INTRODUCTION

Two fig varieties, Improved Brow Turkey (IBT) and Masui Dauphine (MD), have different effects on growth and physiological changes after receiving different concentrations of brassinolide. Brassinolide, first isolated from brassinosteroids as steroidal plant hormones that have unique biological impacts on growth and development (Clouse, 2011). Biomass production for each plant species may vary considerably. The maximum relative growth rate (RGR), the dry weight increase per unit of biomass and time under optimal conditions, may differ for each species. Related to yield, the main growth factors are the functional leaves, dry matter production and leaf area index.

As individual size influences tremendous ecological processes, growth rates are fundamental for plants. As main ecological importance such as survival, reproduction and competitive interactions, growth for plants, rely on individual size. Plants differ widely in RGR both among and within habitats (Paine et al., 2015), with consequences for community structure and dynamics (Kraft, Valencia, & Ackerly, 2008). To investigate the mechanistic basis of inter- and intra-specific variation in growth, RGR can be factored into net assimilation rate (NAR), also called unit leaf rate, specific leaf area (SLA), and leaf mass ratio (LMR).

At a given point of time, increasing dry matter is measured by RGR. The net gain in total dry matter per unit leaf area per unit time (described as NAR) is the rise in dry biomass per unit leaf area and an intricate variable of plant physiology related to rates of respiration and photosynthesis. The leaf area per unit leaf mass (or SLA), a characteristic of morphology, and LMR is the ratio between leaf and plant biomass, which describes a plant’s investment in light capture (Li, Schmid, Wang, & Paine, 2016).

ABSTRACT

There is limited information and insures for next studies on exogenous brassinolide application fig cultivar of Masui Dauphine (MD) and Improved Brown Turkey (IBT). Therefore, the research objective was to determine the impact of concentrations of exogenous brassinolide application on Leaf Area Index (LAI), Leaf Mass Ratio (LMR), Net Assimilation Rate (NAR), Relative Growth Rate (RGR), and Specific Leaf Area (SLA) of fig. Fig resources were propagated using stem cuttings and were transferred into 3:2:1 mixed soil (topsoil:organic matters:sand) media. Two cultivars of fig treated with brassinolide (control, 50, 100 and 200 ml/L) were repeated four times and constructed by Split Plot Randomized Complete Block Design. Changes in the parameter of LAI, LMR, NAR, RGR, and SLA were investigated monthly. There was a significant effect of fig variety alone on LAI, SLA and LMR but it wasn’t affected on NAR and RGR. SLA and LMR were influenced by brassinolide but not on LAI, NAR and RGR. Cultivar IBT showed higher responses to these parameters than cultivar MD after receiving brassinolide treatment. The interaction between brassinolide and variety was significant on LAI, SLA and LMR of fig except in the parameters of NAR and RGR.
There are several possible explanations based on the contribution of these factors to RGR. Firstly, the relative influence of functional characters on RGR may differ with conditions of environment, particularly availability of light. If RGR is principally determined by SLA and NAR for low and high irradiances respectively, the SLA and NAR compensatory effects on RGR would limit variation on RGR in heterogeneous light (Ryser & Wahl, 2001). Secondly, SLA, LMR and NAR can all differ over ontogeny (Rees et al., 2010), influencing contributions on RGR. RGR tends to diminish over ontogeny, as more biomass is distributed to non-photosynthetic tissue, and costs of respiration and the increasing of self-shading (Paine et al., 2012). Thirdly, because of interspecific variation in RGR, NAR, SLA, and LMR, research on one or few plant species may be unreliable for wider patterns (Osnas, Lichstein, Reich, & Pacala, 2013). Finally, Size-varying analyses overemphasize the correlation between RGR and NAR because of absolute growth rate and plant size is related to NAR (Rees et al., 2010).

Leaf area index (LAI) is the total leaf area determined per unit of ground area (Hu, Yan, Mu, & Luo, 2014). Investigating the LAI should have to observe the atmosphere, growth of vegetation, radiation uptake, gas exchange, energy conversion, and precipitation. Essentially, the leaf area represent the vegetation on a deciduous tree, and the tree’s leaves are much more consideration compared to the stems, twigs, and branches. Traditionally, LAI are measured by direct and indirect methods (Pinto-Júnior, Sanches, de Almeida Lobo, Brandão, & de Souza Nogueira, 2011). Destructive sampling, allometry and litter traps are direct method. By measuring radiation transmission through the canopy, the indirect ground method infers LAI (Ryu et al., 2010). The RGR is the product of NAR and SLA, where NAR is widely the net result of carbon gain (namely photosynthesis) and carbon losses (i.e. respiration, exudation, and volatilization) shown per unit leaf area (Osone, Ishida, & Tateno, 2008). There is less literature about exogenous brassinolide utilization on these cultivars. Thus, the research objective was to investigate the impact of different concentrations of exogenous application of BL on LAI, LMR, NAR, RGR, and SLA.

**MATERIALS AND METHODS**

Cuttings were used to propagate Fig and transferred into the 3:2:1 mixed soil (topsoil: organic matters: sand) media. Two fig varieties (MD and IBT) were examined into 0, 50, 100 and 200 ml/L of brassinolide (BL). Different fig cultivar and BL concentrations were considered as main and subplots respectively. This research was arranged by Split Plot Randomized Complete Block Design (SRCBD) with four replications. This study was carried out on an open field at Ladang 15, Faculty of Agriculture, Universiti Putra Malaysia located at 2° 58’ N and 101° 44’04” E in Serdang, Selangor, Malaysia from May to December 2017. Data were recorded monthly.

**Measuring Leaf Area Index**

The ratio of foliage area to ground area (LAI) was measured by using LAI-2000C Plant Canopy Analyzer from LICOR, Inc., USA.

**Net Assimilation Rate Determination**

Net Assimilation Ratio was measured by Beadle formula (Rajput, Rajput, & Jha, 2017). This formula measures the rate of dry matter production per unit leaf area or the net gain in dry weight of the plant per unit leaf area per unit time.

\[
   \text{NAR} = \frac{(W_2 - W_1) \ln(A_2 - A_1)}{(A_2 - A_1) (t_2 - t_1)} 
\]

Where W represent dry weight of whole seedlings (g), A represent leaf area per seedlings (cm²) and t represent time (month).

**Specific Leaf Area Determination**

Specific Leaf Area is ratio between the leafiness of plant and a dry weight (Milla, Reich, Niinemets, & Castro-Diez, 2008).

\[
   \text{SLA} = \frac{\text{Seeding leaf area (cm²)}}{\text{Seeding total weight (g)}} \quad (2)
\]

**Relative Growth Rate Determination**

Relative growth rate is growth rate relative to size (Rajput, Rajput, & Jha, 2017). In plant physiology, the speed of plant growth is quantified by RGR. The increase in dry matter with a given amount of assimilatory material at a given point of time is measured by RGR.

\[
   \text{RGR} = \frac{\ln W_2 - \ln W_1}{(t_2 - t_1)} \quad (3)
\]

Where \( W_t \) represent dry weight of plant/m² recorded at time \( t \) (g), \( W_{t_1} \) represent dry weight of plant/m² recorded at time \( t_1 \) (g), and \( t_{1,2} \) represent interval time (month).

**Leaf Mass Ratio Determination**

Percentage of LMR was measured based on the ratio between leaf biomass and the sum
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of above- and belowground biomass (Li, Schmid, Wang, & Paine, 2016).

\[ \text{LMR} = \left( \frac{\text{Leaf Dry Mass}}{\text{Leaves} + \text{Shoots} + \text{Stems} + \text{Roots}} \right) \times 100\% \]

Statistical Analysis

Data were recorded monthly and analyzed using Statistic Analysis System (SAS) 9.4. Two-way ANOVA and Least Significant Different Test at 5 % were adopted to check the significant differences of mean values and level of differences respectively.

RESULTS AND DISCUSSION

Table 1 describes that fig growth was influenced by brassinolide levels and varieties. Treatment interaction between concentrations of brassinolide and variety of fig was significant at parameter LAI, SLA and LMR at fourth and first Month After Treatment (MAT) respectively. There was no interaction between brassinolide concentrations and fig variety at parameter NAR and RGR. Treatment brassinolide was significant at parameter LAI, NAR and RGR but not significant at parameter SLA and LMR. In addition, treatment cultivar of fig, parameter LAI was significant only at 3 MAT, SLA at 3 and 4 MAT, and LMR at 4 MAT.

Treatment of 50 and 100 ml/L brassinolide increased LAI and it decreased at 200 ml/L brassinolide compared to control. Treatment of 100 ml/L brassinolide resulted in higher LAI for IBT variety and treatment of brassinolide 200 ml/L for MD variety. Variety IBT resulted in higher LAI than variety MD at 4 MAT.

NAR showed increased strongly when increasing concentration of brassinolide (control and 50 ml/L) at 1-2 MAT and 2-3 MAT and decreased strongly when increasing concentration of brassinolide (100 and 200 ml/L) at 3-4 MAT (Fig. 1b). Based on Table 1, showed that treatment of brassinolide was significant only at 2-3 MAT on NAR and there was no effect of fig variety on NAR. SLA showed no consistent pattern among increasing concentrations of BL (Fig. 1c). Meanwhile, RGR value resulted increased strongly when increasing concentration of brassinolide at 1-2 MAT and 2-3 MAT and decreased strongly at 3-4 MAT except at brassinolide concentration 100 ml/L (Fig. 1d). Additionally, LMR (Fig. 1e) was enhanced when increasing brassinolide concentration (50, 100, and 200 ml/L) compared to control.

In this study, the effect of exogenous brassinolide application on LAI, LMR, NAR, RGR, and SLA characters was explored on two varieties of fig which may cause the increase of cell enlargement (Bajguz & Hayat, 2009). Parameter LAI and LMR had a consistent graph. As brassinolide levels improved (50, 100, and 200 ml/L) these parameters were also linearly increased to 11.66 % and 33.25 % respectively bigger than control. The growth excitation was more pronounced on biomass of above-ground than the biomass of below-ground by having bigger shoots than roots. The enhance in growth in this experiment might increase the rate of carboxylation after receiving brassinolide which improved carbon assimilation that was directed to improves in total biomass and leaf area (Raines, 2011). Whilst the parameter of NAR, SLA, and RGR had an inconsistent graph.

The ecological benefit of a high RGR may seem to be clear: In competitive situations, a fast occupation of space is resulted by fast growth (Boyden, Reich, Puettmann, & Baker, 2009). Fast completion of the life cycle on a plant may be facilitated by a high RGR. Having shown that LMR was essential in explaining differences in RGR, future analysis of this factor was imperative. In fast-growing species, allocation of dry matter to leaves (LMR) was higher, but the relationship with RGR was not very tight. Plant species from fertile, productive habitats tend to have inherently higher relative growth rates (RGR) than species from less good environments when they are planted under optimum conditions. Under these conditions, more leaf area and less root mass will be produced by fast-growing species (Sugiura & Tateno, 2011), and it’s related to larger carbon gain per unit plant weight. They have a higher photosynthesis rate per unit leaf dry weight and per unit leaf nitrogen, but not necessarily per unit leaf area, due to their higher leaf area per unit leaf weight. Fast-growing species also produce a higher rate of respiration per unit organ weight, because of demands of a higher rate of nutrient uptake and higher RGR. However, expressed as a fraction of the total amount of carbon fixed per day, they use less in respiration (Poorter et al., 2012).

In explaining variation in RGR, the balance of respiration and photosynthesis per unit leaf area, are not of principal importance based on differences in the carbon economy. The relationship between RGR and LAI on the other hand, was slightly fitted.
Table 1. Effect application of different concentrations of brassinolide on physiological parameters two fig varieties

| Treatments | Leaf Area Index (nm) | Net Assimilation Rate (g/cm²/month) | Specific Leaf Area (cm²/g) | Relative Growth Rate (g/g/month) | Leaf Mass Ratio (g/g) |
|------------|----------------------|-------------------------------------|---------------------------|---------------------------------|----------------------|
|            | Month After Treatment | Month After Treatment | Month After Treatment | Month After Treatment | Month After Treatment |
|            | 1 2 3 4 | 1-2 2-3 3-4 | 1 2 3 4 | 1-2 2-3 3-4 | 1 2 3 4 |
| Control    | 1.09³ 0.18 0.50³ 0.20³ | 0.067 0.214³ 0.046 | 11.96 8.06 8.43 | 4.40 | 0.69 1.41 0.42³ | 7.43 14.99 15.94 8.41 |
| 50ml/L     | 0.61³ 0.68 0.26³ 0.35³ | 0.059 0.103³ 0.106 | 7.45 9.64 7.07 | 5.19 | 0.94 1.51 0.97³ | 12.77 17.01 11.12 9.67 |
| 100ml/L    | 0.90³ 0.58 0.85³ 0.62³ | 0.050 0.095³ 0.246 | 7.97 7.37 6.22 | 4.76 | 0.90 0.92 0.97³ | 15.05 13.22 12.59 8.96 |
| 200ml/L    | 1.14³ 0.57 0.61³ 0.45³ | 0.035 0.267³ 0.137 | 13.73 9.99 7.83 | 5.16 | 0.74 1.70 1.02³ | 29.21 17.04 11.39 9.54 |
| IBT        | 0.91 0.41 0.66³ 0.40 | 0.055 0.159 0.098 | 11.04 8.36 6.84³ | 4.3³ | 0.85 1.35 0.76 | 18.09 14.12 11.41 7.80³ |
| MD         | 0.95 0.60 0.45³ 0.41 | 0.051 0.181 0.169 | 9.51 9.17 7.94³ | 5.4³ | 0.78 1.42 0.93 | 14.14 17.01 14.11 10.49³ |
| IBT + 50ml/L | 0.69 0.69 0.26 0.29³ | 0.030 0.075 0.119 | 7.10³ 11.45 5.99 | 4.42 | 0.84 1.38 0.87 | 15.15³ 17.24 10.06 8.36 |
| IBT + 100ml/L | 1.24 0.56 0.96 0.95³ | 0.029 0.053 0.153 | 12.69³ 8.42 5.58 | 3.66 | 0.80 0.70 1.00 | 18.49³ 14.27 10.58 6.51 |
| IBT + 200ml/L | 0.97 0.08 0.64 0.22³ | 0.058 0.237 0.069 | 8.96³ 9.10 8.18 | 5.48 | 0.90 1.93 0.72 | 27.88³ 16.16 12.24 9.06 |
| MD + 50ml/L | 0.54 0.66 0.25 0.41³ | 0.089 0.131 0.093 | 7.79³ 7.82 8.15 | 5.95 | 1.03 1.64 1.08 | 10.39³ 16.77 12.18 10.97 |
| MD + 100ml/L | 0.56 0.60 0.74 0.29³ | 0.070 0.137 0.339 | 3.25³ 6.33 6.86 | 5.86 | 1.00 1.14 0.94 | 11.62³ 12.17 14.60 11.41 |
| MD + 200ml/L | 1.31 1.07 0.57 0.68³ | 0.012 0.297 0.205 | 18.49³ 10.88 7.48 | 4.84 | 0.59 1.48 1.33 | 30.5³ 17.93 10.53 10.01 |
| LSD V       | 0.12 | 0.88 | 0.62 | 2.58 |
| LSD B       | 0.51 0.53 0.33 | 0.101 | 0.26 |
| LSD V*B     | 0.62*0.36 | 13.25*9.07 | 22.09*15.11 |

Remarks: Means values within a column followed by the different small letters are significant at P < 0.05
The higher LAI at IBT variety and brassinolide 100 ml/L might be due to more number of leaves produced per unit area. Half the total green leaf area per unit ground surface area \( (m^2/m^2) \) is called LAI. It means that a higher number of leaves have higher value of LAI. All treatments (varieties, brassinolides and interaction between varieties and brassinolides) had significant value on LAI (Table 1). A significant interaction between varieties and brassinolides occurred only at 4 MAT. A range of LAI value worth in 0.08-1.31. It is hard to differentiate foliage from woody tissue. In common, the highest values reported previously for LAI are for coniferous canopies, in some cases LAI > 15, even though this is partly a function of how LAI is defined and measured (Liu, Jin, & Qi, 2012). As a locus of physical and biogeochemical processes in an ecosystem, nutrient dynamics, microclimatic conditions, herbivore activities influence the functional and structural attributes of plant canopies. In ecosystem processes such as photosynthesis, transpiration, energy exchange, and other physiological attributes, the amount of foliage, and canopy leaf area play an important role.

At 50 and 100 ml/L brassinolide, specific leaf area was lower than control \( (P \leq 0.05) \). The result indicates that leaves of plants were thicker. The thicker leaf might be owing to improvement in mesophyll layer after receiving brassinolide (Xu, Salih, Ghannoum, & Tissue, 2012). The increase in leaf thickness caused by higher leaf weight ratio in the fourth MAT compared with 1-3 MAT. The leaf area was maintained at lowest SLA. This showed that improved in SLA was in consequence of enhancing in leaf weight compared to enhance in leaf area (Hayat, Alyemeni, & Hasan, 2012; Poorter et al., 2012). The major factor explaining variation in RGR seem to be SLA, it is described by a high SLA had the highest RGR. Seemingly fast-growing plant produces a low investment in biomass of leaves. Distinctions in SLA can be ascribed either to morphology such as the thickness of leaves and structure of vein or to the leaf chemical composition (Ruggiero, Ascione, Punzo, & Vitale, 2012).

The NAR of plants reflects plant growth performance under specified condition (Tomlinson et al., 2014). It is clear that plant under elevated BL has high NAR. Rajput, Rajput, & Jha (2017) reported that increases in plant growth on different planting geometries and depths of SRI increased 30 % total biomass in rice and also increased 4 % NAR compared to control. The NAR reduction caused by ontogenetically development of fig shows the plant reaching maturity.

Consistently, the best predictor of growth rate was NAR, whilst lesser contributions were made by SLA and the allocation of biomass to LMR. In other words, high net assimilation rates and growth fast was provided by the fast-growing plant. Furthermore, NAR was positively related to both leaf N concentration and the maximum rate of photosynthesis.

As another essential plant growth regulator, BL has profound effects on plant performance and leaf photosynthesis. On cotton seedlings under NaCl stress, BL application increased photosynthetic performance (Shu, Guo, Shen, & Ni, 2011). BL application boosted the formation of lateral roots, the occurrence of new roots, and nutrient absorption on cucumber seedlings (El-Feky & Abo-Hamad, 2014).

Because of a negative relationship between SLA and NAR, and this in turn, will have a negative impact, therefore masking the correlation between LAI and RGR. Poorter et al. (2012) offer two explanations for the negative relationship between SLA and NAR. Firstly, an increasing NAR may involve an improved photosynthesis rate, which can be resulted by extra investment in the photosynthetic apparatus, therefore decreasing SLA. Second, the balance between the leaf area and the root amounts could affect the water status of the leaves and hence the rate of photosynthesis. As the negative relationship between NAR and SLA is primarily affected by a negative correlation between SLA and NAR, and the second explanation is the most probable for this result case.

**Correlation Analysis**

Based on the correlation analysis, significant negative relationship was noted between leaf area index and relative growth rate, and between net assimilation rate and specific leaf area (Fig. 2). Enhancing in leaf area index and net assimilation rate was associated with a decrease in relative growth rate, and specific leaf area at 16.08 %, and 59.67 % respectively.
Remarks: Bars represent means followed by the different small letters significant at P<0.05, and ns=not significant

**Fig. 1.** Changes in parameter of fig: a) LAI at fourth MAT as interaction between cultivars and brassinolides; b) NAR as main effect of brassinolides; c) SLA at first MAT as interaction between cultivars and brassinolides; d) RGR as main effect of brassinolide, and e) LMR at first MAT as interaction between cultivars and brassinolides
CONCLUSION AND SUGGESTION

There was a significant effect of fig variety alone on LAI, SLA and LMR but it was not affected on NAR and RGR. SLA and LMR were influenced by brassinolide but not on LAI, NAR and RGR. Cultivar IBT showed higher responses to these parameters than cultivar MD after receiving brassinolide treatment. Treatment of brassinolide 200 ml/L resulted in higher LAI, LMR, NAR, RGR, and SLA than the other concentrations. Treatment of brassinolide and variety had a significant effect on LAI, SLA and LMR of fig except in the parameters NAR and RGR.

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