability for soybean plants.

Soil characteristics	Level of land suitability			
	S1 Very suitable	S2 Suitable	S3 Moderately suitable	N Not suitable
Score	4	3	2	1
Nutrient retention:				
CEC (cmol kg^{-1})	> 25	25 – 15	15 – 5	< 5
pH (H_2O)	6.0 – 7.0	7.1 – 7.5 5.9 – 5.5	7.6 – 8.5 5.4 – 5.0	> 8.5 < 5
Nutrient availability:				
N total (g kg^{-1})	> 1.0 – 0.5	0.5 – 0.2	0.2 – 0.1	< 0.1
P_2O_5 available (Bray 4)(mg kg^{-1})	> 50	50 – 15	< 15	< 5
P_2O_5 available (Olsen 3) (mg kg^{-1})	> 15	15 – 5	< 5	< 2
K available (cmol kg^{-1})	0.8 – 0.4	0.4 – 0.2	0.2 – 0.03	< 0.03
Al saturation (Al/CEC) %	< 20	20 – 30	30 – 40	> 40

Source: CSR-FAO (1983); Landon (1984).

soybeans production were divided into four categories: very suitable (S1), suitable (S2), less (moderate), suitable (S3) and not suitable (N). Table 2 shows the land suitability criteria for soybean from soil chemical characteristics. The acid dry land on all four study sites were in less suitable (moderate) categories for soybeans crop (Table 2).

Biologically Diagnostic of Acid Dry Land Soil

Plants grown at the study site were varied greatly, either soybeans, peanuts, sweet potatoes, cassava or maize. Based on interviews with the farmers, no targeted on crop productivity was caused by non-fertile land but high enough by pest attack. Weed plant species were relatively abundant especially the reeds and grasses puzzles. In general, farmers land in this area did not have too many shade trees, so it did not affect the growth of cultivated plants.

The morphology of the soybean roots in acid dry land and the number of root nodules were a quite low (Figure 1), indicating that Rhizobium bacteria were less able to grow and to form root nodules of soybean in acid dry land. Rhizobium was generally regarded as microbial symbiotic partners of legumes and mainly known for the role in the formation of nitrogen-fixing nodules (Antoun and

Figure 1. Morphology of the roots soybean plants on acid dry land.

Prevost 2005). The roots of soybean crops were observed to have root nodules, although in very small amounts (< 10 seeds) and small pieces (< 2 mm), that were pressured by acidic pH. The optimally growth of Rhizobium bacteria occured at a temperature of 25 – 30 °C and pH 6 – 7 (Ilyas *et al*. 2008).

Table 3. Soil microbial population on the acid dry land.

Soil samples location	Number of cells (CFU g^{-1} of soils)	
	Bacteria	Fungi
Bumi Nabung	13.3×10^4	29.4×10^1
Sari Bakti	42.0×10^3	41.4×10^1
Seputih Banyak	14.8×10^4	31.7×10^2
Rumbia	39.6×10^3	38.2×0^1

Table 3 showed that the microbial population on acid dry land was quite low, ranging between $57.10^3 – 29.10^4$ CFU g^{-1} of soils. On the fertile soils contained more than 100 million microbes per gram of soil (Alexander 1977). Types of microbes that were found on acid dry land consecutive according to the density level were a bacterial $17 \times 10^3 – 29 \times 10^4$ CFU g^{-1} and fungi $21 \times 10^1 – 63 \times 10^2$ CFU g^{-1} soil. Although the population of microbials in acid dry soils were low, but the presence of soil microbials that exist on this land are needed to be studied further about its activity and effectiveness and its tolerant in the natural growing environment.

Beneficial Microbe Isolated from Acid Dry Land Soil

Soil microorganisms are ubiquitous in nature and form a vital component from all known ecosystems on earth. Pandey *et al*. (2006) suggested that bacteria, actinomycetes and fungi are three major groups of soil inhabiting microorganism. The extent of the diversity of soil microorganism is seen to be critical to the maintenance of soil quality, as a wide range of microorganism is involved in important soil function (Garbeva *et al*. 2004).

Free living nitrogen-fixing bacteria (non-symbiotic) was isolated by pour plate and streak plate method on NFB selective medium (Figure 2). The medium color was able to change from green to blue indicated the activity of non-symbiotic N-fixing bacteria. The presence of non-symbiotic N-fixing bacteria allows for the supply of nitrogen to the plants through the process of nitrogen fixation from the air. N_2reduction process from air into ammonia is a reaction that requires high energy. Nitrogenase enzyme is owned by N_2 nitrogen fixation bacteria and sensitive to oxygen (Giller and Wilson 1991). This enzyme is also able to reduce acetylenetoethylene, so that the reduction reaction of acetylene can be used to detect the activity of nitrogenase (Bergersen 1980).

The results of gas chromatography analysis showed that the non-symbiotic nitrogen fixation bacterial activity were ranged 0.15 – 1.53 m*M* 100 ml^{-1} medium hr^{-1} (Table 4). Although these results could not be used to predict the activity of nitrogen fixation in the field, but at least was able to describe the activity of nitrogen fixation each isolate in a laboratory environment.

Phosphate solubilizing microbial from acid dry land was isolated on Pikovskaya selective medium, included free living bacterial and fungal (non-symbiotic), were characterized by the formation of clear zone around the colony (Figure 3).The activity of phosphate solubilizing bacterial and fungal can be shown from the value index (Table 5). The determination of value index reflected the differences in activity between isolates selected in which 1.22 – 6.25 for bacterial and 1.07 – 2.18 for fungal. Table 4 shows a more active bacterial was with P sources of $Ca_3(PO_4)_2$, while the fungal was more active with a source of P from phosphate rock. From the research it was found a variety of

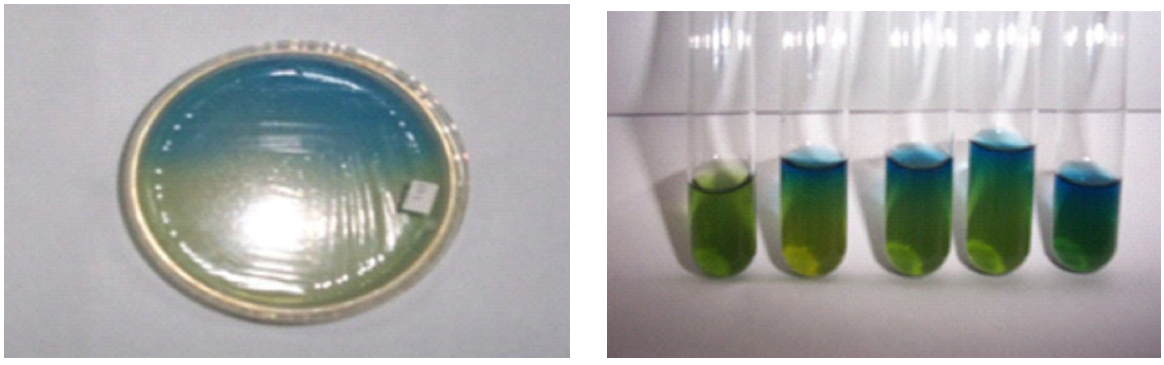

Figure 2. Isolates of non-symbiotic nitrogen-fixing bacterial on NFB medium.

Table 4. Non-symbiotic nitrogen-fixing bacterial activity from acid dry land Central Lampung.

Gram test[1]	Number of bacterial	Value of C_2H_4	Equivalent of N_2 fixation
		..... m*M* 100 ml^{-1} medium hr^{-1}	
+	8	0.58 – 6.10	0.15 – 1.53
–	5	0.62 – 2.44	0.16 – 0.61

Notes: [1]+ = Gram positive bacteria, and – = Gram negative bacteria.

phosphate solubilizing activity of three P sources different. Not all isolates were able to P solubilizing from the three sources of P, which showed that for the development of their utilization needed to be adjusted to isolate the source of P existed in the land. In addition to the considerations in determining the composition and concentration of microbial cells that will be applied.

There were16 bacterial isolates as follows: six isolates of Gram positive bacterial, phosphate solubilizing capable of P sources $Ca_3(PO_4)_2$ and rock phosphate with index value by 3.50 – 5.00 and 1.40 – 3.30; three Gram positive bacterial isolates, capable of activity with the source of P derived from $Ca_3(PO_4)_2$, $AlPO_4$, and rock phosphate by value index 2.75 – 6.25, 1.22 – 3.25 and 1.28 – 1.75, respectively; two isolates were Gram positive bacterial, capable of activity with the source of P from $Ca_3(PO_4)_2$ with the value index by 2.40 – 4.20; four isolates of Gram positive bacterial, capable of activity with the P sources originated from $Ca_3(PO_4)_2$ and $AlPO_4$ by value index 4.00 – 6.00 and 1.64 – 2.38; and one isolate was Gram negative bacteria, capable of activity with the source P from $AlPO_4$ with value index by 4.19.

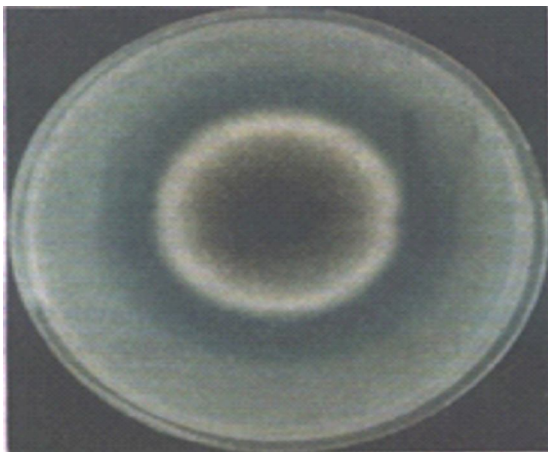

Figure 3. Phosphate solubilizing microbia growth on Pikovskaya medium.

There were 16 (sixteen) fungal isolates that as follows: seven isolates capable of P source from phosphate rock by value index 1.07–1.38; two isolates capable of P source from $Ca_3(PO_4)_2$ by the value index 1.31–2.18; three isolates capable of P source from $AlPO_4$ by value index1.22–2.18, respectively; two isolates capable with P source from $Ca_3(PO_4)_2$ and phosphate rock with value index each

Table 5. Properties of P solubilizing bacterial and fungal from acid dry land Central Lampung

Types of microbial	Number of species	Gram test[1]	The value index on Pikovskaya médium enriched by		
			$Ca_3(PO_4)_2$	$AlPO_4$	Rock phosphate
Bacterial	6	+	3.50 – 5.00	-	1.40 – 3.30
Bacterial	3	+	2.75 – 6.25	1.22 – 3.25	1.28 – 1.75
Bacterial	2	+	2.40 – 4.20	-	-
Bacterial	4	+	4.00 – 6.00	1.64 – 2.38	-
Bacteria	1	–	-	4.19	-
Fungal	7		-	-	1.07 – 1.38
Fungal	2		1.31 – 2.18	-	-
Fungal	3		-	1.22 – 2.18	-
Fungal	2		1.32 – 1.91	-	1.10 – 1.32
Fungal	2		-	1.26 – 1.75	1.50 – 1.69

Notes: [1]+ = Gram positive bacteria and – = Gram negative bacteria.

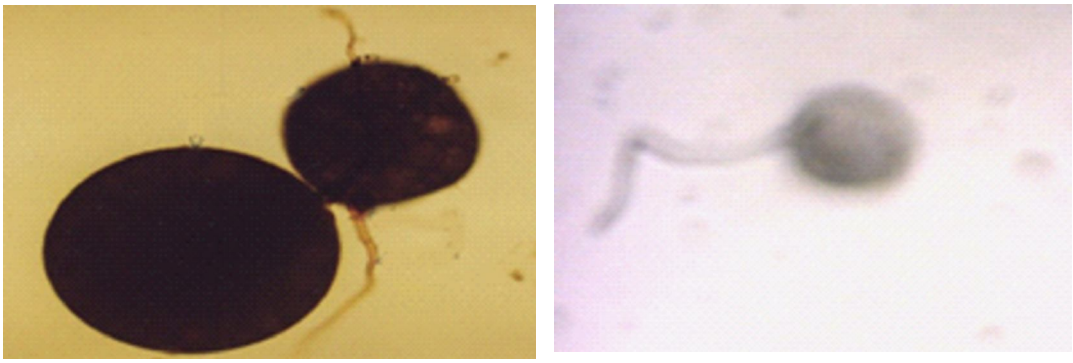

Figure 4. Spores of arbuscular mycorrhizal fungi that dominated on acid dry land.

1.32–1.91 and 1.10–1.32, and two isolates capable of activity with P source from phosphate rock and $AlPO_4$ by value index 1.26–1.75 and 1.50 –1.69, respectively.

The activity of phosphate solubilizing bacterial and fungal in the different P sources made the judgment in the utilization of beneficial microbe which was adapted to soil conditions. Types of bacterial was more active with P source from $Ca_3(PO_4)_2$, while the fungal was more active in P source from rock phosphate.

Spore of mycorrhizae was isolated by filtration multilevel (Sellose 2006). At least eight types of arbuscular mycorrhizal fungi (AMF) spore forming on acid dry land, which were dominated by species *Gigasporamargarita* and *Glomusmoseae* (Figure 4).

All the root preparations observed were positive to AMF colonization. The calculated number of AMF spore known the distribution, so there was uneven distribution. Table 5 shows the results of the level of root colonization by AMF and the amount of spore on the rhizosphere. The level of root colonization by AMF was quite high (averaging over 70%). Arbuscular mycorrhizal fungi were known to grow and thrive in an environment less favorable for the growth of other soil microbial (Daleo *et al*. 2007). The mutualistic association between the mycorrhizal fungi and plants provides the fungus with a renewable food source in return for increasing the surface area of the plants roots to allow more efficient absorption of water and mineral nutrient from the soil (Pozo *et al*. 2002). These associations are particularly beneficial to plants in nutrient poor soils (Weir 2007).

The root colonization level by AMF was high enough to give an indication of the ability of mycorrhizal on acid dry land in the infected plant roots. Generally, the crops studied had the positive response to AMF colonization and this condition might be a consideration to circumstence AMF benefit to be used on the land. Soybean crop is one type of plant that responses to AMF colonization (Hacisalihoglu *et al*. 2005). The root system colonization plants are expected to cope with soil acidity stress conditions (Moyer-Henry 2005). The presence of external fungal mycelium activities that explore the soil outside the root zone depletion can accelerate the diffusion of nutrients from the soil into the roots, so that the uptake of P increased (Welsh *et al*. 2010).

Prospects of Acid Dry Land Management for Soybean Development

The knowledge of processes that regulate nutrient release and plant uptake in soil is an essential prerequisite for sustainable agricultural management (Jia *et al*. 2010). It is essential to appreciate that in soil most nutrients are bound in organic form and the amount of nutrient available for plant growth depends on complex interactions between plant roots and soil microorganism (Coleman and Whitman 2005).

Fitter *et al*. (2005) suggested that soils were home to an extraordinary range of microbial. Biological activities in soils drive many of the key ecosystem processes, especially in the cycling of element such as carbon. nitrogen and phosphorus. The great biodiversity and abundance of soil microorganism and plant seems likely to influence ecosystem function in various ways (Joseph *et al*. 2003). The activity of the microbial biomass is commonly used to characterize the microbiological status of soil and to determine the effect of cultivation and field management on soil microorganism (Logah *et al*. 2010).

The acid dry lands are characterized by acidic pH value and low nutrient content makes the cation exchange capacity on the acid dry land are generally low(< 15 cmol kg^{-1}). Increase in CEC values can generally be done through increasing levels of soil organic C or use of land ameliorant (dolomite and zeolite). Levels of base cations (K, Ca, Mg and Na) on the acid dry lands of Central Lampung were low. thus requiring nutrient additional through fertilization and soil ameliorant. Thus, technical measurement of the soil amelioration would be part of the standard technology components to reduce the Al and Fe activity in acid dry land. The use of agricultural lime in the form of $CaCO_3$, dolomite, and organic material

Table 6. Average of arbuscular mycorrhizal fungi colonization level in the plant root system.

Crop comodity	Root colonization (%)	Number of AMF spore g^{-1} soils
Soybean	74.49 ± 16.00	175.25 ± 86.53
Peanuts	70.50 ± 28.18	61.75 ± 26.77
Maize	79.45 ± 10.57	99.50 ± 48.15
Cassava	90.33 ± 3.47	48.50 ± 32.80
Sweet Potato	79.17 ± 14.05	125.33 ± 110.95
Grassland	54.78 ± 5.52	114.00 ± 35.80

to improve land productivity has long been recommended and done (Mengel *et al*. 1987). Liming can be effective if the saturation acidity (Al + H) > 10% and soil pH < 5 (Wade *et al*. 1986).

The use of organic fertilizer is a standard component in soybean cultivation in acid dry land to increase levels of soil organic matter, soil moisture holding capacity, cation exchange capacity (CEC) and amount of soil organic colloids can be constraint resulting from Al and Fe compounds. There is needed to control acidity and plant poisoning by Al and Fe. The addition of P and K are needed to increase the availability of P and K on acid dry land which are generally low.

The neutralizing action of soil pH is through liming and amelioration. Then indirectly provide a better growing environment for soil microbes, especially the Rhizobium who initially present in an amount sufficiently low. It is known that the neutralization of soil with calcium hydroxide or potassium carbonate to restore the conditions become more favorable for the proliferation of Rhizobium (Niemi *et al*. 2008).

The results of diagnostic studies on the biological of acid dry land in Central Lampung Regency showed the soil microbial population was quite low, but species diversity was quite high especially the beneficial microbes. Soil microbial populations on acid dry land contained bacterial in an average of $17 \times 10^3 – 29 \times 10^4$ CFU g^{-1} of soil and fungal $21.10^1 – 63 \times 10^2$ CFU g^{-1} of soil. The types of beneficial microbe found in acid dry land included non-symbiotic N-fixing bacteria, AMF, and phosphate solubilizing microbia. Nitrogen fixation activity of non-symbiotic bacterial reached 0.15 – 1.53 m*M* 100 ml^{-1} medium hr^{-1}, the activity of phosphate solubilizing bacterial and fungal, respectively, reached 2.40 – 6.25 and 1.31 – 2.18 to the P source from $Ca_3(PO_4)_2$. 1.22 – 4.19 and 1.75 – 2.18 to the P source from $AlPO_4$. and 1.28 – 3.30 and 1.07 – 1.69 to the P source from rock phosphate. The level of mycorrhizal colonization in plant roots in acid dry land reached 70.50 – 90.33% and the number of spore 49 – 175 spores g^{-1} of soil were high enough.

The existence of the abundance of microbial populations in acid dry land was low enough, then the absolute biological management efforts required input microbial cultures. The process of inoculation of microbes that are useful for plant growth are intended to enrich the population and are resistant to natural microbial populations, and can move in the supply of nutrients (Parham *et al*. 2003). The microbial inoculation that was applied to the acid dry land must be able to grow and move in condition. A good relationship synergism between certain types of microbial and plants will have a positive impact on crop yields. The nutrient efficiency management in cropping system could lead to build up of microbial biomass overtime.

CONCLUSIONS

The acid dry land in the district of Bumi Nabung, Sari Bakti, Seputih Banyak, and Rumbia Central Lampung regency was classified as less suitable for soybeans crops. In an effort to realize a sustainable farming system, the expansion of soybean acreage planted in acid dry land required an increase of soil microbial populations, particularly of the type of beneficial microbe. This activity can be done through technology inoculation/input microbes which are beneficial for improving acid dry land productivity through maintenance technology and utilization of natural microbes to be developed in acid dry land grows by creating the appropriate environment. The success of these activities, in addition, to provide the elements of nitrogen and phosphate to the plants biologically as well as, to establish a stable cycle of these nutrients naturally.

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