

The Effect of Phytoremediation of Heavy Metal by Orok-Orok (Crotalaria sp.) On the Growth of Aloe Vera.

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Abstract: One of the most important problems in peatland is contamination of soil and water by toxic substances, mainly heavy metal such as Al and Pb. Phytoremediation could be used as an alternative technique to overcome this problem. Phytoremediation is defined as clean up of pollutants primarily mediated by hyperaccumulator plants. These plants have several beneficial characteristics such as the ability to accumulate metal in their roots and an especially high tolerance to heavy metals. This research was carried out to study the potencies of local species to accumulate Al and Pb. Four place, two species were collected from several location and then the Al and Pb accumulated in each species were analyzed. The result showed that some species accumulated Al and Pb in high concentration such as *Crotalaria juncea* L. (1999.43mg kg⁻¹ Aluminium (83%)) and *Crotalaria mucronata* (1555.33 mg kg⁻¹ (80%)). A series of research is needed to prove that these species are potential as heavy metal accumulators.

Keyword: phytoemediation, heavy metal, hyperaccumulator plants.

I. Introduction

Aloe vera is one of the agricultural commodities in the tropical area that has great opportunity to be developed particularly in West Kalimantan Province. The quality of Aloevera flour that contains heavy metal causes a decrease in import and export in these few years. Since 2010 this decrease in export brings out some opinions that soil as the media of Aloevera itself contains a very high heavy metal, even surpassing the normal line. One of the effort or way to diminish the content of heavy metal both in soil and Aloevera's midrib gel is by applying hyperaccumulator plant treatment as phytoremediation alternative technology.

This phytoremediation method is one of technology's breakthroughs due to its reachable cost and appropriateness towards farmers. Phytoremediation itself means the use of specific plant to remove, transfer, stabilize, or eliminate the pollutants, both organic and inorganic (Foth, 2001). The plant used for phytoremediation should have high adsorption and would be able to grow well in contaminated soil (Vangronsveld et al., 2000). Recently, the reclamation engineering by phytoremediation has grown rapidly due to it is cheaper than other method, such as the planting of hyper-accumulators to adsorb heavy metal from the soil (Chaney et al., 2007). Two types of hyper-accumulators, which were used, have been able to adsorb metal in varying levels, but some of them have been able to accumulate specific element of metal in high concentration (Charlena, 2004).

Some results of the research have proven the success of plant use for remediation purpose, and few of them, which have been proven as hyper-accumulator, are derived from tropical areas. For instance, the application of hyper-accumulator (*Crotalaria* sp.) has been able to adsorb some heavy metals from the soil, such as Aluminum (Al) for about 2421 – 3363 mg ka⁻¹, and could adsorb Cadmium (Cd), Lead (Pb), Copper (Cu) for about 24, 542, and 2162 mg/m², respectively (Hidayati et al., 2004). It is expected that the application of such hyper-accumulator would be able to reduce toxicity caused by heavy metals, such as Aluminum and Lead, as well as increase the optimal growth of the aloe vera in the peat soil, both quality and quantity.

Along with the spread of contaminated soil by heavy metals and the rapid growth of phytoremediation, the rehabilitation engineering is an economical alternative and effective to be developed. Therefore, it requires further research that concerning with types of plants, which would be able to accumulate heavy metals in order to make the peat land to be secured for health and environment. Objective of the research was to study effectiveness of the *Crotalaria* sp. Application on the adsorption ability on heavy metals in the soil in order to increase production and improve quality of the aloe vera in the soil that having high level of heavy metals.

II. Material And Methods

The research used the medium in polybags and conducted at UPTD Aloevera Center in Pontianak, West Kalimantan Province, at the altitude of ± 50 m asl by the average temperature of 34° C, average humidity is about 99.58% and rainfalls is about 3000-4000 mm/day. The research was conducted from October 2013 to February 2014.

Preliminary test on analysis of the nutrient levels for N, P, K, C- Organic, metal content of Aluminum, Lead, and acidity level of the soil (pH) was done in August 2013 at the Laboratory of Soil Science and Land

Resources, UNTAN. Meanwhile, for intermediate test on Aluminum and Lead contained in the roots of *Crotalaria sp.*, the roots of aloe vera, gel of the succulent leaf of the aloe vera, and pH level of the soil, heavy metals in the soil, which include Aluminum and Lead, was done at the Laboratory of Natural Resources Institute and Research on Plants in the West Kalimantan Province.

This research used the peat soil of four locations, in which each location has different characteristics. In general, the peat soils from those four locations have tolerable maturity level (hemic), decomposition of the organic materials has not completed yet by C-Organic from the first to the fourth location successively (50.79%, 49.10%, 50.77%, and 51.34%), low total nitrogen (1.84%, 1.4%, 1.4%, and 1.53%), the content level of Aluminum (1747, 1479, 1064, and 680 mg kg⁻¹), the content level of Lead (112.51, 0.08, 28.40, and 22.76 mg kg⁻¹) and reaction of highly acid soil (pH = 4.24, 4.41, 4.47, and 4.75). Therefore, the soil has low fertility level. The seeds of Aloe vera were taken from the aloe vera plantlets at the Aloe vera Seedling Center using Aloe verachinensis, which has morphological characteristics, such as 27 cm height, 4 succulent leaves and at the age of ± 4 weeks. A part of the hyper-accumulators, which were being studied in this research, were selected following the previous result of the research, viz. inventory of the tropical plant types (Sambas, 2002). The hyper-accumulator plants included *Crotalaria sp.* of *Crotalaria juncea*L. and *Crotalaria mucronata*. The organic fertilizers include the stable manure derived from farmer around the Aloe vera Center farming.

This research used the Randomized Block Design (RBD) by 3 replications and 12 treatments. The treatments included P1: Concentration of Al :1747 mg kg⁻¹, Pb : 112.51 mg kg⁻¹ by planting aloe vera, P2: Concentration of Al :1479 mg kg⁻¹, Pb : 0.08 mg kg⁻¹ by planting aloe vera, P3: Concentration of Al :1064 mg kg⁻¹, Pb : 28.40 mg kg⁻¹ by planting aloe vera, P4: Concentration of Al : 80 mg kg⁻¹, Pb : 22.76 mg kg⁻¹ by planting aloe vera, P5: Concentration of Al : 1747 mg kg⁻¹, Pb : 112.51 mg kg⁻¹ by planting aloe vera + *Crotalaria mucronata*, P6: Concentration of Al :1479 mg kg⁻¹, Pb : 0.08 mg kg⁻¹ by planting aloe vera + *Crotalaria mucronata*, P7: Concentration of Al :1064 mg kg⁻¹, Pb : 28.40 mg kg⁻¹ by planting aloe vera+ *Crotalaria mucronata*, P8: Concentration of Al : 80 mg kg⁻¹, Pb : 22.76 mg kg⁻¹ by planting aloe vera + *Crotalaria mucronata*, P9: Concentration of Al : 1747 mg kg⁻¹, Pb : 112.51 mg kg⁻¹ by planting aloe vera + *Crotalaria juncea* L., P10: Concentration of Al :1479 mg kg⁻¹, Pb : 0.08 mg kg⁻¹ by planting aloe vera+ *Crotalaria juncea* L., P11: Concentration of Al :1064 mg kg⁻¹, Pb : 28.40 mg kg⁻¹ by planting aloe vera + *Crotalaria juncea* L. and P12: Concentration of Al : 80 mg kg⁻¹, Pb : 22.76 mg kg⁻¹ by planting aloe vera+ *Crotalaria juncea* L. Data analysis used analysis of variance by F-test at the significant level of 5%. Moreover, in order to find out the difference among treatments, the Smallest Significant Difference (SSD) test has been done at the level of 5%.

Composition of the planting medium has been obtained from result of the preliminary research that derived from four different locations, which have different levels of heavy metals along with the amount of planting medium in polybags of 10 kg each. The seeds of aloe vera were taken from the seedbed. Then the seeds were planted in the polybags. The research used Aloe verachinensis. Meanwhile, *Crotalaria juncea* L. and *Crotalaria mucronata* were planted around the aloe vera, 5 seeds per polybag by 4 times planting cycles. Each planting cycle of *Crotalaria sp.* was 28 days. Stable manure was applied, 250 g polybag⁻¹ for each treatment. The stable manure was applied pre-planting of aloe vera and *Crotalaria sp.* was disseminated around the aloe vera.

III. Results And Discussion

Observation was done on aloe vera, hyper-accumulator (*Crotalaria sp.*) and analysis of the heavy metal content in the soil and water. Numbers of the observed hyper-accumulator plants (*Crotalaria sp.*) were 5 plants polybag⁻¹. While for the aloe vera included 5 plants treatment⁻¹. Parameters of the growth included plant height and numbers of leaf for *Crotalaria sp.*, which were observed once a week. Numbers of succulent leaf, plant height, length and width of the succulent leaves were observed once a month. Meanwhile, analysis on heavy metal content in the roots of *Crotalaria sp.* was done once a month, and the roots of aloe vera as well as gel of the succulent leaf of aloe vera were observed at 120 days after planting. Measurement of heavy metals, particularly in the parts of the plant was done using Atomic Absorption Spectrophotometer. Aloe vera has succulent leaves, which are taken to be processed. There are no specific criteria or standard for the succulent leaves, which are ready to be harvested. Criteria or requirements for the harvested succulent leaves must be adjusted to export purpose, in which the minimal weight must be 0.8 kg, no wound or lesion on the succulent leaves, the tip part of the succulent leaf should not dried or burnt, the color is pure green, unrotten, hard texture, and length of the succulent leaf is about 50-60 cm, and the width is about 10-14 cm, as well as the flesh thickness is about 2-3 cm. Up to the present, quality standard of aloe vera is still based on the physical criteria, and not related to the nutrient content of the aloe vera (qualitative). Therefore, this qualification relates to the method about the proper harvesting, appropriate breeding techniques, and the right maintenance of the aloe vera (Feller, A.K. 2000). High level of damages or defects on the succulent leaf will affect on the reduced yield of the qualified products. The increasing yield and quality of the plant and soil as medium for aloe vera breeding can be done through phytoremediation.

Some efforts to remediate the heavy metal-contaminated land that is hospitable to environment, are easily to be applied and efficient, and it is so-called Phytoremediation (Cunningham and Ow, 1996; Cunningham et al., 1997). The plants, which can be applied for phytoremediation, should have high biomass production, have been able to accumulate the contaminant at the entire parts of the plant that has beyond the contaminant level in the soil (hyper-accumulator) and tolerable to environment (Chee Y.K. and C.P. Chen. 2002).

The cause of different heavy metal-accumulation in both types of *Crotalaria* sp. has not known yet. However, the hyper-accumulator plants should meet the requirements for phytoremediation, such as tolerability to high concentration of metal in the roots of the plant (Bingham et al., 1989; Piccini and Malavolta, 1992). Therefore, before applying the phytoremediation technique, the hyper-accumulator plant should be learnt in relation to the plant's response, ability of the plant in accumulation and tolerant to the contaminated soil.

The important thing that must be known in phytoremediation is whether the plant has hyper-accumulator features in order to reach the success of phytoremediation in the metal-contaminated land. In general, such hyper-accumulator plants have been able to accumulate or extract heavy metals to the part of the plant and rapid growth of the plant. Deep rooting system and high biomass production are the main factors of success in using hyper-accumulator (Kidd et al., 2009). Efficient phytoremediation depends on high concentration in biomass and produce them in greater numbers.

Rapid growth of the plant will directly affect the remediation process. The application of phytoreaction method for the remediation success requires specific plant that has the ability of high metal-accumulation, rapid growth and high biomass yield, as well as easily to be bred without any complicated adjustment of location (Vangrosveld and Cunningham, 1998). Rapid growth of the plant will shorten the required time to reach high biomass. Selection on breeding plants has been done for phytoremediation in relation to the ability to grow rapidly and produce high biomass.

Result of the research showed that the accumulator plant, *Crotalaria mucronata*, has been able to produce lower biomass than *Crotalaria juncea*L., for about two folds (Table 1). The average values values of both types of plants, *Crotalaria juncea*L. tended to have higher Aluminum accumulation than *Crotalaria mucronata*, for about 1919.43 mg kg⁻¹. Metal availability and mobility around the rooting system (rhizosphere) are the important aspect in metal adsorption from the soil to the roots (Maestriet al., 2010). As reported by Puschenreiter et al. (2005), metal availability around the hyper-accumulator's roots were higher than on non-accumulator. Root exudates, such as organic acid and phytosiderophores as well as acidity of the soil have played important roles in metal mobilization from the soil to the roots (Marschener, 2002).

Table 1. Result of heavy metal adsorption by the roots of *Crotalaria* sp. (mg kg⁻¹)

Treatment	Aluminum content per planting period				Lead content per planting period			
	I	II	III	Amount of Al adsorption	I	II	III	Amount of Pb adsorption
P5	754.0	734	268	1756.0 d	0,02	3,36	0,911	4,291 b
P6	701.9	656	266.4	1624.3 c	0,02	0,645	0,101	0,766 a
P7	698.6	544	250	1492.6 b	0,02	1,68	0,70	2,4 ab
P8	606.4	536	206	1348.4 a	0,02	0,772	0,505	1,30 a
P9	878.5	1960	148	2986.5 h	0,02	2,90	1,81	4,73 c
P10	702.3	879	99.4	1680.7 g	0,02	0,45	0,53	1 a
P11	656.2	820	78.3	1554.5 f	0,02	2,86	1,43	4,31 b
P12	624.6	754	77.4	1456.0 e	0,02	0,472	0,56	1,052 a
SSD 5%			3.67					2.01

Notes : Numbers followed by the same letter in the same column show insignificant difference based on SSD test at the level of 5%.

- P5 : (Al:1747mg kg⁻¹,Pb:112.51 mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
- P6 : (Al:1479 mg kg⁻¹,Pb: 0.08mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
- P7 : (Al:1064mg kg⁻¹,Pb:28.40mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
- P8 : (Al:680 mg kg⁻¹,Pb:22.76 mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
- P9 : (Al:1747 mg kg⁻¹,Pb:112.51mg kg⁻¹)+ Aloe vera + *Crotalaria juncea* L.
- P10 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹)+ Aloe vera + *Crotalaria juncea* L.
- P11 : (Al: 1064 mg kg⁻¹,Pb:28.40 mg kg⁻¹) + Aloe vera + *Crotalaria juncea* L.
- P12 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera + *Crotalaria juncea* L.

During the observation, it showed that the planting of aloe vera and hyper-accumulator (*Crotalaria* sp.) has positive effect, so that it will be able to increase the accumulation ability of Aluminum in the roots. According to Maestriet al., (2010) the role of microorganism around the rooting system of *Crotalaria* sp. affected the metal adsorption and the plant growth. The bacterial colony, which grew on the root nodule, will be filled with *Rhizobium* bacteria that would be useful for symbiosis with the plant. Meanwhile, the roots have

cooperated to form the root nodule, which play actively in attaching nitrogen for the soil (de Resende et al. 2003). Besides that, the bacteria in the Rhizobium could affect mobility and availability of metal in some ways, such as reducing pH of the soil, producing antibiotic compounds, organic acid, hormone (for instance: IAA) and metal-chelate agent, which is able to increase the root biomass. Apparently, rooting environment of both hyper-accumulator and non-accumulator species provide different effect on metal availability and root adsorption (Maestri et al., 2010).

Relating to the above observation, it seems that the growth of hyper-accumulator and aloe vera will grow well even in Al-contaminated medium (Figure 1b). The aloe vera, which were planted solely, have been able to make an adaptation, up to the fourth month, by the Aluminum level for about 1747 mg kg⁻¹, but some characteristics were seen, such as retarded growth and metal toxicity (Figure 1a). If it is ignored, along with the time passing by, the metal content in the soil will keep increasing and become toxicity to the aloe vera.



Figure 1. Aloe vera. (a) indication of the succulent leaf, which is contaminated by heavy metals, (b) normal succulent leaves

Meanwhile, result of the research showed that the peat soil is identical to heavy metal contamination, and if it is planted with hyper-accumulator, it is expected that it is going to be the best medium for planting aloe vera. This has been supported by the yield of aloe vera without *Crotalaria* sp. that has plant height, number of succulent leaves, length or width of the succulent leaves, lower fresh and dry weights in comparison with the aloe vera by the treatment of planting hyper-accumulator. The increasing yield of fresh weight has reached 98-100% on the average (Figure 2).

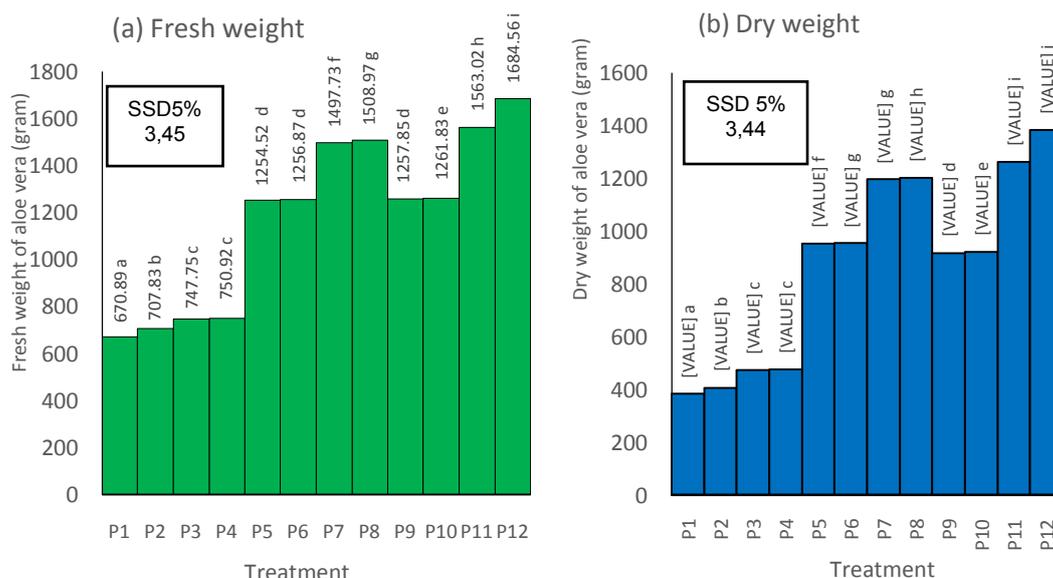


Figure 2. Graphic for fresh weight (a) and dry weight of the aloe vera (b)

- P1 : (Al: 1747 mg kg⁻¹, Pb: 112.51 mg kg⁻¹) + Aloe vera
- P2 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹) + Aloe vera
- P3 : (Al: 1064 mg kg⁻¹, Pb: 28.40 mg kg⁻¹) + Aloe vera
- P4 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera
- P5 : (Al:1747mg kg⁻¹,Pb:112.51 mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P6 : (Al:1479 mg kg⁻¹,Pb: 0.08mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P7 : (Al:1064mg kg⁻¹,Pb:28.40mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P8 : (Al:680 mg kg⁻¹,Pb:22.76 mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P9 : (Al:1747 mg kg⁻¹,Pb:112.51mg kg⁻¹)+ Aloe vera + Crotalaria juncea L.
- P10 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹)+ Aloe vera + Crotalaria juncea L.
- P11 : (Al: 1064 mg kg⁻¹,Pb:28.40 mg kg⁻¹) + Aloe vera + Crotalaria juncea L.
- P12 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera + Crotalaria juncea L.

Meanwhile, parameters of fresh weight and dry weight of gel and roots of the aloe vera, which were not planted with hyper-accumulator and planted with hyper-accumulator (*Crotalaria mucronata* or *Crotalaria juncea* L.), showed significant difference of the result on weight of both fresh and dry roots, as well as gel of the succulent leaves of aloe vera. Table 2 shows comparison between aloe vera, which is planted solely and planted with *Crotalaria mucronata* or *Crotalaria juncea* L. that showed different result in which the aloe vera, which was planted individually had the lowest yield of fresh and dry weight of the roots and the gel in comparison with the aloe vera, which was planted with hyper-accumulator (*Crotalaria mucronata* and *Crotalaria juncea* L.). This shows that the increasing number of hyper-accumulator, which has reduced the heavy metal level of Aluminum in the soil, would be able to increase the yield of fresh and dry weights of roots and gel, for about 2-to-4-fold (\pm 200%).

Table 2. Fresh weight and dry weight of the roots and gel of the aloe vera under diverse treatment of Aluminum content in the soil (g)

Treatment	Fresh weight		Dry weight	
	Root	Gel	Root	Gel
P1	30.22a	570.89a	11.53 a	303.55 a
P2	31.95ab	607.83b	13.84 ab	326.50 b
P3	34.35b	645.75c	14.35 bc	365.75 c
P4	34.63b	649.61c	16.63 c	369.61 d
P5	54.52c	1138.87d	30.73 d	745.53 e
P6	94.20e	1147.85e	72.20 f	747.85 e
P7	127.99g	1395.06h	101.99g	967.06 g
P8	139.57i	1412.90i	110.90i	1020.77h
P9	68.52d	1151.83f	47.79 e	748.49 e
P10	102.36f	1159.12g	75.09 f	760.45 f
P11	132.49h	1453.69j	105.81h	1085.55i
P12	141.03i	1584.56k	113.03i	1203.69 j
SSD 5%	2.67	3.45	2.64	3.42

Notes : Numbers followed by the same letter in the same column show insignificant difference based on SSD test at the level of 5%.

- P1 : (Al: 1747 mg kg⁻¹, Pb: 112.51 mg kg⁻¹) + Aloe vera
- P2 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹) + Aloe vera
- P3 : (Al: 1064 mg kg⁻¹, Pb: 28.40 mg kg⁻¹) + Aloe vera
- P4 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera
- P5 : (Al:1747mg kg⁻¹,Pb:112.51 mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P6 : (Al:1479 mg kg⁻¹,Pb: 0.08mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P7 : (Al:1064mg kg⁻¹,Pb:28.40mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P8 : (Al:680 mg kg⁻¹,Pb:22.76 mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P9 : (Al:1747 mg kg⁻¹,Pb:112.51mg kg⁻¹)+ Aloe vera + Crotalaria juncea L.
- P10 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹)+ Aloe vera + Crotalaria juncea L.
- P11 : (Al: 1064 mg kg⁻¹,Pb:28.40 mg kg⁻¹) + Aloe vera + Crotalaria juncea L.
- P12 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera + Crotalaria juncea L.

Subject that must to be concerned in relation to the use of aloe vera was the yield, particularly the succulent leaf, which contained heavy metals, should not be consumed by animals and human. Result of the analysis on aloe vera, which was planted solely, showed the aluminum value in the succulent leaf ranged 7.03-10.04 mg kg⁻¹, and the Lead ranged 0.202-0.552 mg kg⁻¹. This fact is really worrying and has beyond the normal threshold of the standard and criteria of the export quality. Meanwhile, the average value of Aluminum in gel of

the succulent leaf for planting the hyper-accumulator was about 0.01 mg kg⁻¹ and the Lead was about 0.02 mg kg⁻¹, which was far from the analysis result of heavy metal on the succulent leaf's gel of the aloe vera, which was planted solely (Table 3).

Table 3. The content of heavy metals in aloe vera (mg kg⁻¹)

Treatment	Aluminum Content		Lead Content	
	Gel	Root	Gel	Root
P1	10.04c	169.1 i	0.552 f	0.931 j
P2	9.07 bc	124.3 h	0.322 e	0.853 i
P3	8.05 bc	90.9 g	0.302 d	0.831 h
P4	7.03 b	80.8 f	0.202 c	0.731 g
P5	0.01 a	54.0 d	0.05 b	0.153 e
P6	0.01 a	52.3 cd	0.02 a	0.087 c
P7	0.01 a	51.2 bc	0.02 a	0.069 b
P8	0.01 a	48.9 b	0.02 a	0.044 a
P9	0.01 a	60.1 e	0.04 b	0.244 f
P10	0.01 a	57.6 e	0.02 a	0.131 d
P11	0.01 a	51.2 bc	0.02 a	0.100 c
P12	0.01 a	41.1 a	0.02 a	0.066 b
SSD 5%	2.56	2.56	0.017	0.017

Notes :Numbers followed by the same letter at the same age show insignificant difference based on SSD test at the level of 5%, insig. means insignificant.

- P1 : (Al: 1747 mg kg⁻¹, Pb: 112.51 mg kg⁻¹) + Aloe vera
- P2 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹) + Aloe vera
- P3 : (Al: 1064 mg kg⁻¹, Pb: 28.40 mg kg⁻¹) + Aloe vera
- P4 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera
- P5 : (Al:1747mg kg⁻¹,Pb:112.51 mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P6 : (Al:1479 mg kg⁻¹,Pb: 0.08mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P7 : (Al:1064mg kg⁻¹,Pb:28.40mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P8 : (Al:680 mg kg⁻¹,Pb:22.76 mg kg⁻¹)+ Aloe vera +Crotalaria mucronata
- P9 : (Al:1747 mg kg⁻¹,Pb:112.51mg kg⁻¹)+ Aloe vera + Crotalaria juncea L.
- P10 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹)+ Aloe vera + Crotalaria juncea L.
- P11 : (Al: 1064 mg kg⁻¹,Pb:28.40 mg kg⁻¹) + Aloe vera + Crotalaria juncea L.
- P12 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera + Crotalaria juncea L.

Some descriptions about contamination at the parts of the plant have been affected by planting medium. Based on result of analysis on the soil during harvest time (at the end of the research) that has been done in accordance with Ministry of State for Population Environment of Indonesia and Dalhousie, 2002 described that maximum range of heavy metal content in the soil for Aluminum was about 100-450 mg kg⁻¹. Average content of Al for treatment P1 to P4 (planting the aloe vera individually) was 1239.25 mg kg⁻¹ and aloe vera that has been treated with hyper-accumulator of Crotalaria mucronata, 347.57 mg kg⁻¹, and Crotalaria junceaL., 311.15 mg kg⁻¹. Concentration of more than 450 mg kg⁻¹ is categorized as contaminated, so that treatments P1 to P4 (planting aloe vera only) has been categorized as Aluminum (Al)-contaminated soil. Result of the analysis on pH soil showed that aloe vera, which was planted individually and together with hyper-accumulator plants of Crotalaria mucronata andCrotalaria juncea L. had different results. In which, treatment P1 to P4 (aloe vera only) had lower yield in comparison with treatments P5 to P12 (aloe vera and hyper-accumulator of Crotalaria sp.). pH of the soil under the treatment of P1 to P4 included 4.00, 4.20, 4.30, and 4.50. According to Fonte (2009), highly acid soil is < 5.00. So that, treatment P1 to P4 has been included as highly acid soil (Table 4).

Condition of the soil that has acid pH caused heavy metals in the soil would be soluble and actively being adsorbed by the plant, besides that, according to Barber in Connell and Miller (2005), the existence of heavy metals would limit the amount of phosphor, kalium, and iron in the root tissues that would inhibit the root growth and development of the meristem tissues. This is supported by Fitter and Hay (2001), in which the retarded growth of aloe vera was due to acid soil that is generally related to soil reaction. Low pH/highly acid may create a combination of Aluminum (Al) and Lead (Pb) toxicity. However, the most dominant effect and the worst cause of the retarded growth in aloe vera is Aluminum toxicity.

Table 4. Chemical analysis of the soil pre-and-post-research

Treatment	pH H ₂ O		Aluminum (mg kg ⁻¹)		Lead (mg kg ⁻¹)	
	Before planting	Harvest	Before planting	Harvest	Before planting	Harvest
P1	4.24	4.00 a	1747	1909.0 i	112.51	95.43 d
P2	4.41	4.20 a	1479	1402.6 h	0.08	4.48 b
P3	4.47	4.30 a	1064	1055.6 g	28.4	21.82 c
P4	4.75	4.50 a	680	589.8 f	22.76	22.55 c
P5	4.24	5.01 b	1747	440.7 e	112.51	6.06 b
P6	4.41	5.03 b	1479	371.8 d	0.08	0.03 a
P7	4.47	5.04 b	1064	306.7 c	28.4	1.30 a
P8	4.75	5.33 b	680	271.1 b	22.76	2.00 a
P9	4.24	4.99 b	1747	437.2 e	112.51	5.95 b
P10	4.41	5.05 b	1479	373.0 d	0.08	0.07 a
P11	4.47	5.06 b	1064	308.1 c	28.4	2.86 a
P12	4.75	5.71 c	680	126.3 a	22.76	1.89 a
SSD 5%		0.85		3.85		3.85

Notes :Numbers followed by the same letter at the same column show insignificant difference based on SSD test at the level of 5%.

- P1 : (Al: 1747 mg kg⁻¹, Pb: 112.51 mg kg⁻¹) + Aloe vera
P2 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹) + Aloe vera
P3 : (Al: 1064 mg kg⁻¹, Pb: 28.40 mg kg⁻¹) + Aloe vera
P4 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera
P5 : (Al:1747mg kg⁻¹,Pb:112.51 mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
P6 : (Al:1479 mg kg⁻¹,Pb: 0.08mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
P7 : (Al:1064mg kg⁻¹,Pb:28.40mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
P8 : (Al:680 mg kg⁻¹,Pb:22.76 mg kg⁻¹)+ Aloe vera +*Crotalaria mucronata*
P9 : (Al:1747 mg kg⁻¹,Pb:112.51mg kg⁻¹)+ Aloe vera + *Crotalaria juncea* L.
P10 : (Al: 1479 mg kg⁻¹, Pb: 0.08 mg kg⁻¹)+ Aloe vera + *Crotalaria juncea* L.
P11 : (Al: 1064 mg kg⁻¹,Pb:28.40 mg kg⁻¹) + Aloe vera + *Crotalaria juncea* L.
P12 : (Al: 680 mg kg⁻¹, Pb: 22.76 mg kg⁻¹) + Aloe vera + *Crotalaria juncea* L.

The ability to remediate the metal-contaminated soil, which is counted in accordance with the potential accumulative ability of Aluminum from the soil and adsorbed by the roots of *Crotalaria sp.* is the main key for the success of the remediation process. Based on the potential phytoremediation, it showed that the aloe vera breeding in such contaminated soil along with the planting of hyper-accumulator of *Crotalaria sp.* has been able to absorb the contaminant, so that it would increase the production of succulent leaves of the aloe vera, both physical appearance and metal content in gel of the succulent leaves of the aloe vera.

IV. Conclusions

The application of *Crotalaria sp.* has been able to bind heavy metals in the soil and high accumulative ability has been obtained under the treatment of P9 (Aluminum :1747 mg kg⁻¹, Pb:112.51 mg kg⁻¹) + Aloe vera+ *Crotalaria juncea*L.) is about 1309.8 mg kg⁻¹. The higher content of heavy metals in the soil is, the higher adsorption of Aluminum by the roots of *Crotalaria sp.*

The decreasing content of heavy metals in the soil would reduce the adsorption of heavy metals by the aloe vera, particularly at the part of succulent leaves, so that in accordance with the quality aspect of succulent leaf, the aloe vera has been able to meet the quality standard for the export. Value of the fresh weight of the succulent leaves at 120 days after planting along with the treatment of *Crotalaria juncea* L., is about 1584.56 gram blade⁻¹

Different types of hyper-accumulator (*Crotalaria sp.*) show different adsorption of different metal. *Crotalaria juncea* L. shows better ability than *Crotalaria mucronata*. It has been proven by high adsorption of Aluminum by the roots of *Crotalaria juncea*L. (P9) is about 2986.5 mg kg⁻¹ (90%) by the adsorption of Lead is about 4.73 mg kg⁻¹ (70%). High adsorptive ability by the roots of *Crotalaria juncea* L. will reduce the adsorptive ability by the roots of aloe vera, Al: 41.1 mg kg⁻¹, Pb: 0.066 mg kg⁻¹ and metal adsorption in gel succulent leaves of the aloe vera is Al: 0.01 mg kg⁻¹ and Pb: 0.02 mg kg⁻¹. Also, these values are lower in comparison with the aloe vera without accompanied with *Crotalaria sp.* (P1), in which Al content in gel of the aloe vera is about 10.04 and Pb is 0.5 mg kg⁻¹

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