

LEAF GAS EXCHANGE PROPERTIES OF THREE VARIETIES OF
LABISIA PUMILA BENTH. UNDER GREENHOUSE CONDITIONS

Jaafar, H.Z.E.^{1*}, Ibrahim, M.H.¹ and Philip, E.²

¹Department of Crop Science, Faculty of Agriculture,
Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia

²Forest Research Institute Malaysia (FRIM), Kepong, 52109 Selangor

*E-mail: hawazej@agri.upm.edu.my

ABSTRACT

Leaf gas exchange properties of three varieties of *Labisia pumila* Benth. (varieties *alata*, *lanceolata* and *pumila*) were measured using LICOR 6400 portable photosynthesis meter under greenhouse conditions in a single factor Complete Randomized Design replicated 10 times. Results indicated that there were no varietal preferences on stomata conductance, water use efficiency and transpiration rate. However, net photosynthesis was 33% and 26% significantly higher ($p \leq 0.01$) in var *lanceolata* ($3.47 \mu\text{mol/m}^2/\text{s}$) compared to varieties *alata* ($2.33 \mu\text{mol/m}^2/\text{s}$) and *pumila* ($2.58 \mu\text{mol/m}^2/\text{s}$), respectively. Although other leaf gas exchange parameters did not differ significantly among the varieties, var *lanceolata* consistently recorded higher values for stomata conductance and water use efficiency against other varieties. The results may imply that var *lanceolata* is a more adaptable, hence, more suitable for the propagation under greenhouse conditions than the other two varieties. However, further research needs to be conducted to confirm these findings as the effects observed might be attributed to the differences in stomata sensitivity of the plants studied. Variety *alata*, conversely, had consistently exhibited the lowest values for all its leaf gas exchange characteristics suggesting that the variety is more challenging to be raised under the greenhouse conditions for domestication purposes. Selection of variety for propagation should also, however, be based upon varietal potential to produce and accumulate secondary metabolites under greenhouse for the purpose of enhancing local herbal industry.

Keywords: Kacip Fatimah, water use efficiency, stomata conductance, transpiration rate, net photosynthesis

INTRODUCTION

Kacip Fatimah (*Myrsinaceae* family; *Labisia pumila* Benth) or locally known in Malaysia as *Selusoh Fati mah*, *Tadah Matahari*, *Pokok Pingan g* and *Belangkas hutan* is a woody, small subherbaceous plant with creeping stem (Jamia et al. 2003). This plant is shade loving and best thrives under heavy canopy and mostly found on the hill and lowland forest of Peninsular Malaysia. *Labisia pumila* is slow growing and very sensitive to its surrounding microenvironment. Exposure to direct open

sunlight reduces growth, and establishment of this plant (Zhari et al. 1999) making it more challenging for domestication and conservation purposes.

There are three varieties of *Labisia pumila* in Malaysia (Stone 1988), namely var *alata*, var *pumila* and var *lanceolata*. Of the three, var *lanceolata* is the least studied. The plant is highly demanded for its medicinal value as female health tonic due to the discovery of the plant phyto-estrogenic properties (IMR 2002; Jaafar et al. 2008). Each variety commands different use. For instance, *Labisia pumila* var *alata* is known to be used in the pre-partum and *Labisia pumila* var *pumila*, in the post partum (Jamia et al. 2004; Jaafar et al. 2008). In addition, the plant also has anti-microbial properties and acts as anti-dysmenorrhoea, anti-flatulence, anti-rheumatism and anti-gonorrhoea. Due to its high health benefit, recently a lot of local entrepreneurs have established energy drink based on this plant such as Livita Femina, Pearl, and Kacip Fatimah Orang Kampung.

Due to high demand of the plant as herbal remedy and for use in herbal formulation, the demand for raw material has soared. With the current high rate of demand and methods of harvesting, particularly from the wild, the natural population sizes of different varieties of *Labisia pumila* may soon dwindle. This has made research on domestication, propagation and establishment of robust, high quality *Labisia pumila* plants more urgent than ever, especially when raising and establishing the plant is difficult due to lack of information on the agronomic and propagation practices. There has been some limited works on propagation of *Labisia pumila* under shelters (Rozihawati et al. 2004), however, findings were not conclusive.

The aspects of leaf gas exchange *Labisia pumila*, especially in the different varieties, have not been well researched and measured. This information is vital to differentiate the adaptability and capacity of different varieties to thrive well under greenhouse micro-environment based on their photosynthetic and water use efficiency performances. This information has been used widely in many crops as a selection tool for best progenies or varieties that suit certain set of environment as varieties display different acclimation of leaf gas exchange to different environment (Roslan & Haniff 2006). Thus, the aim of the present work was to determine the variety that could thrive well under greenhouse conditions based on the leaf gas exchange characteristics measured with portable photosynthesis system.

MATERIALS AND METHODS

Population of *Labisia pumila* and experimental design

The experiment was carried out at Faculty of Agriculture Greenhouse, Universiti Putra Malaysia on 18 month old greenhouse-acclimatized accessions of *Labisia pumila* after collection from different population: *Labisia pumila* var *alata* from Sungkai, Perak, var *lanceolata* from Kota Tinggi, Johore, and var *pumila* from

Kuala Langat, Selangor. The study used a complete randomized design (CRD) replicated 10 times with *Labisia pumila* varieties as the treatments.

Greenhouse microclimatic conditions and plant growth

The mean microclimatic conditions of the greenhouse are presented in Table 1. Under these conditions, all the three varieties of *Labisia pumila* were observed to grow well (Plate 1) and were able to bear flowers and set fruits (Plate 2).

Table 1. The mean microclimatic conditions under greenhouse cultivated with three varieties of *Labisia pumila* Benth.

Microclimate Parameters	Quantification
Relative humidity (RH)	77.63%
Photosynthetic photon flux density (PPFD)	13.13 $\mu\text{mol}/\text{m}^2/\text{s}$
Ambient CO ₂	351.67 $\mu\text{mol}/\text{mol}$
Canopy air temperature	26.71 °C
Soil temperature	25.85 °C

Leaf gas exchange parameters

In this study, net photosynthesis (A), stomata conductance (g_s), transpiration rate (E), and instantaneous water use efficiency (WUE) were measured simultaneously using a LICOR-6400 portable photosynthesis system (IRGA: LICOR Inc. Nebraska, USA). A homogenous population was randomly selected from each of the treatment varieties. Prior to measurements, standard cuvette conditions were observed i.e. PPFD, at 1000 $\mu\text{mol}/\text{m}^2/\text{s}$, CO₂ reference, at 400 $\mu\text{mol}/\text{mol}$, relative humidity at 50 – 60 % and leaf temperature set at 30°C. All measurements were made on fully expanded leaf. Before measurement, the instrument was warmed-up for 30 minutes and then calibrated. Instantaneous water use efficiency was calculated using the equation by Tanner and Sinclair (1983):

$$w = P_n / E_p$$

where P_n is the net assimilation rate ($\mu\text{mol}/\text{m}^2/\text{s}$) and E_p is evapo-transpiration rate ($\text{mmol}/\text{m}^2/\text{s}$). A mean of 10 plants were sampled for each varieties, and in each plant a mean of three leaf samples were used. Data were subjected to Analysis of Variance using PC-SPSS program, and means separation test were differentiated

using Duncan's multiple range test (DMRT) and standard error of differences (SED).



Plate 1. Mixed population of *Labisia pumila* collected from different locations in Malaysia. The plants have been acclimatized under greenhouse condition for eighteen months.

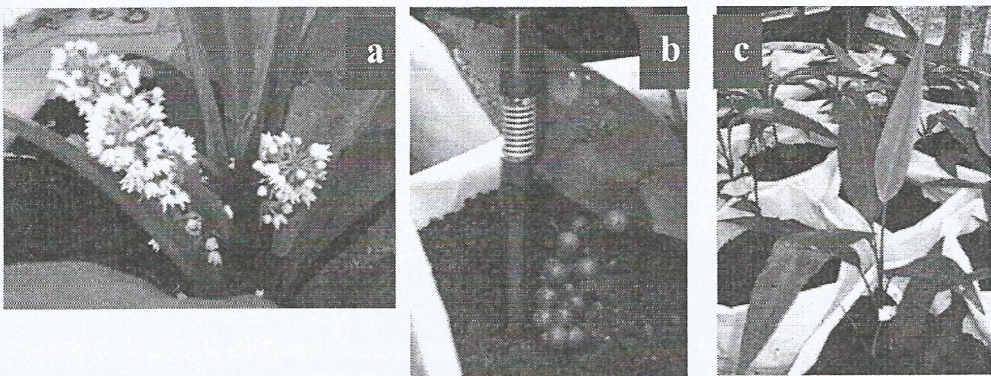


Plate 2. Under greenhouse microclimatic conditions, *Labisia pumila* was able to bear good flowers (a), set fruits (b), and enhanced the growth of all varieties of *L. pumila* for example var *lanceolata* (c).

RESULTS

There were no statistical significant differences of leaf gas exchange traits of stomata conductance, transpiration rate and water use efficiency ($p \geq 0.05$; Table 2) among varieties observed except for net photosynthesis. *Labisia pumila* var *lanceolata* had higher net photosynthesis ($3.47 \mu\text{mol/m}^2/\text{s}$; Figure 1) followed by var *pumila* and var *alata* (2.58 and $2.33 \mu\text{mol/m}^2/\text{s}$, respectively). Enhancement of net photosynthesis in var *lanceolata* was almost 33% more than that recorded in var *alata*, and 26% of var *pumila*. Meanwhile, although stomata conductance and water use efficiency among the varieties did not differ notably, var *lanceolata* again exhibited higher values more than var *pumila* and *alata* by 29% and 25%, respectively for stomata conductance (Figure 2), and about 22% more in water use efficiency than var *alata* (Figure 3). However, var *pumila* recorded higher rate of transpiration followed by var *lanceolata* and *alata* by respective 9% and 13% (Figure 4).

Table 2. Mean squares for net photosynthesis (A), stomata conductance (g_s) transpiration rate (E) and water use efficiency (WUE) of *Labisia pumila* under glasshouse conditions.

Leaf gas exchange parameters	S.V.	S.S.	Df	M.S.	F	Sig.
A	Variety	6.96	2	3.48	8.22	0.00163**
	Error	11.44	27	0.42		
	Total	18.40	29			
g_s	Variety	0.00	2	0.00	0.19	0.826255
	Error	0.02	27	0.00		
	Total	0.02	29			
E	Variety	0.61	2	0.31	0.88	0.427515
	Error	9.43	27	0.35		
	Total	10.04	29			
WUE	Variety	0.64	2	0.32	0.61	0.54822
	Error	14.04	27	0.52		
	Total	14.63	29			

Notes:

** , significant at $p \leq 0.01$; S.V. = source of variation; S.S. = sum of squares; M.S. = mean of squares; Df = degree of freedom; Sig. = significant

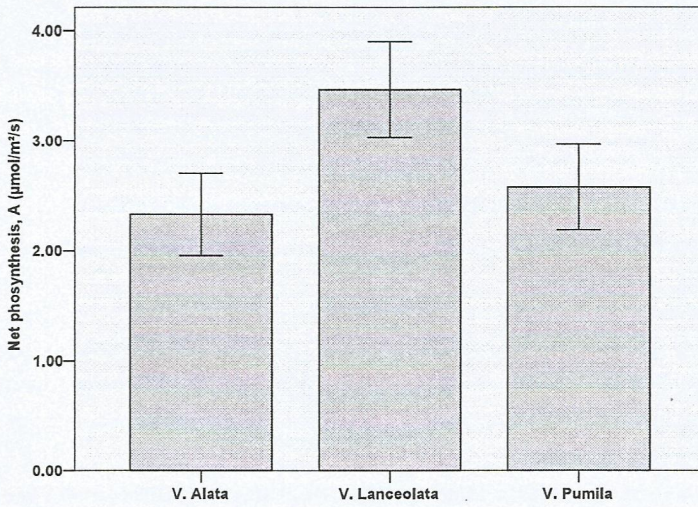


Figure 1. Varietal differences on *Labisia pumila* net photosynthesis. N = 10. Bars represent standard error of differences between means (SED).

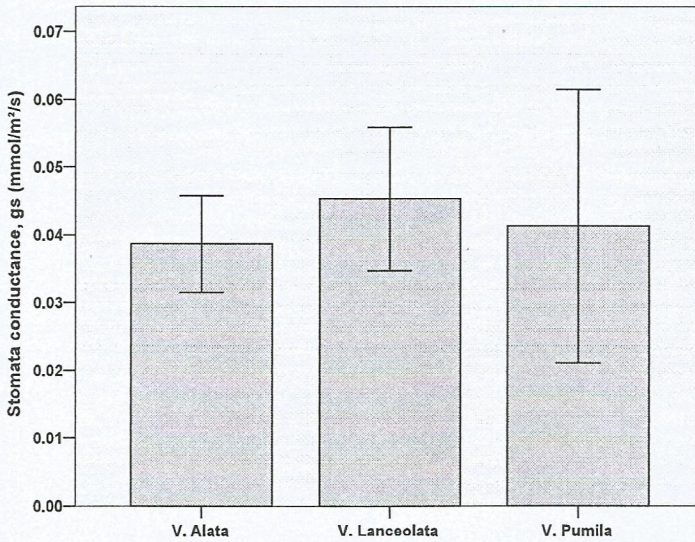


Figure 2. Varietal differences on *Labisia pumila* stomata conductance. N = 10. Bars represent standard error of differences between means (SED).

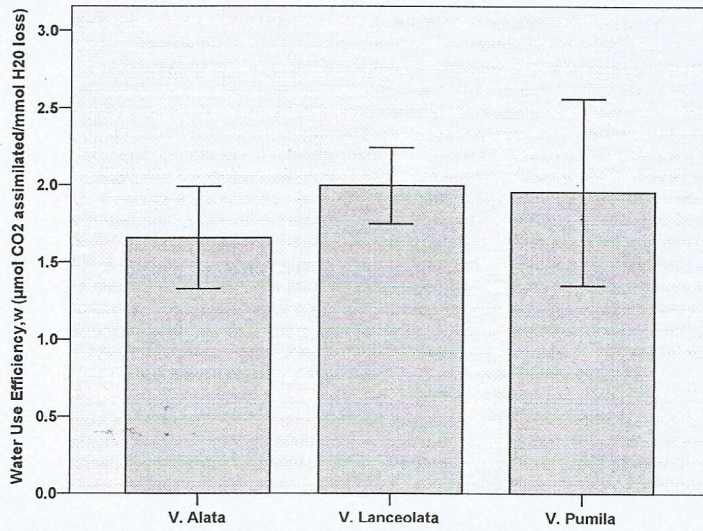


Figure 3. Varietal difference on *Labisa pumila* water use efficiency. N = 10. Bars represent standard error of differences between means (SED).

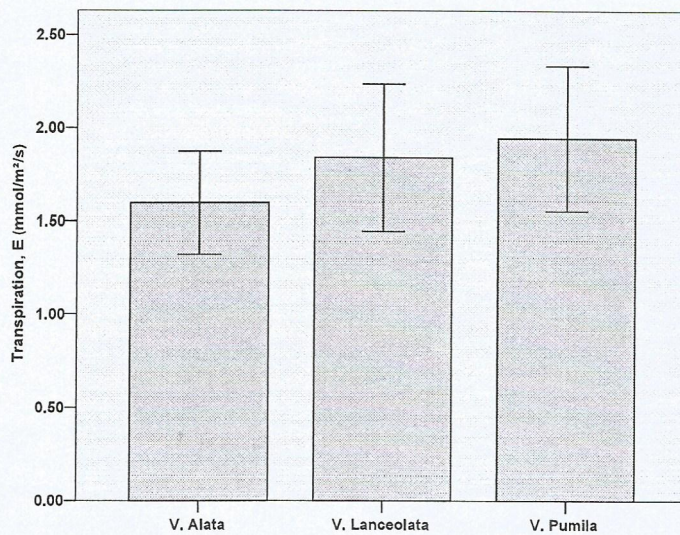


Figure 4. Varietal differences on *Labisa pumila* transpiration rate. N = 10. Bars represent standard error of differences between means (SED).

DISCUSSION AND CONCLUSION

All the three varieties of *Labisia pumila* had almost similar characteristics of the leaf gas exchange except for net photosynthesis, which recorded significantly higher value in *Labisia pumila* var *lanceolata* compared to other two varieties. At the same time var *lanceolata* also exhibited higher values in stomata conductance and water use efficiency although they were not drastically different among varieties. Increase in net photosynthesis could be attributed to increase in stomata conductance as observed by the strong relationship between the two parameters, which allows more CO₂ to be fixed at the same time allowing the transpiration rate to increase (Ibrahim et al. 2009). Enhanced stomata conductance is known to improve the sub-stomata intercellular CO₂ concentration (C_i), hence enhances the net photosynthesis (Ainsworth & Rogers 2007; Ibrahim et al. 2009).

The increase in net photosynthesis of var. *lanceolata* might also be attributed to higher Rubisco per unit chlorophyll surface area that can enhance net photosynthesis (Terashima et al. 1995). On the other hand, lower net photosynthesis values recorded in varieties *alata* and *pumila* could result from differences in stomata sensitivity of the plants. For example in some sensitive plants, even small stress induced could increase the resistance of sub-stomata and, thus, reduced the net photosynthesis (Kogami et al. 2001). Among the varieties studied, *Labisia pumila* var *alata* seemed to be most sensitive to the greenhouse conditions set at it had consistently registered lower values in all the gas exchange parameters measured.

Reduced stomata conductance could also be attributed to thick cell wall trait that decreases stomata conductance (Angelos et al. 2003). However, in the present study measurements of the stomata characteristics were not carried out. Subsequently, with decrease in stomata conductance as a result of stomata sensitivity/characteristics could apprehend water use efficiency as both leaf gas exchange traits are very strongly related as observed by Ibrahim et al. (2009) in oil palm seedling exposed to elevated carbon dioxide. Hence, pre-selection or pre-determination of more adaptable / tolerant varieties to greenhouse conditions for the purpose of propagation and domestication could be addressed by the characteristics of the leaf gas exchange.

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