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Effects of Soil Amendments on the Growth and Total Phenolic Content in *Labisia Pumila* var. *Alata* at Nursery Stage

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Abstract* – *Labisia pumila* is an important herbal plant belonging to the Primulaceae family. Traditionally, the water decoction of this herb was used for pre and post-partum treatment by the Malay women. *L. pumila* has the ability to regulate body weight, prevent photo-aging and has anti-bacterial and anti-fungal effects. As natural supplies of this herb are dwindling due to over exploitation, it is imperative that attempts be made to cultivate this local popular herb. Thus, it is important to determine the best agronomy practices of this herb which may assist to increase the yield and phythochemical compounds. Fertilizer applications are essential in order to supply the important nutrients that increase growth, vigour, yield and maintain the health of plants. Besides optimum growth, it is important to identify how plant nutrition can increase the phytochemical compounds of *L. pumila.* Maintenance and/or enhancement of phytochemicals present via fertilization may garner high quality plants for production of phythochemical compounds. The objectives of this research were to determine the effects of different types of soil amendments on 12 months *old L. pumila* at nursery stage. Seven treatments were used in this study i.e. i) Inorganic fertilizer (NPK Green 15: 15: 15), 90 kg N/ha (T1) (positive control); ii) Inorganic fertilizer (NPK Green 15: 15: 15), 90 kg N/ha + Biochar, 5% of media (T2); iii) Controlled release fertilizer (AJIB NPK Granular 10: 15: 17: 2: 5), 90 kg N/ha (T3); iv) Controlled release fertilizer (AJIB NPK Granular 10: 15: 17: 2: 5) + Biochar, 5% of media (T4); v) Controlled release fertilizer (AJIB NPK Granular 10: 15: 17: 2: 5) + Compost (T5); vi) Controlled release fertilizer (AJIB NPK Granular 10: 15: 17: 2: 5) + Compost + Biochar, 5% (T6); and vii) Without fertilizer (negative control). The findings showed that T5 gave the best growth performances in terms of stem height, leaves number, leaves width, leaves length and collar diameter. Whereas, for total phenolic content, T6 gave the highest value of 310 mg/100 GAE followed by T5 with 202 mg/100 GAE. In conclusion, soil health plays important roles in growth and phytochemical response of *L. pumila* at nursery stage.

***Keywords* – Fertilizer, Biochar, Total Phenolic Content, Growth Performance, Planting Stock.**

I. Introduction

Labisia pumila is an important herbal plant belonging to the Primulaceae family. *L. pumila* var. *alata* is one of the variety that mostly used by researchers in their research as it was abundant in Malaysia (Abdul Kadir et al. 2017; Mannerås et al. 2010; (Shuid et al. 2011). Traditionally, the water decoction of this plant was used for pre and post-partum treatment by the Malay women (Jamia 2006). *Labisia pumila* has the ability to regulate body weight (Fazliana *et al*. 2009), prevent photo-aging and has anti-bacterial and anti-fungal effects (Karimi *et al*. 2011; Ali & Khan 2011). As natural supplies are dwindling due to over exploitation, it is imperative that attempts be made to increase the production of planting stocks by cultivate this local popular herb. Thus, it is important to determine the agronomy practices of this herb which may assist to increase the growth and *L. pumila* quality.

For this purpose, nutrients needed by the plants must be supply in correct quantity, proportion and in usable form at the right time. Soil amendments and fertilizers are a wide array of materials could be added to topsoil to improve plant growth. It can be organic and inorganic. Inorganic fertilizer such as NPK are essential nutrients that must be supplied to the plants to increase growth, vigour, yield and maintain the health of plants. Besides, soil conditioners such as biochar and compost will help to improve soil structure. Biochar is as carbon-rich by product resulting from pyrolyzing biomass (Biedermen & Harpole, 2013) will help to ameliorate the soil fertility and plant growth (Schulz & Glaser, 2012). In addition, this study have tried use of improved fertilizer technologies such as controlled release fertilizer (CRF) to minimize the nutrient loss and to obtain optimum growth of *L. pumila* so that more economically viable once it is cultivated commercially. For medicinal plants like *L. pumila*, phytochemical compounds such as phenolics and flavonoids are the two main economical products that made this herb attractive for medicinal purpose. In the present study, seven treatments of soil amendments were applied on 12 months *L. pumila* var. *alata* plants and the effects of these treatments to the growth and total phenolic content (TPC) of *L. pumila* var. *alata* at nursery stage were investigated.

II. Materials and Methods

A total of 12 plants at the age of 12 months derived from tissue culture technique were transferred into 8’ x 10’ polybags (3 kg) in September 2016. Growing media used were topsoil, sand and coconut husk (3:1:1), whereby 7 treatments were represented by 2 replications for observation purposes. The seven treatments applied for every five months were:

1. Inorganic fertilizer (NPK Green 15:15: 15), 90 kg N/ha (T1) (positive control).
2. Inorganic fertilizer (NPK Green 15:15: 15), 90 kg N/ha

+ Biochar, 5% of media (T2).
3. Controlled release fertilizer (AJIB NPK Granular 10: 15: 17: 2: 5), 90 kg N/ha (T3).
4. Controlled release fertilizer (AJIB NPK Granular 10: 15: 17: 2: 5)+ Biochar, 5% of media (T4)
5. Controlled release fertilizer (AJIB NPK Granular 10: 15:17:2:5) + Compost (T5);
6. Controlled release fertilizer (AJIB NPK Granular 10: 15: 17: 2: 5) + Compost + Biochar, 5%
7. Without fertilizer (negative control).Stem height (cm), leaves number, leaves width (cm), leaves length (cm) and collar diameter (mm) were measured at interval of 3 months using a digital calliper and a standard ruler. At the end of the study, the initial data measurement and the 12th month measurement were summarized using the relative growth rate (RGR) equation as below for stem height (cm), leaves number, leaves width (cm), leaves length (cm) and collar diameter (mm). Analysis of variance (ANOVA) was also conducted to compare the means for each treatment. The total phenolic content (TPC) of the plants was also determined using Folin Ciocalteu method (Singleton & Rossi 1965). RGR = $[\ln (M_o - M_t)]/ t$
M_o = Data at initial M_t = Data at final t = time (months).

III. Results and Discussion

Figure 1 showed that T5 [CRF + Compost] gave high effects to the stem height of *L. pumila* var. *alata* (0.17 cm) compared to control (0.06 cm). *Labisia pumila* var. *alata* plants applied with NPK (T1) gave low value of relative growth rate for stem height (0.07 cm). However, there is an increment of 0.03 cm when added biochar with normal fertilizer, NPK. It is expected that the addition of biochar to normal fertilizer treatments will increased plant productivity. This is because biochar has the potential to improve water-holding capacity of soils and increased plant growth with the addition of fertilizers (Woolf *et al*. 2010). Whereas, treatments T3, T4 and T6 showed no significance differences at $P < 0.05$.

Figure 2 indicated that T5 [CRF + Compost] gave high relative growth rate in number of leaves for *L. pumila* var. *alata* followed by treatments, T3 [CRF] and T6 [CRF + Compost + Biochar]. The results showed that *L. pumila* var. *alata* applied with CRF gave positive effects to the growth and increment of leaf number compared to plants grown with NPK fertilizer. It is recommended to have high number of leaf for the species because it can increase biomass rate (fresh and dry) during harvesting period. Similar results were also found for the traits of leaf length (cm) and leaf width (cm) of *L. pumila* var. *alata* (Figure 3 and 4). Treatment T5 [CRF + Compost] gave significant effects to leaf length (0.11 cm) and leaf width (0.12 cm) compared to control and other treatments. Treatment 1 and 7 (positive and negative control) gave the lowest value for relative growth rate especially in leaf length and leaf width.

In terms of collar diameter, result indicated that T5 [CRF + Compost] gave the highest value of 0.08 mm, even though it has no significant different with T3 [CRF] and T6 [CRF + Compost + Biochar] (Figure 5). The low values of RGR for collar diameter showed by T7 (negative control), followed by T1, T2 and T4, respectively at 0.05.

From the findings, it was found that application of controlled release fertilizer (CRF) into growing media of *L. pumila* var. *alata* enhanced the growth of this herb. The effects of CRF were also prominent compared to control as the nutrients are released gradually due to the semi-permeable coating in line with plant growth. High effects are seen when compost was added with CRF. It is believed that, nutrients released from CRF were retained and strongly hold by compost due to its physical structure of compost with tiny holes and capable to holds air, moisture and nutrients (Dorte, 2004) and thus, make it available for roots of *L. pumila* var. *alata* plants to absorb the nutrients. This incorporation reduce the nutrient losses into environment and increase the nutrient efficiency.

Beside relative growth rates, analysis of total phenolic content (TPC) was also conducted in this study. Results found that *L. pumila* var. *alata* leaves extract from T6 [CRF + Compost + Biochar] gave the highest value of TPC (310.2 mg/100g GAE) and significantly different compared to other treatments (Figure 6). This is possibly due to the nutrients (especially nitrogen) released by CRF, compost and biochar were in small portions compared to normal fertilizer, NPK. According to Li et. al (2008), high nitrogen supply could decreased the TPC whereas sulphur supply may increase the TPC of the plants. Furthermore, it believed that other nutrients incorporated in the controlled release fertilizer such as magnesium and micronutrients might also enhanced the TPC values. Micronutrients are known to assist in the phenol metabolism of plants (Marschner 1995).

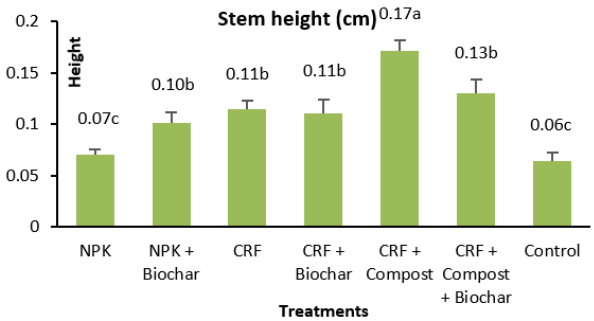

Fig. 1. Effects of different soil ammendments on the relative growth rate of stem height (cm) of *Labisia pumila var. alata*

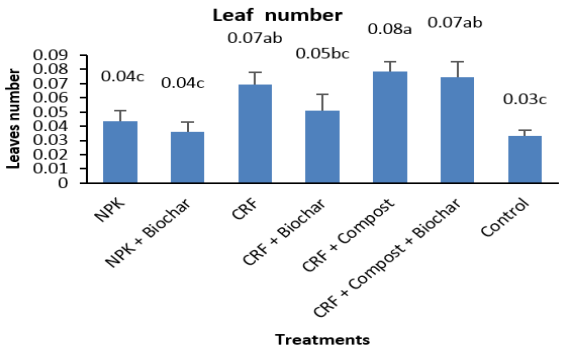

Fig. 2. Effects of different soil ammendments on the relative growth rate of leaf number of *Labisia pumila var. alata*

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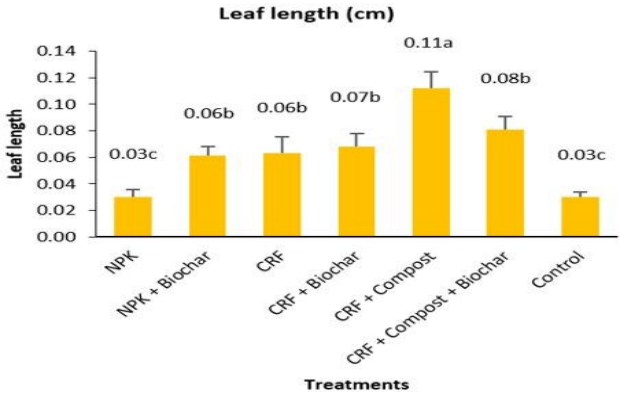

Fig. 3. Effects of different soil ammendments on the relative growth rate of leaf length (cm) of *Labisia pumila var. alata*

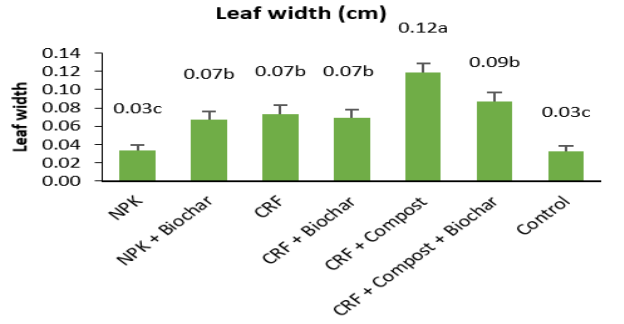

Fig. 4. Effects of different soil ammendments on the relative growth rate of leaf width (cm) of *Labisia pumila* var. *alata*

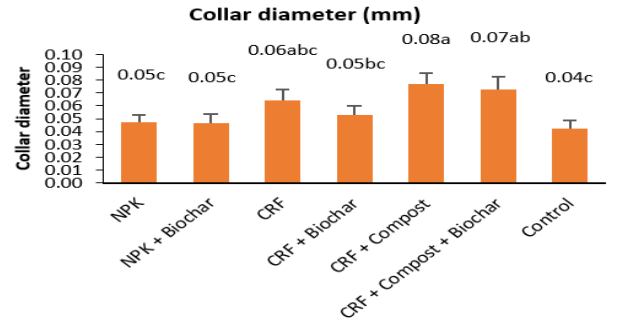

Fig. 5. Effects of different soil ammendments on the relative growth rate of collar diameter (mm) of L. pumila var. alata

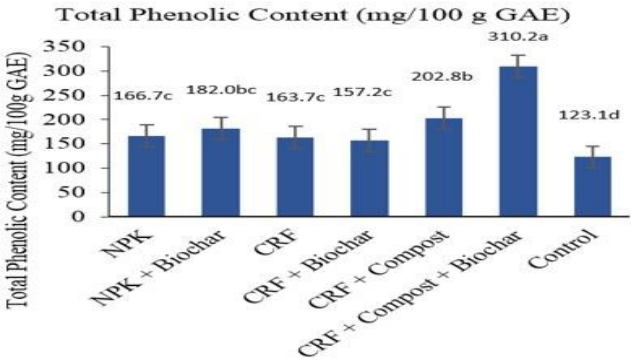

Fig. 6. Effects of different soil ammendments on the total phenolic content of Labisia pumila var. alata

IV. Conclusion

In general, findings from this study showed that T5 [CRF + Compost] promotes good growth performances in *L. pumila* plants due to the increasing relative growth rate compared to control and other treatments. Total phenolic content of *L. pumila* plants were also enhanced after adding biochar into CRF and Compost (T6). Further investigation should be conducted on chlorophyll level and gallic acid compound to develop better understanding on the growth requirement of *L. pumila* var. *alata*. It is also concluded that soil health plays an important role in environmental sustainability and growth response of the plants.

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Norhayati, S. was born on 13th June 1989 in Teluk Intan, Perak. She gained admission to Kolej Kolej Marikulasi Negeri Sembilan, Kuala Pilah, Negeri Sembilan for one year on 2007-2008. Then she further her study in Bachelor of Agricultural Biotechnology at Universiti Sultan Zainal Abidin (UniSZA), Terengganu. On 2012, she pursued her study at UiTM Shah Alam in Master of Science, Agro-Biotechnology. At the same time, she was joined Forest Research Institute Malaysia (FRIM) as a Research Officer in Herbs and Tree Improvement Branch, Forest Biotechnology Division, 52109, Kepong, Selangor from 2012 until recent. There are a lot of publications and works regarding conservation, propagation, and improvement of herbal Kacip Fatimah that have been done together with her team. She also involved in the study of genotype by environment interaction for Kacip Fatimah and screening selected clones until it recognized as elite clone for commercialization purpose at the end.