

**Research Article**

**Utilization of LCC (Legume Cover Crop) and bokashi fertilizer for the efficiency of Fe and Mn uptake of former coal mine land**

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**Abstract:** Coal mining activities have a negative impact on the ecosystem and lead to the disruption of the environment due to waste disposal containing a high concentration of Fe and Mn. In addressing the problem, the biological reclamation approaches using LCC (Legume Cover Crop) is performed to anticipate the acidic condition of soil pH and high concentration of Fe and Mn. This study aimed to determine the effect of a combination of several types of LCCs, i.e., *Centrosema pubescens*, *Calopogonium mucunoides* and *Pueraria javanica* in combination with Bokashi fertilizers application for improving soil chemical properties and the efficiency of Fe and Mn uptake. The results showed that the combination of LCC and bokashi fertilizers had a significant impact on raising soil C-Organic, P and K along with the increasing of Fe and Mn uptake. *Pueraria javanica* had the highest value of BCF (BioConcentration Factor) of Fe and Mn uptake at the level of 0.72, and 0.56 %, respectively and this crop is more potential crop as phytoremediator than *Centrosema pubescens* and *Calopogonium mucunoides*. Canonical Variate Analysis could distinguish the position and distance among the treatments based on selected parameters.

**Keywords:** land rehabilitation, mining, revegetation

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**Introduction**

Coal is one of the most abundant mineral export commodities in Indonesia, where based on data from the Ministry of Energy and Mineral Resources, Directorate General of Mineral and Coal (2015), coal production in (2015) reached 264,443,663.52 tons, but mining activities have a negative impact to the ecosystem and environment. This is due to the waste produced during mining activities enriched with Fe and Mn in high concentration, and the pH becomes acidic, resulting in a various problem to surrounding environment mainly when mining process is not properly managed. Decreasing the pH value may due to the establishment of by pyritic rocks which are exposed to open air and reacts with oxygen to produce high acidity and could reach the values less than (<2) (Dowarah et al., 2009; Widyati,

2009). Apart from that, the coal mining process is usually carried out with an open-pit system, where all natural vegetation is lost which results in permanent changes in landscape (topography). The process of extracting topsoil soils can damage the geological structure of the soil which can interfere with the hydrological system (Shrestha and Lal, 2006). Many mining areas in Sumatra, are usually low in soil fertility, high Fe, Al and Mn content so that they are grouped into the Ultisols. Rehabilitation of ex-coal mining land that has been carried out by PT Bukit Asam Persero Tbk is to reuse top soil into the ex-mining land as a planting medium for pioneer plants to maintain the physical properties of the soil. Application of dolomite lime or rock phosphate to improve soil chemical properties has not been effective, due to high rainfall and large erosion rates. Therefore, it is necessary to explore

alternative ways of rehabilitating ex-mine land by planting LCC (Legume Cover Crop) (Mukhopadhyay et al., 2013; Mukhopadhyay et al., 2014). The LCC can produce biomass in rapid quantities and can fix N from the air (Sheoran et al., 2010). Singh et al (2002) recommended that LCC plants used should be local species that have adapted to the existing environment. The LCCs have been tested in areas of former coal mines which have a significant effect on increasing the C-organic content, N-total and soil pH (Agus et al., 2013), but the information on how efficient Fe and Mn uptake by LCC is very limited. On the other hand, the application of LCC alone is hypothesized not to give satisfactory results so that a combination of soil enhancing ingredients such as bokashi fertilizer is needed so that this plant can grow and develop well in the former coal mine land.

Bokashi fertilizer contains essential microorganisms and can provide nutrients for plants (Zahrah, 2011). Previous research found that the use of organic fertilizers including bokashi fertilizer could reduce the adverse effects of Ultisols through improvements in soil physical, chemical and biological properties (Karimuna, 2007). This is an alternative way to improve soil fertility through the use of organic materials that have been decomposed by effective microorganisms (EM4), which when applied with organic materials derived from *Chromolaena odorata* L. (Karimuna, 2000), has a very positive impact because it can also be associated with mycorrhiza (Halim, 2009; Halim, 2012). The application of organic materials from *Chromolaena odorata*, *Imperata cylindrica* L. Beauv, and *Colopogonium mucunoides* L. combined with bokashi fertilizer can increase the yield of maize and peanuts twice as much (Karimuna et al., 2012; Karimuna et al., 2009).

This study aimed to determine the effect and effectiveness of bokashi fertilizer combined with organic mulch from LCCs (*Centrosema pubescens*, *Calopogonium mucunoides* and *Pueraria javanica*) on the improvement of soil chemical properties of an Ultisol at the former Bukit Asam Coal mine.

## Materials and Methods

This research was conducted from July 2015 to February 2016 at PT Bukit Asam, Jl. Parigi No. 1 Tanjung Enim 31716 on the reclaimed (Agropremium) land in the Pit. 3 East Banko Barat with an elevation of 135 m with coordinates 48 M, X: 0370587 and Y: 9587496. This study uses a randomized block design method with six treatments and three replications. The treatments

used in this study were combinations of bokashi fertilizer and three types of LCC (Legume Cover Crop) as follows: (1) *Centrosema pubescens* (CP0), (2) *Calopogonium mucunoides* (CM0), (3) *Pueraria javanica* (PJ0), (4) *Centrosema pubescens* + 100 g bokashi fertilizer / plant (CP1), (5) *Calopogonium mucunoides* + 100 g bokashi fertilizer / plant (CM1) and (6) *Pueraria javanica* + 100 g bokashi fertilizer / plant (PJ1).

Analysis of soil chemical properties carried out at the Chemical Laboratory of the Soil Department, Faculty of Agriculture, Brawijaya University, Malang included pH (pH meter), organic C (Walkley and Black method), P (Olsen method), and K (HCl 25% method), Fe and Mn uptake by plants were determined using a spectrophotometer. Plant growth variables observed were plant height and biomass. Data were subjected to Analysis of Variance followed by Duncan's test at 5% level of significance. Besides, correlation tests were also conducted to determine the closeness of the relationship between treatments. The results of the correlation test were continued with multivariate regression and analysis with the Canonical Variate approach using the application GenStat Discovery 10<sup>th</sup> Edition. While for the efficiency of Fe and Mn absorption/uptake, BCF calculation was done by the equation as follow:  $BCF = \text{Cons aboveground} / \text{Cons soil}$ , where Cons aboveground is the concentration of metals in the stem and leaf tissue and Cons soil is the concentration of metals in the soil (Li et al, 2009).

## Results and Discussion

The initial pH of soil was categorized as very acidic (3.84). Treatment of bokashi application combined with LCCs significantly affected soil pH (H<sub>2</sub>O) (p <0.05). The highest increase in soil pH was found in the treatment of bokashi fertilizer combined with *Centrosema pubescens* (CP) although it was not significantly different to other LCC treatments, except with the application of CP without the addition of bokashi (Figure 1). The low value of soil pH is thought to originate from the oxidation process of Fe in the pyrite minerals which increases the H<sup>+</sup> ion released into the soil (Wilkin and Barnes, 1996). The addition of bokashi can bind the excess of oxidized Fe or excess H<sup>+</sup> ions through the chelatilization process. The highest soil organic-C was found in *Centrocema pubescens* treatment with bokashi application, which was significantly different (p <0.05) with all LCC treatments without bokashi application, with an average value of 6.63%, whereas the organic-C content in the treatment without bokashi fertilizer decreased to 4.18%

(Figure 2). Increased levels of organic-C in all bokashi treatments are thought to come from the decomposition and mineralization of organic compounds contained in bokashi. Soil pH which tends to lead to neutral conditions also helps the decomposition and mineralization of organic matter (Stevenson, 1994). The highest available soil phosphorus content was found in *Centrosema pubescens* which was not significantly different from *Calopogonium mucunoides* with bokashi fertilizer ( $p < 0.05$ ). Besides, all LLC treatment with bokashi application tended to be significantly different from those treatments without bokashi.

Soil with *Pueraria javanica* with the application of bokashi containing available P of ten times higher than those treatments without bokashi (Figure 3). This shows that some of the inorganic-P has been released into the soil through bokashi decomposition and mineralization process. The increase in soil phosphorus was 5-7 times higher than the initial condition, in average. Six months after planting, both types of LCC (*Pueraria javanica* and *Calopogonium mucunoides*) could overburden former tin mining land with an average life ability performance at 83.3%.

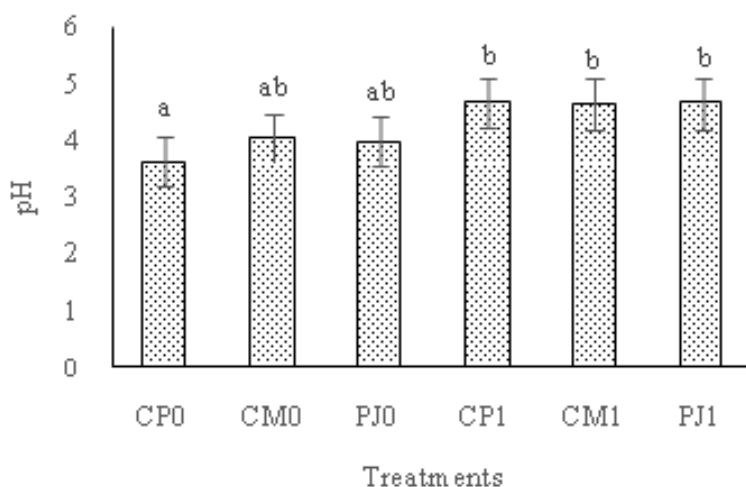


Figure 1. pH(H<sub>2</sub>O) due to the application of LCC and bokashi

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

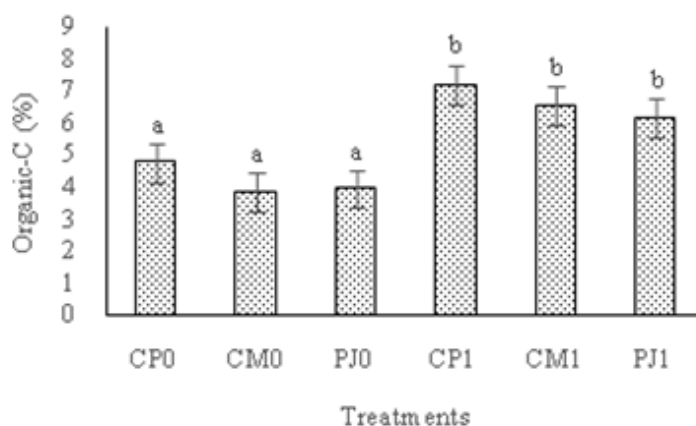


Figure 2. Organic-C (%) due to the application of LCC and bokashi

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

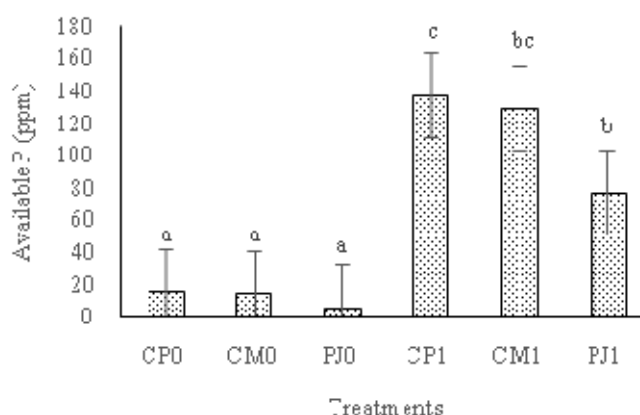


Figure 3. Available P (ppm) due to the application of LCC and bokashi

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

*Pueraria javanica* had a higher ability to cover soil surface and produced biomass at the level of 308.8 g compared to *Calopogonium mucunoides*. Inline cropping pattern of LCC can cover land surface (1.2 m<sup>2</sup>) faster than a scattered pattern and produced higher biomass (380.7 g) (Narendra and Pratiwi, 2014). *Pueraria javanica* consistently produced the highest biomass of 10.2 t/ha/year, whereas *Calopogonium mucunoides* only produced 3.8 t/ha/year. In general, every kilogram of LCC biomass was significantly capable of contributing 1.9-4.6 g of organic-C and 0.52-0.78

g of total N. P-Bray and K values also increased respectively 1.3-2.3 and 3.2-4.2 mg/100 g of biomass. Nutrient additions to this soil were significantly found greater in *Pueraria javanica* as a response to the high organic matter (Dinesh et al., 2004). Treatments of *Centrosema pubescens* and *Calopogonium mucunoides* with bokashi fertilizer resulted in significant differences in soil K content ( $p < 0.05$ ) with a two-fold increase from the initial conditions when compared to other treatments (Figure 4).

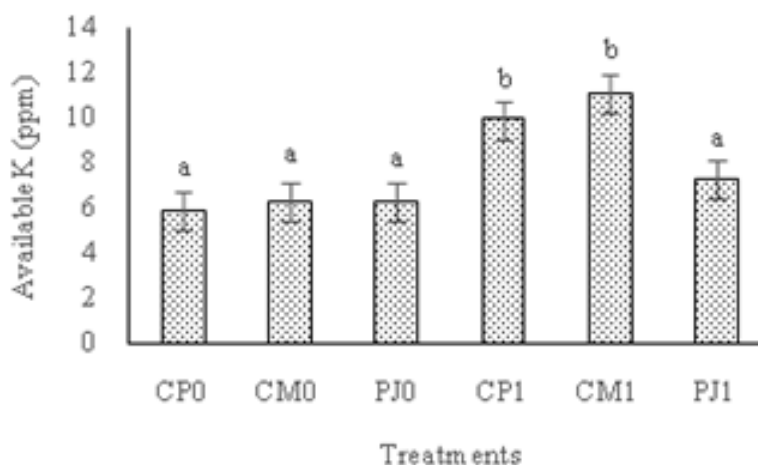


Figure 4. Available K (ppm) due to the application of LCC and bokashi

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

The release of available K in the soil can be triggered by an increase in organic-C which can increase soil microorganism activity and pH leading to neutral dramatically affects the availability of K and P for plants (Kastono, 2005; Hanafiah, 2012; Rosmarkam, 2013). Bokashi fertilizer fermented with EM<sub>4</sub> can dissolve phosphate compounds that are not available to plants (Wididana, 1998). Crop length since the first week (M1) showed a significant difference between the treatments of with (+B) and without (-B) bokashi application ( $p < 0.05$ ), until the period

of the fourth week after planting (M4) (Figure 5). The lowest crop length was observed under treatment without bokashi fertilizer in *Pueraria javanica*, while the highest crop length was found under bokashi application in *Centrosema pubescens* although it was not significantly different to *Calopogonium mucunoides*. The LCC treated with bokashi had an average crop length at a value of 40 cm, while under the treatments of LCC without bokashi fertilizer, it only reached an average length of 14 cm at fourth week (M4) after planting (Figure 5).

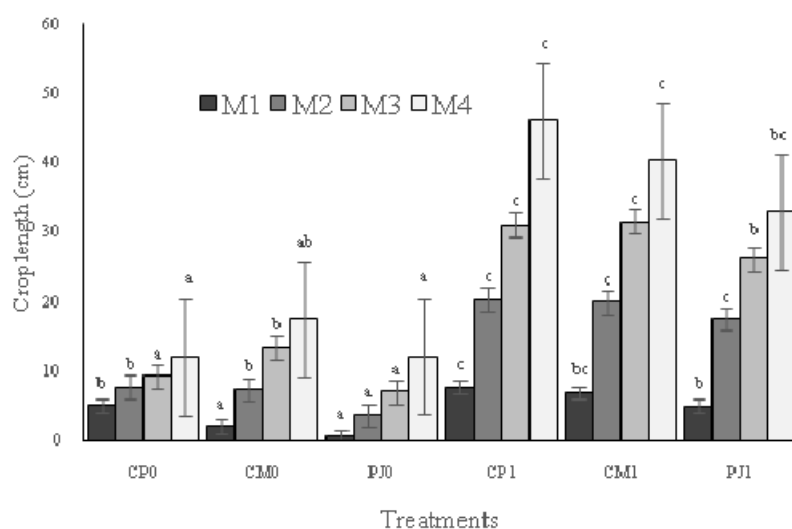


Figure 5. Crop length (cm) due to the application of LCC and bokashi

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

LCC growth became inhibited if there were no bokashi applications; with bokashi the length of LCC increased 2-3 times compared to other treatments without bokashi. *Pueraria javanica* had the lowest crop length in which the length

was not significantly different at 4 weeks after planting. Visual effects of bokashi addition to *Pueraria javanica*, *Centrosema pubescens* and *Calopogonium mucunoides* are presented in Figures 6, 7 and 8.



Figure 6. The difference in crop performance of *Centrosema pubescens* due to bokashi application (+B) and without (-B)

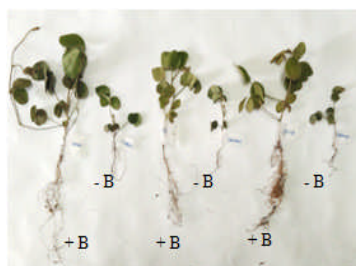


Figure 7. The difference in crop performance of *Calopogonium mucunoides* due to bokashi application (+B) and without (-B)



Figure 8. The difference in crop performance of *Pueraria javanica* due to bokashi application (+B) and without (-B)

The application of bokashi gave a significant effect ( $p < 0.05$ ) on increasing the value of fresh weight (FW) and dry weight (DW) of LCC. The highest fresh weight and dry weight of plants were found in the treatment of *Calopogonium mucunoides* + bokashi, despite the highest data on plant length and soil chemical properties (C, P and K) found under *Centrocema pubescens* + bokashi

treatment (Figure 9). The treatment of bokashi addition to LCC increased FW and DW up to two times compared to no bokashi application treatment. This is consistent with the research from Syarif (2009) that *Calopogonium mucunoides* plant has the highest biomass accumulation compared to other plants on ex-gold mining land.

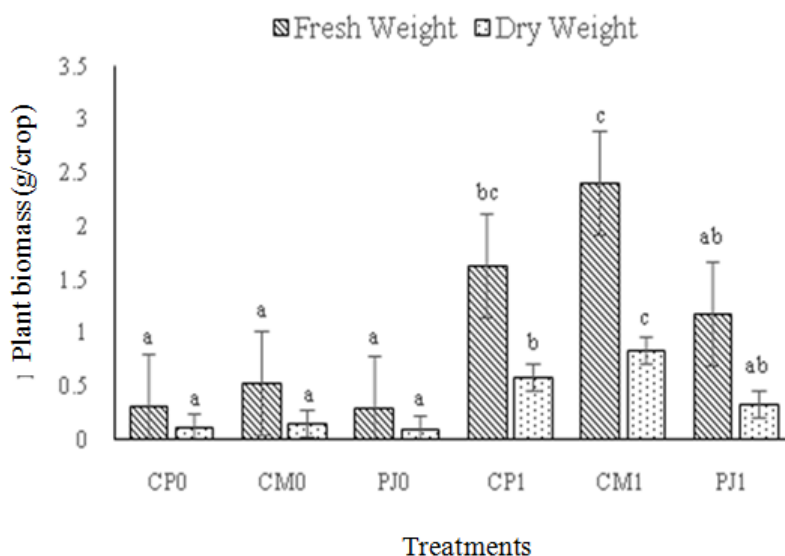


Figure 9. Fresh weight (FW) and dry weight (DW) of LCC due to bokashi application

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

This LCC is being able to adapt to nutrient-poor and acidic soils with a pH of 4.5-5.0, can survive throughout the year, and tolerant to water less, tolerant of long droughts (Jamin et al., 2002; Cook, 2005). This type of LCC has rapid initial growth and ability to clean contaminant metal in gold mining waste (Armecin et al., 2004). *Pueraria javanica* absorbed the highest Fe to their biomass, with an average of 26.62 ppm which was significantly different ( $p < 0.05$ ) to those absorption found under *Calopogonium mucunoides*. Both had significantly different ( $p < 0.05$ ) to that Fe absorption of other treatments. The average Fe absorption of the treatment without the addition of bokashi was at the level of 12 ppm (Figure 10). The highest Mn uptake was found in *Pueraria javanica* with bokashi addition at a value of 187.05 ppm which was significantly different ( $p < 0.05$ ) to other treatments. The value was three times more than *Calopogonium mucunoides* without the bokashi application (57.1 ppm) resulting in the lowest Mn absorption value

(Figure 11). BCF values that showed differences in elemental concentrations in soil and plant tissue showed that the treatment of *Pueraria javanica* and *Calopogonium mucunoides* with bokashi addition were not significantly different ( $p < 0.05$ ), although both showed the highest values among treatments. The BCF value of Fe with bokashi treatment was almost two times higher than the treatment of LCC without bokashi (Figure 12). The BCF value of Mn had a similar pattern to Fe uptake, where the highest value was detected on *Pueraria javanica* with bokashi addition. Although *Calopogonium mucunoides* added with bokashi treatment did not show a significant difference in BCF values ( $p < 0.05$ ) with the treatment of *Centrocema pubescens* and *Pueraria javanica* without addition of bokashi, but the lowest BCF value was found in the LCC treatment of *Calopogonium mucunoides* without bokashi (Figure 13). Fe and Mn uptake of *Pueraria javanica* with bokashi fertilizer application had the highest effectiveness value of 0.72 and 0.65 %, respectively.

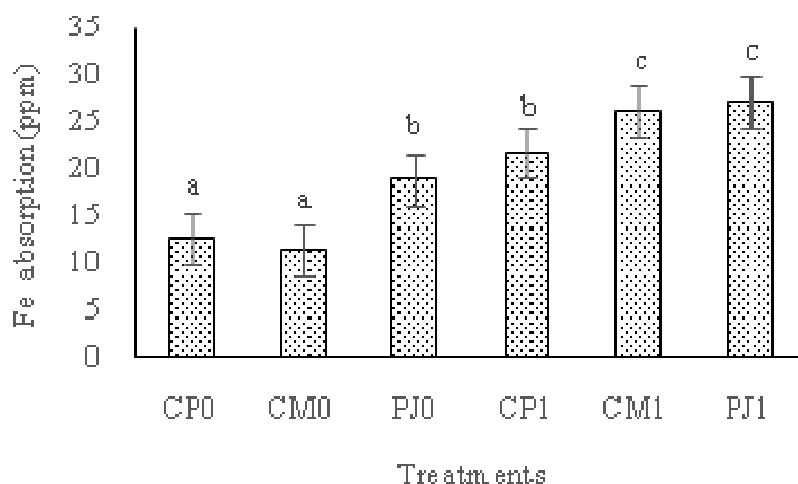


Figure 10. Fe absorption of LCC due to bokashi application

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

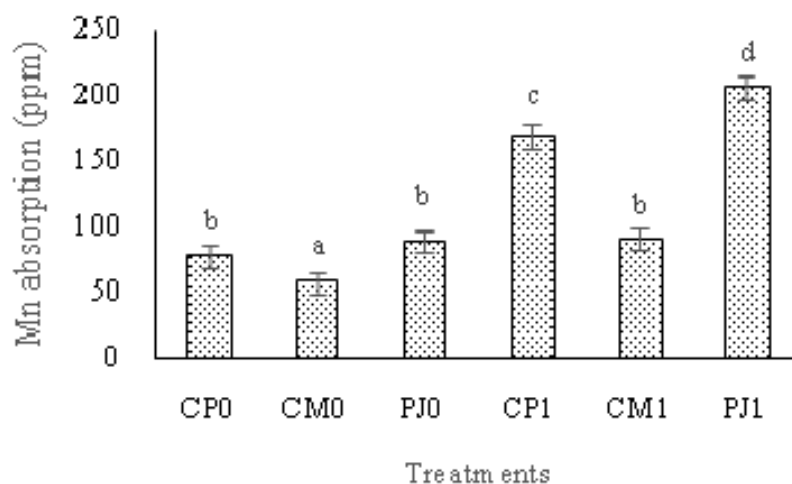


Figure 11. Mangan (Mn) absorption

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

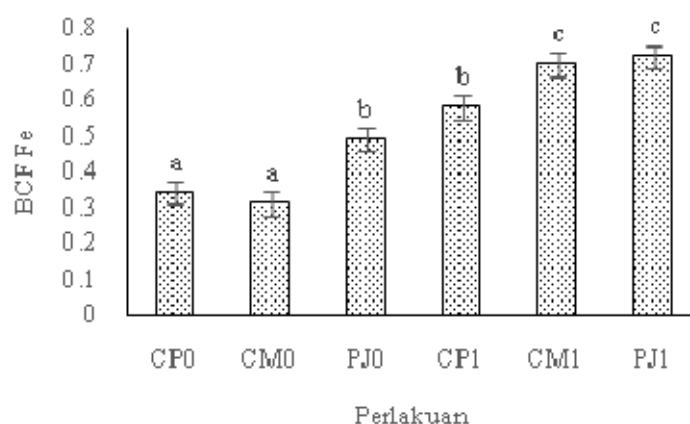


Figure 12. BCF (Bio Concentration Factor) of iron (Fe)

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

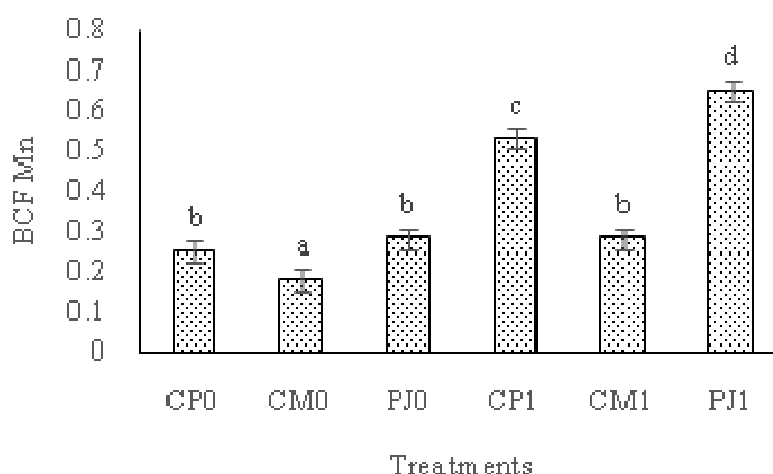


Figure 13. BCF (Bio Concentration Factor) of mangan (Mn)

Remark: CP0 (*Centrosema pubescens*), CM0 (*Calopogonium mucunoides*), PJ0 (*Pueraria javanica*), CP1 (*Centrosema pubescens* + bokashi fertilizer), CM1 (*Calopogonium mucunoides* + bokashi fertilizer) and PJ1 (*Pueraria javanica* + bokashi fertilizer); Figures followed by the same letter show no significant difference based on Duncan's test level of 5%.

Moreover, *Centrosema pubescens* and *Calopogonium mucunoides* had an effective absorption of Fe and Mn at the value of 0.58 and 0.53%; 0.70 and 0.28%. Higher BCF value can be assumed to the higher effectiveness of metal absorption by LCC; therefore, the LCC can be used as a phytoremediation plant (Yoon et al., 2006). Furthermore, the correlation test showed that the pH of the soil with the uptake of Fe and

Mn had a strong relationship with the values of  $r = 0.58$  and  $r = 0.53$ . Increasing soil pH to neutral will increase the effectiveness of Fe and Mn uptake into plants. It can be seen that the increase in pH towards neutral is closely related to the increase in soil organic-C ( $r = 0.53$ ), where the increase in organic-C also affects K and P available with the correlation values of  $r = 0.50$  and  $r = 0.65$ , respectively (Table 1).



Table 1. Correlation test results between observed parameters

	Organic C	Total N	Available K	Available P	pH	Fe uptake	Mn uptake
<b>Organic C</b>	1						
<b>Total N</b>	-0.83	1					
<b>Available K</b>	0.50	-0.44	1				
<b>Available P</b>	0.65	-0.56	0.90	1			
<b>pH</b>	0.53	-0.44	0.65	0.76	1		
<b>Fe uptake</b>	0.62	-0.41	0.60	0.63	0.57	1	
<b>Mn uptake</b>	0.59	-0.48	0.28	0.51	0.53	0.69	1

Canonical Variate Analysis presented in Figure 14 shows that between CP0, CM0, PJ0, CP1, CM1 and PJ1 treatments were significantly different. This is indicated by a circle of confidence intervals (95%) that are intersecting or not intersecting. In the PJ1 and CP1 treatment had a positive correlation to the axis of Canonical Variate 1 with a percentage variation of 83.69% higher than the axis of Canonical Variate 2 of 13.78% where the treatment of CP1 and CM1 had

a positive correlation to this axis. As for the CP0 treatment, CM0 and PJ0 had a negative correlation with both Canonical Variate axes. So it can be concluded that in each observation parameter in one treatment has a relationship with each other which are mutually binding, where pH, organic C, total N, available P and K, uptake of Fe and Mn, and BCF of Fe and Mn affect each other in one treatment.

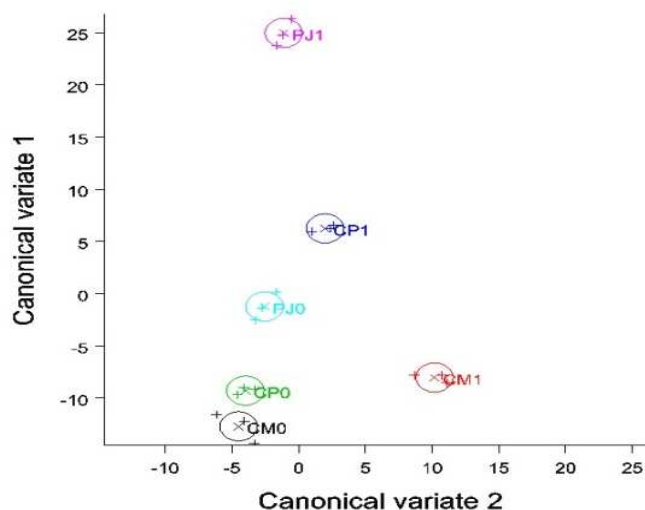


Figure 14. Canonical Variate Analysis based on the selected parameter for clustering across the treatments

**Conclusion**

The combination of LCC and bokashi fertilizer gave a positive influence in increasing soil fertility as seen from the increase of essential nutrients in the soil such as pH, organic-C, available soil P and K. *Centrosema pubescens* had a positive impact on increasing soil nutrients (C-organic, P and K available) when added with bokashi application. In term of biomass and crop growth, the best performance was in *Calopogonium mucunoides* with bokashi

treatment. On the other hand, *Purearia javanica* plant has a better efficiency in the absorption of Fe and compared to *Centrosema pubescens* and *Calopogonium mucunoides*. Canonical Variate Analysis could distinguish the position and distance among the treatments

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