Growth and Accumulation Of Pb In Native Hymenachne (*Hymenachne acutigluma)* In Tailings Area As Affected By Organic and Inorganic Fertilizer

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***Abstract*—Tin mining in the Islands of Bangka and Belitung left behind a vast area with low fertility. The tailings area of ex-tin mining is different from common soil both in terms of mineral and chemical compositions, with characteristics such as the unstable geochemical aspect, extreme pH value, cation-exchange capacity, low organic matter, and heavy metals pollution. Native hymenachne (*Hymenachnes*p) is an invasive type of grass. Invasive vegetation could dominate the layers of soil and change the structure, composition, and the ecosystem's ecological function. Invasive grass can compete in the terms of utilization of water, nutrients, and light. Native hymenachne (*Hymenachne acutigluma* (Steud)) is a type of perennial grass that has been used for long time by the local people in America, Australia, and Asia since it has good economic value as high-nutrient feed. The existence of this plant in the tailings areas have not been vigorously studied yet. Thus this study was aimed to determine the growth rate and accumulation of Pb in native hymenachne as affected by organic or inorganic fertilizer. The results showed that the hymenachne grew better after treatment with natural fertilizer and nitrogen. The accumulation of Pb at the tip of the leaves before and after treatment with organic and inorganic fertilizers was beyond the safe limit for feed. At the early utilization stage of tailings area after tin mining, native hymenachne (*Hymenachne acutigluma* (Steud)) has the potentials as pioneer vegetation and as phytoaccumulation of Pb. However, the utilization of hymenachne as animal feed has to consider the level of Pb in it if they come from tailings area, where the safe level of Pb in vegetables and its products based on SNI 2009 should not be over 0.5 mg/kg.**

Keywords— Tin mining, Accumulation, Native grass, Hymenachne acutigluma, Tailings

I. INTRODUCTION

Mining activity often leaves damage to the soil and affects the vegetation that causes damage to the area [1]. Tinmining in Bangka and Belitung islands has left behind a vast area of tailings with low fertility. Based on the mineral and chemical compositions, tailings area is different from common soil, due to the unstable geochemistry and extreme toxicity. Other effects of mining is heavy metals pollution, extreme pH value, cation-exchange capacity, and low organic matter [3].

Efforts are needed to restore the ecological need of the ex-mining areas, including the management of physical, chemical, and biological aspects of the soil, such as soil pH, fertility, and microbiological community and variety of soil nutrients, which had degraded the productivity of the soil [1]. The existence of heavy metals in agricultural area has to be paid attention to since they are toxic and lethal in certain dosage and one of the heavy metals is tin (Pb). Other than affecting the health of humans and animals, absorption and translocation of Pb in plant tissue can also cause toxic effect that decreases the production of biomass. However, several types of plants could stand in the condition of high Pb level, thus they can be used to reduce the level of Pb (phytoremediation). The plants used in the strategy of tailings reclamation should be able to survive in low nutrient condition and high environmental stress; they should also be able to absorb, transform, and store contaminants and to promote degradation of organic contaminant through the activity of rhizoferous microorganisms [4].

Native hymenachne (*Hymenachne sp)*is a native grass from Sumatera dan Kalimantan that is invasive. *H. amplexicaulis* has interesting agronomic characteristics, including high nitrogen and protein content (13.9 – 15.8% d.m.), high production of green materials, and the ability to survive in dry season [9].

Treatment with organic matters on *H.pernambucense* was shown to have positive effect on the production of dry matters [10]. Organic fertilizers could influence the physical, chemical, and biological aspects of the soil. Moreover, organic fertilizer also has significant effect on crude protein, phosphor, and digestibility of organic matters of the grass (*H.amplexicaulis*[11]. A study showed that application of organic matters on plants could affect the plant growth and its ability to yield high biomass and support proliferation and activity of microorganisms [4]. The productivity of tailings area could be improved by adding natural fertilizer such as compost that could act to enhance the activity of microorganisms to provide nutrients (N, P) and organic carbons in the soil [1]. The utilization of grass plants *Thysanolaena maxima* and *Vetiveria zizanioides* in mining areas has been combined with the use of organic matters, where they could show positive effects in terms of absorption of Pb in the tailings area [12].

In order to support the growth of plants in areas with low nutrients, inorganic fertilizer containing N, P, and K is needed; moreover, the fertilizer has to be able to release those minerals quickly. For short term, the initial storage of nutrients is provided by the inorganicfertilizer [13]. A study showed that addition of lime could help to produce dissolved organo-metal complex with dissolved organic carbon (DOC), which can be easily absorbed by the roots of the rye grass (*Lolium temulentum*) [14].

The utilization of hymenachne grass as pioneer vegetation is possible due to its invasive characteristic that is supported by the supply of nutrients from inorganic fertilizer and pH improvement from lime as well as from addition of organic matters that could provide physical, chemical, and biological improvement of the soil, including reduction of contamination of heavy metals; therefore, it is expected that there would be economical value of the grass in the future.

II. METHODS

A. Materials And Methods

The tailings area selected in this study was used to be a tin mining area that had been left 20 years ago. The organic matter used was mixed compost from cow's manure, hay, and rice bran, with effective addition of microorganism and inorganic fertilizer with 15% N, 25% P, and 15% K, agricultural lime or dolomite, and plantation media forhymenachne that was collected from the area surrounding the campus of Bangka Belitung University, BalunjukVillage, Merawang District. The tools used were hoes, spades, balances, and double-layer polybags sized 20 x 25 cm and 25 x 30 cm. The inner layer of the polybags was punctured with six holes at the bottom part and two holes at the sides. The position of the side hole was 5 cm from the ground level. The outer layer of the polybag had holes only on the sides with the set at the height of 15 cm from the ground level and 4 holes at each side. There were also watering cans, weed pullers, stationery, and laboratory equipments needed for the analysis of Pb.

The research was started with the analysis of Pb before the hymenachne was planted. The young grass plant used was 15-cm height and each polybag was planted with 5 young plants. The treatments were as follow: t0 = 100 % of tailings soil, t1=tailings soil with addition of NPK (15:15:15) at 100 kg/ha (6 g per polybag) that was given at 2 weeks after planting, t2 = tailings soil with addition of organic matters at 5% of the total volume of the tailings media in each polybag (600 mL per polybag) that was given 1 week after planting,, t3 = tailings soil with addition of lime and NPK (lime at 6.8 tones/ha and 100 g of NPK/ha) that was given 1 week prior to planting, and t4 = tailings soil with addition of organic matter (5% compost of the total media per polybag) and NPK (100 kg/ha). Care was given to the grass plants by manual watering and weeds removal. Parameters of the grass growth included height, number of young plants, wet mass of the grass, dry mass of the roots, total dry mass, index of chlorophyll, and specific area of the leaves. Accumulation of Pb was determined by analyzing the mineral using the Morgan method and atomic absorption spectroscopy (AAS). Level of Pb was analyzed at the initial and end points of the planting media as well as in the grass at135daysafter planting; while concentration of bioaccumulation was calculated from the ratio between the concentration of Pb in the grass and in the soil, and accumulation of grass (mg/g) was determined from the ratio between Pb in the grass and the total weight of plants.

B. Data analysis.

The experimental design used in the study was completely randomized design with 5 treatments. To get the best treatment, post-hoc Duncan test was calculated. Correlation coefficient (r) was used to see the relationship among growth factors. If the r value was smaller than 0.5, the correlation value of the variables was considered low. Meanwhile, determinant coefficient value (r^2) was used to judge the effect of grass height, number of young plants, chlorophyll index, dry mass of the grass, dry mass of the roots, and specific area of the leaves on the parameter of total dry mass of the plants.

III. RESULTS

A. Growth of hymenachneintin tailings area

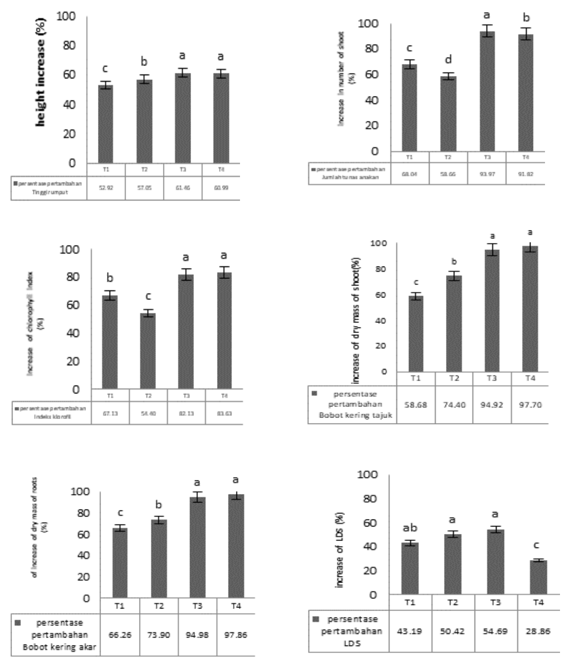


Fig. 1. The percentage (%) of increase of growth of the grass due to accumulation from t1, t2, t3,, and t4 as compared to control (t0) Notes: types of plantingmedia:t1=tailings with addition of inorganic fertilizer NPK (15:15:15) 100kg/ha, t2=tailings with addition of organic matters 5% of total volume of tailings soil/polybag, t3=tailings with addition ofLime+NPK (lime 6,8 ton /ha and 100kg NPK/ha), t4= tailings with addition of organic matter and inorganic fertilizer NPK (compost of 5% of total volume of media/polybag) and NPK (100kg/ha). SAL= specific area of leaves, as compared to t0 (tailings with no additional treatment). Different alphabets in a graph and histogram shows significant differences of Duncan post-hoc test at 0.05.types of planting media: t1 = t2 = tailings soil with addition of inorganic fertilizer at 5 % of the total volume of the tailings per polybag.

Increased growth as affected by the addition of inorganic fertilizer in the form of NPK (t1), organic matters in the form of compost (t2), addition of dolomite along with addition of inorganic fertilizer of NPK (t3), and addition of organicfertilizer combined with NPK (t4). Addition of organic matters combined with NPK fertilizer gave the highest dry and wet masses of the hymenachne grass as compared to other treatments (Figure 1)

Control treatment was treatment with tailings soil without any fertilizer (t0). There was increase in grass height due to addition of inorganic fertilizer NPK (t1) at 52.92 %. Next, the grass height would increase more at 57.05% if the inorganic fertilizer NPK was replaced with organic compost (t2). Similar trend was also shown in the parameters of dry mass of grass, dry mass of roots, and specific leaves area. Slightly different trend was seen in the parameters of number of young plants or sprouts and chlorophyll index where inorganic fertilizer NPK (t1) could improve the number of young plants and chlorophyll index better if compared with the treatment with addition of organic matter (t2).

B. *Coefficient of correlation and determination coefficient between parameters of growth of the hymenachne grass at the tailings of tin mining*

Based on the coefficient correlation (r) in Table 1, in treatment t0, the parameters that had strong relationship with the total dry mass of the grass were chlorophyll index and dry mass of the grass and roots.

TABLE I. DETERMINATION COEFFICIENT VALUE (R^2) AND CORRELATION COEFFICIENT (R) OR TOTAL DRY MASS WITH GRASS HEIGHT, NUMBER OF YOUNG PLANTS, CHLOROPHYLL INDEX, DRY MASS OF GRASS, DRY MASS OF ROOTS AND SPECIFIC LEAVES AREA OF THE HYMENACHNE GRASS

Parameter	*values*	**Total dry mass of hymenachne grass**				
		t0	*t1*	*t2*	*t3*	*t4*
Grass height	r^2	0.108	0.027	0.156	0.381	0.124
	r	0.329	0.164	0.395	0.617	0.352
Number of young plants	r^2	0.033	0.751	0.025	0.316	0.819
	r	0.182	0.867	0.158	0.562	0.905
Chlorophy ll iindex	r^2	0.874	0.209	0.458	0.458	0.37
	r	0.935	0.457	0.677	0.677	0.608
Dry mass of grass	r^2	0.,982	0.102	0.428	0.858	0.984
	r	0.991	0.319	0.654	0.926	0.992
Dry mass of roots	r^2	0.994	0.224	0.883	0.958	0.984
	r	0.997	0.473	0.940	0.979	0.992
Specific leaves area	r^2	0.133	0.935	0.446	0.572	0.274
	r	0.365	0.967	0.668	0.756	0.523

Notes: r2= values of determination coefficient (r^2) and correlation coefficient (r), t0= 100%tailings soil, t1=tailings with addition of NPK (15:15:15) 100kg/ha, t2= tailings with addition of organic matter 5% from the total media of tailing/polybag, t3=tailings with addition of lime+NPK (lime 6,8 ton /ha and 100kg NPK/ha), t4= tailings with addition of organic matter and NPK (5% compost of the total media/polybag) and NPK (100kg/ha).

Similar trend was also seen in t2 and t3, while plant height, number of young plants and specific leaves area did not have correlation with the total dry mass of the plant. In the treatment t1, the parameters of number of young plants and specific leaves area had stronger relationship with total dry mass of the grass, while in treatment t4, almost all growth parameters had strong relationship with total dry mass of the plant, except with grass height.

Based on the high determinatiion coefficient, in treatment t0, the roots dry mass was the most influential in affecting the total dry mass. In treatment t1, the total dry mass was seen to be affected by the specific leaves area. Meanwhile, the treatment t2 showed total dry mass that was affected by roots dry mass; while t3 showed total dry mass that was affected by specific leaves area. Moreover, in t4, it was shown that number of young plants had strong effect on total dry mass (Table 1).

C. *Concentration of Pb in the tailings and hymenachne grass*

Table 2 shows the concentration of Pbin the hymenachne grass, as affected by several treatments of combination of the tailings. The trend showed decrease of Pb after planting of the grass. Concentration of Pb in the grass of the hymenachne was 9.05 ppm and the highest level was found in t4. In Table 2, all treatments are shown to have high Pb concentration more than the national standard of SNI 2009 for lead in vegetables and its products that should not be more than 0.5 mg/kg.

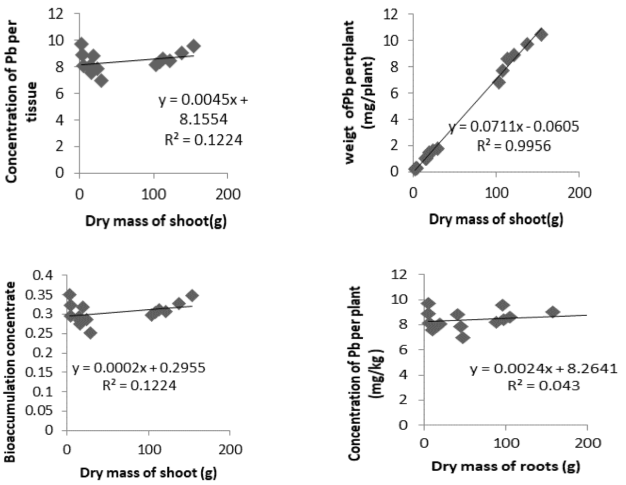
TABLE II. AVERAGE CONCENTRATION OF PB AFTER 135 DAYS OF PLANTING, PB CONTENT OF THE ROOTS, AND PB CONTENT OF THE HYMENACHNEGRASS AS AFFECTED BY THE TREATMENTS

Treatment (type of media)	Final Pb content of the tailings (mg/kg)	Concentration of Pb in grass (mg/kg)	Mass of Pb in the grass (mg/plant)	Bio accumulation concentration (BAC)
t0	6.80	8.91	0.27	0.32
t1	6.40	7.85	1.00	0.28
t2	6.80	7.87	1.65	0.29
t3	7.10	8.40	7.69	0.30
t4	7.08	9.05	9.75	0.33

Notes: t0= 100% tailings, t1=NPK (15:15:15) 100kg/ha, t2= organic matters at 5% of total volume of tailings/polybag, t3=lime+npk (lime 6.8 ton /ha and 100kg npk/ha), t4= organic matters and NPK (compost at 5% of total volume of media/polybag) and NPK (100kg/ha).

The determination coefficient (r^2) of dry mass of grass and dry mass of roots on the concentration of Pb in the tissue (mg/kg), the level of Pb per plant (mg/plant) and bioaccumulation concentration of the hymenachne grass in the tailings media are all shown in Figure 2.

The correlation value (r) of the grass dry mass and roots dry mass with the level of Pb in grass and bioaccumulation concentration (BAC) waslower than 0.50; thus, it was concluded that there was no significant relationship between increase of grass dry mass and roots dry mass on the level of Pb in the plants (mg/kg) and BAC, although the correlation tended to show positive value. However, based on the value of determination coefficient of dry mass of grass and roots on the level of Pb in the plant, the value of r^2 was 99.5%. This value indicated that the larger the grass, the more Pb was accumulated in the grass tissue. Similarly, roots dry mass would affect the level of Pbin the plant as much as 76.9%. This phenomenon indicated the absorption of Pb from the tailings media by the roots that was delivered to the grass of the hymenachne.



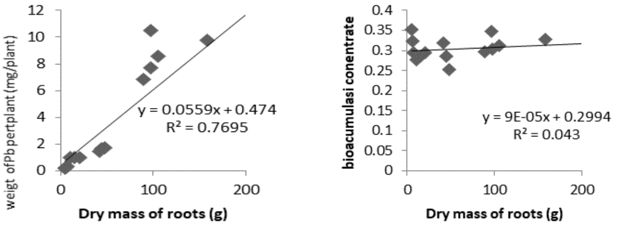


Fig. 2. The values of determination coefficient (r^2) of grass dry mass and roots dry mass with concentration of Pb in the tissue (mg/kg), level of Pb in the plant (mg/plant) and bioaccumulation concentration of the hymenachne grass in tailings media

IV. DISCUSSION

A. *Growth rate of the hymenachne grass in tailings area of tin mining*

Growth of the grass in the tin tailings was affected by the treatment given to the tailings. All variables showed that at 100% tailings (t0), the growth was low. Until 135 days of plantation, the hymenachne grass could still survive at 100% tailings (t0) with low growth as compared to other treatments. Addition of organic and inorganic matters showed tendency of different growth rates. The role of organic matter was better for the height, dry mass of grass, and dry mass of roots; however, the effect of the inorganic fertilizer NPK was more dominant on the increase of number of young plants, chlorophyll index, NLD, and NBD (Figure 1).

Addition of lime with inorganic fertilizer NPK (t3) could increase the plant height, the number of young plants, and specific leaves area as compared to the use of the fertilizer NPK combined with organic matter (t4). However, the replacement of lime with organic matters (t4) was relatively better in improving the chlorophyll index, dry mass of grass, and dry mass of roots (Figure 1). This phenomenon was due to the addition of organic matter that could improve the physical and biological characteristics of the media; moreover, addition of nutrients also helped improve the growth of the plants. Availability of nutrients from the organic matter was relatively longer than the inorganic fertilizer due to the slow release characteristics of the inorganic fertilizer.

Organic fertilizer (t2) was generally more able to improve the grass dry mass and roots dry mass in tailings media by 20% as compared to the inorganic fertilizer (t1). Improvementof tailings condition with the application of organic matter could help improve the production of dry biomass and level of chlorophyll of rye grass grown in tailings area [14]. Single application of inorganic matter of NPK (t1) would be better if accompanied with lime (t3) or organic matter (t4). Improvement of dry mass of grass would increase from 58.6% to 94.9% with the addition of lime (t3) and 97.7% with the addition of organic matters (t4). Roots dry mass increased from 56.26% to 94.8% when NPK was added with lime (t3) and 97.8% if NPK was added with organic matters (t4). Table 1 shows that in treatment t0, the variable that had strong relationship with the increase of total dry mass of the hymenachne grass were dry mass of grass, dry mass of roots, and chlorophyll index. Meanwhile, the plant height, the number of young plants, and specific leave area did not affect improvement of total dry mass of grass in treatment t0. In treatment t1, the variable that affected grass dry mass were specific leaves area and number of young plants. In treatment t2, total dry mass of the grass was affected only by the increase of roots dry mass

Addition of organic matter in treatment t2 could only improve the growth of roots that significantly affected the improvement of total dry mass of the plants. Addition of lime and the NPK fertilizer (t3) could improve the grass dry mass, roots, and leaves. However, it was not significantly affected the number of young plants. Meanwhile, the use of NPK fertilizer and organic matters (t4) showed an increase of total plant dry mass that was affected by the number of young plants (r2=0.819) and dry mass of grass and roots (r2=0.984). Increase of number of young plants, dry mass of grass, and dry mass of roots in treatment t4 could affect the increase of total dry mass of the plants in the tailings media. This phenomenon was due to the addition of organic and inorganic fertilizers that could improve the chemical and microbiological characteristics of the tailings and the results showed the trend of significant improvement of plant yield.

The correlation value (r^2) between plant height and total dry mass was lower than 0.50, which showed high tendency of hymenachne grass to correlate negatively with the increase of total dry mass of the plants in all treatments. This phenomenon was due to the increase of length of grass due to horizontal growth of the grass that made it compact, such as in the Rhodes grass. Increase of plant length was not followed by the increase of young plants and number of leaves, which showed that the growth parameter did not significantly affect the observed parameters [16]. Organic amendment in tailings area was especially used to trigger micro-aggregation of tailings through organo-mineral interaction since there was an increase in the organic matter (C) that was related with the aggregate [17]. The number ofyoung leaves with r^2 value of more than 0.5 was seen in treatments t1 and t4. Addition of inorganic fertilizer NPK (t1) and addition of organic fertilizer combined with NPK fertilizer (t4) showed the relationship between the increase of number of young plants with the increase of total dry mass of the plants. The relationship between the number of young plants of hymenachne grass with the total dry mass of the plants was lower than 0.5 for the treatments t0, t2, and t3. This results showed that growth of young plants due to addition of organic matter and lime in tailings of tin mining did not significantly affect the increase of total dry mass of the hymenachne grass.

Based on the correlation coefficient (r) shown in Table 1, in treatment t0 , the variables that had strong relationship with total dry mass of the hymenachne grass were chlorophyll index, dry mass of grass and dry mass of roots. Similar trend was also seen in t2 and t3, while plant height, number of young plants, and specific leaves area did not have strong relationship with the total dry mass of the plant. In treatment t1, the variables of number of young plants and specific leaves area were seen to have relationship with total dry mass of the grass, while in treatment t4, almost all growth variables had strong relationship with total dry mass of the plant, except for grass height. The combination between organic and chemical fertilizers given to the tailings area could improve the chemical and microbiological characteristics of tailings, which lead to increase of plant yield.

B. *Level of Pb in the hymenachne grass grown in tailings area of tin mining*

Table 2 shows that plants that were treated with organic matters had high level of Pb, especially samples of t3 and t4. Addition of compost, lime, and commercial fertilizer could help improve the poor condition of mine tailings [14]. The larger and the more number of leaves, the higher the level of Pb that could be absorbed. Absorption of Pbis affected by dry mass of the plants (Figure 2). Neutralization of tailings area with lime and the application of organic lime could improve the biomass production and level of chlorophyll in rye grass. Addition of lime combined with biosolid decreased the toxicity level of Cu in rye grass (*Lolium temulentum*) [14].

In the hymenachne grass, the larger the biomass, the more risk of the plant to have high level of Pb in the tissue. Iincrease of dry mass of the grass was affected by the growth of the plants; thus, grass with faster growth would have more Pb in its tissue. The growth of the grass in the treatments t3 and t4 affected not only the dry mass of the grass and the roots, but also the increase of total dry mass that was also caused by the number of young plants, chlorophyl index, and leaves area. In the treatmentt2, the number of sprout or young plants did not significantly affect the total dry mass of the hymenachne grass that was planted in the tin mining tailings (Table 1). The correlation between dry mass of the grass with the concentration of Pbin the tissue (mg/kg), the concentration of Pb in the grass (mg/plant), and the bioaccumulation concentration (BAC) of Pb was determined from the coefficient value (r^2) (Figure 2).

The increase of Pb level was caused by the increase of dry mass of each grass unit. The more number of leaves and the larger the leaves, thus the more Pb was absorbed. Absorption of Pb was affected by dry mass of the plant. Most of heavy metals are stored in the stalk of a plant since it has high biomass. Compost had positive effect on metals (especially in reduction of concentration of Cu) with the support of phytoremediation capacity of *Brassica juncea* L. [3]. The use of organic matters could help to providenutrients, increase cation-exchange capacity andsoil porosity, and improve soil conditionthroughimprovement of microorganism activity that could fasten degradation process of the contaminants [4]. Plants that are tolerant to heavy metals are considered effective to treat soil that is contaminated with heavy metals and acid [1].

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