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Formulation of Liquid Biofertilizer for Enhance of Soil Nutrients in Peatland

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Abstract: The main problem of peatland utilization was low fertility of land. The plant growth promoting microorganisms thrive in soils naturally, but their populations are scarce. *Optimation and implementation of fertilizer based on beneficial microbes having potential to* enhance the solubility of soil nutrients and to increase of yield crop. This study aims were to examine: 1) the effectiveness of liquid media for enchance of biofertilizer potency; 2) composition of liquid biofertilizer which potential for enchance of soil nutrients in peatland. This research was an experimental research in a field. Independent variables consist of: 8 compositions of liquid biofertilizer, included positive control (EM4), and negative control (aquadest), with 3 replication of each. Dependent variables were soil nutriens, namely: N, P, K,C. Soil nutriens measured following method of Soil Analysis procedure, using AAS. All data were analysed using ANOVA statistical method. The results showed: liquid media composed by 15% of sugar and 85% of coconut water, better more to support of biofertilizer potency for enhanced of soybean growth in peatland. Composition microbes KHY +IBT better more for increase of pottasium (K-dd) content and N-Total in peatland, compared than another compositions. Treatment with liquid biofertilizer from local microorganisms, had increased of soil nutrients N-total 69,7%, phosphate 4,7%, and potassium 28%, compared with negative control. Composition of potential microorganisms in liquid biofertilizer were Pseudomonas sp., Bacillus sp., Kebsiella sp., Aspergillus sp., Azotobacter sp. **Keywords:** *liquid biofertilizer; local microorganisms; peatland*

I. Introduction

Peatlands in Central Kalimantan are estimated to occupy an area of 3,472 million hectares or around 21.98% of the total land area of Central Kalimantan province, which reaches 15,798 million hectares (Central Kalimantan, go.id). The problem with the use of peatlands is low soil fertility.

In general, the chemical properties of peat soils are dominated by organic acids which are accumulations of plant remains. Physically peat soils are more porous than mineral soils, resulting in rapid movement of water on peat. The total N content is high, but not available for plants because of the high C / N ratio. The content of micro elements, especially Cu, B, and Zn is very low. Nutrients N, P, K, Ca, Mg are also minimal.

The existence of potential microorganisms to fertilize the soil (biofertilizer), naturally the amount is very small (Rifat Hayat, Ummay Amara, Safdar Ali, 2010). Optimization and implementation of potential microorganism-based fertilizers to increase the availability of soil nutrients and to increase crop yields are still very much needed.

Until now, a group of potential isolates for biofertilizer from mining areas in Central Kalimantan have been found. The results of the analysis of the composition of microorganisms at the Microbiology Laboratory, Faculty of Biology, UGM in 2014, showed that the group of microorganisms that were coded for IBT contained 59.6 x 107 cfu / ml cellulitic molds, 16.8×107 cfu / ml nitrogen-fixing bacteria, solvent bacteria phosphate 2,8 x 107 cfu / ml, and cellulite bacteria 180 x 107 cfu / ml. KP microorganisms group contained 3.37 x 107 cfu / ml cellulite molds, 235.6×107 nitrogen fixing bacteria, phosphate solvent

bacteria 2.4 x 107 cfu / ml, and 212 x 107 cfu / ml cellulitic bacteria. Both groups of isolates have also been shown to have the potential to conduct bioremediation of mercury, both in liquid media and in soil (Neneng, et al. 2007 - 2014). Field test results show the potential of isolates to restore land productivity after gold mining and increase plant fertility (Neneng, Liswara, T. Yusintha, 2012), as well as increase peat soil fertility (Winarti & Liswara, 2013).

Several liquid biofertilizer products are available on the market, which also contain potential microorganisms with various advantages, but the success of the application of these microorganisms in the field / on the ground is highly dependent on their ability to adapt to local environmental conditions (Vesna Jerman, Barbara Kraigher, 2014). In this study, the potential of indigenous microorganisms which have been developed as liquid biofertilizers, is expected to have a higher adaptability when applied in peatlands. Compared to microorganisms introduced from areas with different characteristics from peatlands.

This study aims to examine: 1) Effectiveness of liquid media composition supporting the potential of biofertilizer, 2) Effect of liquid biofertilizer on nutrient enhancement in peat soils, 3) Composition of liquid biofertilizer which has the potential to increase nutrients in peat soils.

II. Research Methods

2.1 Microorganism Composition

There are 4 combinations of a consortium of microorganisms used as biofertilizers in this study, namely:

- 1. Consortium of IBT microorganisms consisting of: cellulite fungi, nitrogen-fixing bacteria, phosphate solvent bacteria, and cellulitic bacteria.
- 2. Consortium of IGT microorganisms, consisting of: cellulitic fungi, nitrogen fixing bacteria, phosphate solvent bacteria.
- 3. Consortium of KHY microorganisms, consisting of: cellulite fungi, nitrogen-fixing bacteria, phosphate solvent bacteria, and cellulitic bacteria.
- 4. Consortium of Effective Microorganisms (EM4), liquid biofertilizer as a positive control.

2.2 Liquid Media Material for Biofertilizer

There are 3 liquid media compositions, as shown in Table 1 below:

Liquid Media Composition				
Formula I	Coconut water (70%) +			
	brown sugar (20%) + bran			
	(1%) + charcoal (1%) +			
	livestock waste (4%) +			
	fertile soil (4%)			
Formula II	Coconut water (85%) +			
	sugar (15%)			
Formula III	Coconut water (50%) +			
	brown sugar (40%) + bran			
	(5%) + charcoal (5%)			

Table 1. Liquid Media Composition

Source: Processed Research Data, 2018

2.3 Preparation of Liquid Biofertilizer

All liquid media materials were sterilized in an autoclave, then using aseptic techniques, microorganism culture was inoculated into a sterile liquid medium, with a microorganism culture ratio (20%): liquid media (80%). for 7 days the liquid biofertilizer is then incubated at room temperature.

The experimental design was treated, and repeated 3 times, as shown in Table 2.

Code	Microorganism	Liquid Media Composition			
	composition	Formula I	Formula II	Formula III	
F1	KHY	V	V	V	
F2	IBT	V	V	V	
F3	KHY + IBT	V	V	V	
F4	KHY + IGT	V	V	V	
F5	IBT + IGT	V	V	V	
F6	KHY + IBT + IGT	V	V	V	
F7	EM4	-	-	-	
F8	Air	-	-	-	

Table 2. Experimental Design

Source: Processed Research Data, 2018

a. Field Study

Field studies have been carried out on agricultural land located on peat soils in Jekan Raya District, Central Kalimantan. Application of liquid biofertilizers is done 15 days before planting, and 15 days after planting.

b. Data analysis

Measurement of research parameters include: soybean plant growth (plant height, number of leaves, number of branches, and number of flowers), and soil nutrients (N, P, K, C). Plant growth is measured manually after 40 days of planting. Soil nutrients are measured using the method of soil analysis procedures using the Atomic Absorption Spectro photometer (AAS). All data were analyzed using the ANOVA statistical method.

III. Results and Discussion

3.1 Effectiveness of Liquid Media to Support the Potential of Biofertilizers

The results of this study indicate that there is no significant difference between formula I, formula II, and formula III in terms of supporting the growth of soybean plants in peat soils, on all parameters (Table 3).

Table 3. Comparison of Liquid Biofertilizer Formula in Supporting Soybean Growth in
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Treatment	Rod Height	Number	Number of Interest			
	(cm)	of Leaves	Branches	Amount		
Formula I	22,15	15,08	4,42	249		
Formula II	23,48	17,29	5,21	284		
Formula III	23,30	14,29	4,63	216		
p-value	0,517 ^{ns}	0,096 ^{ns}	0,151 ^{ns}	0,228 ^{ns}		

Source: Processed Research Data, 2018

Based on the measurement results on all parameters of soybean plant growth, it shows that biofertilizer grown in liquid type II formula is more capable of supporting soybean plant growth compared to formula I and forumula III. The composition of formula II consists of simpler ingredients, namely: 85% coconut water and 15% sugar. According to (Atlas, 2019), various types of natural media have long been used to multiply microorganisms, for example urine, meat extracts, potato slices, soy flour, etc., which are pioneering media for the growth of microorganisms.

Microorganisms need a number of macro and micronurients in the right proportion to support optimal growth and metabolism. Microorganisms also need carbon and nitrogen as the main source for growth, while micro elements such as sulfur, phosphorus, vitamins, and others are needed nutrients in small amounts. Coconut water contains 95.5% water, 4% sugar, 0.1% fat, 0.02% calcium, 0.01% phosphorus, 0.5% iron, a number of amino acids, mineral salts, vitamin B complex, vitamin C, and cytokines (Narendrakumar Sekar, 1 Soumya Kariyadan Veetil, 2013). Furthermore (Vigliar R1, Sdepanian VL, 2006), (Narendrakumar Sekar, Soumya Kariyadan Veetil, 2013), states Coconut water contains isotonic solutions. Coconut water contains 4.7 percent of total solids, 2.6 percent sugar, 0.55 percent protein, 0.74 percent fat, and 0.46 percent mineral.

High nutrient content in coconut water, causing it is widely recommended for use as a medium for tissue culture, fungi growth media, and also for the growth of other microorganisms (Jean W. H. Yong, Liya Ge, Yan Fei Ng, 2009).

3.2 Increased Soil Nutrients

a. Measurement Results N-total Nutrient Content

The results of measurements of total N-nutrient content per gram of soil sample, after treatment, are shown in Figure 1 diagram.



Figure 1. Nitrogen Nutrient Level Measurement Results

The graph above shows the average increase in nitrogen levels in the treatment, ranging from 65% to 81.5% in the treatments F3 and F4. Increased levels of soil nitrogen in liquid biofertilizers from local microorganisms in F3 and F4 treatments, on average, were higher by 12.88% when compared to positive controls. Based on international standards (SI), the availability of N-total in the treatment is very high, while the availability of N-total in the negative control, is classified as low, because it is less than 0.5% (Narendrakumar Sekar, Soumya Kariyadan Veetil, 2013).

Nitrogen fixation by free-living bacteria in the soil, belonging to the non-symbiotic group. These soil bacteria can be classified as either autotropic or heterotropic bacteria, which are not in direct symbiosis with plants, but can carry out nitrogen fixation. This

bacterium can be associated with decomposed plant remains, decomposed organic matter aggregates, and in thermite habitats (Roper & Gupta, 2016).

b. Measurement of C-Organic Nutrient Content

The results of the measurement of Carbon nutrient content per gram of soil sample, after treatment, appear in the diagram in Figure 2.



Figure 2. Measurement Results of Carbon Nutrient Levels

The amount of carbon measured in the F4 - F6 treatment, on average is lower than other treatments, including the positive control and negative control treatments. Based on the soil chemical standards listed in international standards, it is known that the amount of organic C> 5%, already classified in the category of very high. Referring to these standards, it can be said that all treatments including controls have a very high organic C content. This can be understood, because the soil media used in this study is peat soils.

The important components of organic matter are C and N. The content of organic matter is determined indirectly by multiplying the C content by a factor that is generally as follows: organic matter content = C x 1,724. If the amount of C-organic in the soil can be known then the content of soil organic matter can also be calculated.

Soil organic matter is all material derived from living things, both animals and plants, which are returned to the soil after the process. When returned to the soil, organic material can contain a series of materials in the form of intact tissue of animals and plants, or in the form of a decomposed mixture, known as (Bot, Alexandra; Benites, n.d.).

The content of organic matter at each horizon is an indication of the amount of accumulation of organic matter under different environmental conditions. The content of organic matter is an indicator of soil fertility. C-organic soil shows the level of organic matter contained in the soil. Peat soils usually have higher levels of organic C than mineral soils. C-organic levels indicate the level of peat maturity. Peat from fibric types will have a higher C-organic level compared to saprik and hemik (Mustamo, Hyvärinen, Ronkanen, & Kløve, 2016).

c. Measurement Result of Phosphate Nutrient Content

The results of Phosphate nutrient content measurements per gram of soil sample, after treatment, are shown in Figure 3 diagram.



Figure 3. Measurement Results of Phosphate Nutrient Content

Based on the data in Figure 3, there was an increase in phosphate nutrients in the F1, F4 and F5 biofertilizer treatments, compared with negative controls. The increase in phosphate nutrients in the treatment ranged from 5.3% to the highest, in the F6 treatment, which was 21%. Overall, phosphate nutrients in positive control treatments were on average higher than other treatments.

d. Measurement Results of Potassium Nutrient Levels

The results of measurement of potassium nutrient content per gram of soil sample, after treatment, can be seen in the diagram in Figure 4.



Figure 4. Measurement Results of Potassium Nutrient Levels

The data in Figure 4 shows that the average value of potassium in the average treatment is higher than in the positive control or negative control. The increase in potassium nutrients in the treatment ranged from 30% to 47%.

The increase in N-total nutrients (%) was an average of 69.7%, an increase in phosphate nutrients, an average of 4.7%, and an increase in potassium nutrients an average of 28%. The increase in potassium nutrients in the biofertilizer treatment is better than the negative and positive controls.

3.3 Effect of Liquid Biofertilizer on Soil Nutrients on Peatlands

The Anova statistical analysis results in Table 4, show that the composition of biofertilizer has a significant effect on nutrient content in peat soils, especially potassium and nitrogen nutrients.

Code	Composition	Nutrient Content in Peat soils			
		K-dd		P-BrayI	N-Total
		(me/100 g)	C-Org (%)	(ppm)	(%)
F1	KHY	0,49 ab	53,30	330,97	1,26 b
F2	IBT	0,49 ab	52,88	273,28	1,35 b
F3	KHY + IBT	0,46 ab	52,08	229,40	1,63 c
F4	KHY + IGT	0,37 a	49,60	302,38	1,63 c
F5	IBT + IGT	0,16 a	46,96	303,15	1,29 b
F6	KHY + IBT + IGT	0,21 a	48,34	363,34	1,34 b
F7	EM4	0,12 a	54,04	446,49	1,42 bc
F8	Control – (Aquadest)	0,26 a	54,66	286,27	0,43 a
P-value		0,045885*	0,175712 ^{ns}	0,1016 ^{ns}	5,47E-07*

Table 4. Effect of Liquid Biofertilizer on N, P, K and C Nutrients in Peatlands

Source: Processed Research Data, 2018

The combination of biofertilizer formulas from the KHY and IBT groups showed a significant effect in increasing N and K nutrients, compared to other liquid biofertilizer groups.

The nutrients available to the soil come from various sources, including the availability of minerals present in the soil, weathering processes, and even various natural processes such as lightning to contribute nitrogen nutrients to the soil. According to Hakim, et al. (1985) The main main way nutrients (nitrogen) enter the soil is the result of the activity of microorganisms, both living freely and in symbiosis with plants. In the latter case the bound nitrogen is used in amino and protein synthesis by the host plant. If plants or microorganisms bind nitrogen-free, decomposing bacteria liberate amino acids from proteins, ammonification bacteria free ammonium from amino groups, which are then dissolved in soil solution. Ammonium is absorbed by plants, or absorbed after being converted into nitrates by nitrifying bacteria (Cooperation, 2014).

Element K is needed by plants in large quantities, which is the second largest after N nutrients. In fertile soils, the K content in tissue is almost the same as N. K does not become a structural component in organic compounds, but is merely ionic, K + is in solution or bound by negative charges from the surface of the tissue for example: R-COO – K +. The main function of K is to activate the enzymes and maintain cell water.

Most soils have high levels of total K, they have more K than other nutrients, whereas for sandy soil naturally the K content is low, the source of K is the mineral feldspar and mica, which will be available slowly, this becomes a source of K in long term, K available represent a small part of the K total.

In the soil, the main source of potassium is minerals, which release this element in the weathering process. This process makes potassium available to plants. Another source of K comes from organic matter. Most K is easily dissolved from plant litter, the release is not related to the level of reshuffle as N or P, this is because K is not a component in the structure of organic compounds (Hillel, 2008).

3.4 Optimal Formula Liquid Biofertilizer Improving Soil Nutrient in Peatlands

Microorganisms play an important role in the decomposition process, and the supply of available nutrients for the soil. The composition of the liquid biofertilizer used in the treatment contains various types of microorganisms that are capable of tethering nitrogen, breaking down cellulitic compounds, and also dissolving phosphates. The composition of microorganisms contained in the KHY + IBT formula is a combination of types of soil-fertilizing microorganisms, namely: *Pseudomonas sp., Bacillus sp., Kebsiella sp., Aspergillus sp., Azotobacter sp.*

Azotobacter is a non-symbiotic gram-negative aerobic bacterium that functions as a free N binding so that these bacteria have an influence on the physical and chemical properties of the soil in increasing soil fertility (Supriyadi, 2009).

Then in (Try & Rahayu, 2007) stated giving the bacteria *Pseudomonas aeruginosa* in Lapindo mud and sand were added *blotong* (sugar factory waste) significantly increase the nutrient phosphorus. Besides being resistant to drought and saline conditions, the fungus *Aspegillus niger and Penicillium sp.* also has the ability to break down cellulose and lignin compounds into simple carbon compounds needed by soil microbes as an energy source (Carbon source).

In another part according to Lakshmikant (1990) reported that the fungus Aspergillus niger, Chaetomium globosum, Scopulariopsis brevicaulis, Trichoderma koningii and Trichothecium roseum have cellulase activity in litter media and wheat straw, so that the fungi can decompose cellulose. Penicillium sp. LM-2 can reduce the color of lignin 0.6 g / L within 4 days, this culture was incubated on a shaker at 25oC (Andriastini, Ramona, & Proborini, 2018).

IV. Conclusion

The liquid biofertilizer treatment, on average, has increased soil nutrients in the form of N, P, K, in the treatment compared to negative controls. Increased N-total nutrients (%) by an average of 69.7%, an increase in phosphate nutrients an average of 4.7%, and an increase in potassium nutrients an average of 28%. The potential microorganism composition in liquid biofertilizer is Pseudomonas sp., Bacillus sp., Kebsiella sp., Aspergillus sp., Azotobacter sp.

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