

The Response of Soybean (*glycine max (l.) Meer.*) Varieties from the Tropical Region to Five Watering Regimes under a Controlled Environment

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Submitted: 01 June 2018; Accepted: 15 June 2018; Published: 04 July 2018

Abstract

In some rice dominated tropical regions, such as in Indonesia, soybeans are an increasingly important dry season crop which are often exposed to periods of drought stress. The morphological and physiological responses, which could lead to some tolerance to water stress, may vary between varieties. By better understanding the plant response to drought stress and finding if these responses vary between varieties better dry season production could be achieved. An experiment was conducted to compare the response of four varieties of soybean (*glycine max (l.) Meer.*) to five watering regimes, with the objective of determining the response of common soybean varieties across a wide range of water supply. Plant response to water supply was measured using gas exchange measurement with the rate of photo synthesis decreasing progressively from well watered to dry conditions across the four varieties. A correlation of stomatal conductance and transpiration rate has a close relationship with photosynthetic rate, where stomatal conductance of Burangrang variety has higher value than other varieties. Varieties Burangrang and Argomulyo stomatal conductances are higher value than those of Anjasmoro and Grobogan varieties. In a deficit of water condition, the Argomulyo varieties have a higher value of transpiration efficiency and significantly different than the other three varieties. The transpiration efficiency significantly declined for treatments watered once every two or three weeks. The transpiration efficiency values of Argomulyo and Burangrang varieties were significantly higher than another varieties.

Keywords: Soybean – Varieties – Watering Regimes

Introduction

Increasingly in rice growing regions, such as in Indonesia, dry season grain legume crops such as soybeans are planted in the dry season for their valuable grain and for their positive rotational effects [1,2]. Soybean is an important strategic commodity for Indonesia's food security and a popular supplement for rice, the national staple food [3,4]. Its shortage could trigger economic and political instability for many Indonesian people especially for low-income people [5]. As an inexpensive protein source, it is a well known and used in a variety of food products besides rice and maize [6].

Furthermore, despite the main cropping system in Indonesia being rice-rice and mostly irrigated, the second rice crops are planted late in the season and normally exposed to high drought risk particularly in areas that are located at the end tail of the irrigation system [7]. Indeed, in the season where irrigation water does not reach the area, the dry season crops depend on soil moisture remaining from the wet season and any rainfall [8]. Based on this background, the opportunities for intensifying soybean production with crops following rice in a rotation is imperative.

A water deficit is a major factor limiting agricultural production worldwide and a major constraint to plant growth and productivity [9,10]. To survive drought, plants optimize their morphology, physiology and metabolism at the organ and cellular levels [11].

In the literature, controversy continues as to whether drought limits photosynthesis through diffusion resistance or by metabolic disturbances [12]. Reviews of in vitro and in vivo measurements, it have concluded that drought stress mainly affects the diffusion of CO₂ in the leaves via decreased stomatal and mesophyll conductance, reduces leaf size, stem extension and root proliferation, disturbs plant water relations and reduces water-use efficiency [13,14].

In addition to photosynthetic response, phenology is indeed an important component in the adaptation of plants to any given stress [15]. Morphological and physiological responses that occur vary to some plants and varieties, being able to adapt drought stress. In general, drought tolerance strategies can be identified, involving diverse plant mechanisms that give plants the ability to respond and survive drought [11].

Given soybean is common grow under rain fed conditions in the

semi-arid tropics, drought is one of the most intractable problems for production [16]. Plants have developed a variety of strategies and mechanisms in response to drought.

In general, this article reviews the current state of understanding about the response of different varieties to water stress. It could potentially be beneficial to develop strategies soybean cultivation in the dry season and also in drought prone areas.

Materials and Methods

Experimental Design

A glasshouse based pot experiment was conducted following randomised complete design with three replications at the Georg-August-University in Göttingen, Germany. (51°33'29"N, 9°57'9" W) from 22 July to 29 October 2013. Pots were rearranged weekly to give a random distribution in the glasshouse. The treatments included 4 Soybean common varieties from Indonesia (Argomulyo, Burangrang, Anjasmoro and Grobogan). Pots of 28 cm diameter and 18 cm depth, with an internal capacity of ~8 L, were filled with mix of clay, loam soil, compost, organic matter at a ratio 1:1:2:3 this mix had a field capacity (VWC) of ~50% and a permanent wilting point of ~12%. Temperature in the Glasshouse was set at 34/22°C day/night. Three seeds were planted into hole at 3 cm depth and later thinned to one plant per pot.

After an initial period of watering to field capacity by repeatedly watering during one week, germination and then thinning 5 watering treatments were imposed. (1) watered three days a week; (2) watered two days a week; (3) watered once a week; (4) watered once every two weeks; (5) watered once every three weeks. At each watering event, the pots were re-watered to field capacity with 0.5 to 2.5 L depending on the water treatment and stage of growth.

Measurements

Three parameters of plant physiology, i.e stomatal conductance (gs), transpiration rate (E) and photosynthetic rate (A); were measured using LCi Portable Photosynthesis System (ADC BioScientific Ltd. Hoddesdon, Herts, EN11 0DB) taken before and after each watering event. All measurements were taken at a light intensity of 1305 mol m⁻² s⁻¹ averaged over 5 minutes. This light intensity which was chosen after measuring photosynthetic rate from 0 to 3000 mol m⁻² s⁻¹ and found to be a stable indicator leaf PAR under greenhouse conditions. The Transpiration Efficiency (TE) was calculated from Photosynthetic rate (A) divided by Transpiration rate (E) on the same leaf.

Soil volumetric water content (VWC) was measured frequently with a soil moisture sensor (HS2 HydroSense II) Both plant and soil parameters were measured one day before and after watering.

Statistical Analysis

Data were subjected to Analysis of Variance (ANOVA) procedures (The STATISTICA Version 10) to test for significance between treatments. Treatment effects and interactions that were considered statistically significant when they occurred at ≤0.05 level of probability. If fixed main effects or a fixed interaction were significant in the ANOVA, then mean separation was tested using least-squares means. To control the experiment wise error rate within the means separation t-tests with Post-Hoc analysis, the Fisher's adjustment was used.

Results

Volumetric Water Content (VWC)

Investigating soil-water parameter was conducted to determine the relationship between a soil water content in this case equated with VWC with photosynthesis activity. Based on the results of VWC measurements from week 6 to week 11, It was found that the value of VWC decreased under the more water stress regime. VWC will be declined dramatically as the soil was further dried on watered once a week to watered once every three weeks (Figure 11).

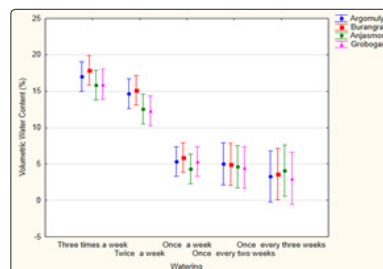


Figure 11: Volumetric Water Content (VWC) at some level of watered regime

The results showed that the highest VWC are at week 7 and 8; and varieties Burangrang VWC showed the highest value compared to the other three varieties (Figure 12).

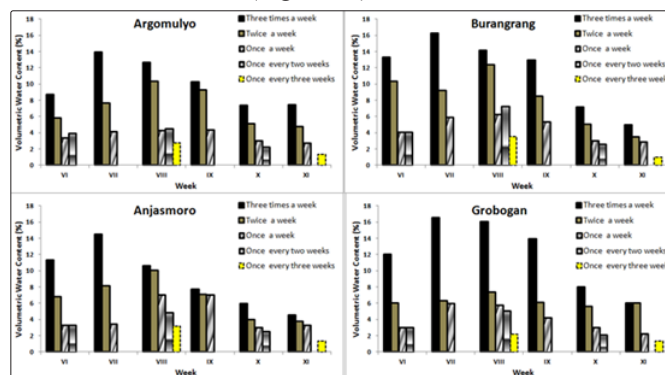


Figure 12: VWC values at the different varieties and water regimes

VWC relationship with photosynthetic rate at week 8 are presented in Figure 13. It is shown that there is a close relationship between the VWC and photosynthetic rate on Burangrang varieties and Grobogan with R² values of 0.71 each and 0.68.

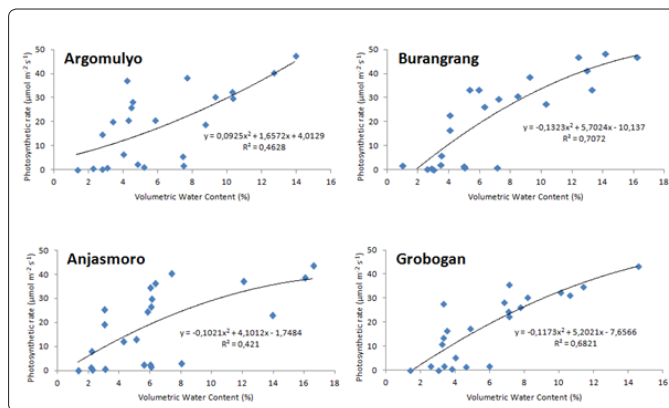


Figure 13: The correlation between VWC with photosynthetic rate in the four weeks to 8 varieties Photosynthesis (A) and associated gas

exchange measurements (stomatal conductance, gs; transpiration, E)

A photosynthetic rate (A) profiles in well watered treatment (three times a week) through time are presented in Figure 1 and during the photosynthetic rate of the experimental period before and after watering are configured in Figure 2. Observations were taken from the end of the vegetative stage (sixth weeks), until the pod filling in week eleven. Photosynthesis, transpiration and stomatal conductance declined with crop development. The highest photosynthesis occurred after the seventh and eighth week, and before decreasing after the ninth week. The differences between varieties in well watered treatment were often small, however Burangrang variety had the highest rate of photosynthesis compared to another varieties (Figure 1) either before or after irrigation (Figure 2).

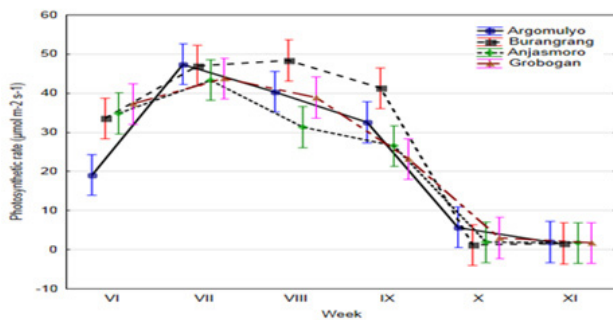


Figure 1: Photosynthetic rate of four soybean varieties grown under well watered conditions

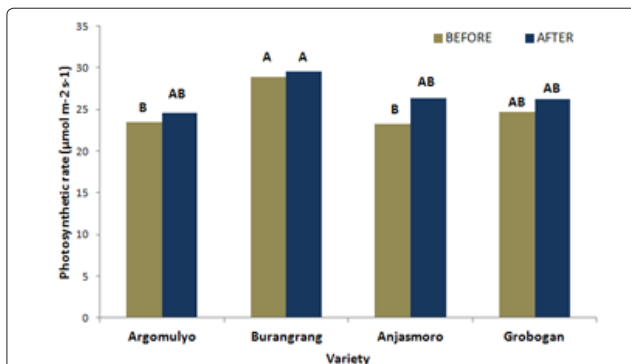


Figure 2: Photosynthetic rate of four soybean varieties grown under well watered conditions before and after a watering event

Photosynthetic rate was strongly affected by watering regime with water stress reducing photosynthetic rate (Figure 3). Then, when the water supply was applied once a week, once every two weeks, and once every three weeks; the rate of photosynthesis would decrease respectively 28-33%, 45-54% and 69-86% from well watered in these four varieties (Figure 3a and Figure 4). An evidence indicates that photosynthesis is highly sensitive to water stress. Meanwhile the addition of water to field capacity will recover to photosynthesis rate from before watered as indicated by increasing of the photosynthetic rate (Figure 3b and Figure 4). There were no significant effect among varieties as well as interaction between watering regime and varieties, however in general, Burangrang variety has a higher photosynthetic rate value than another varieties.

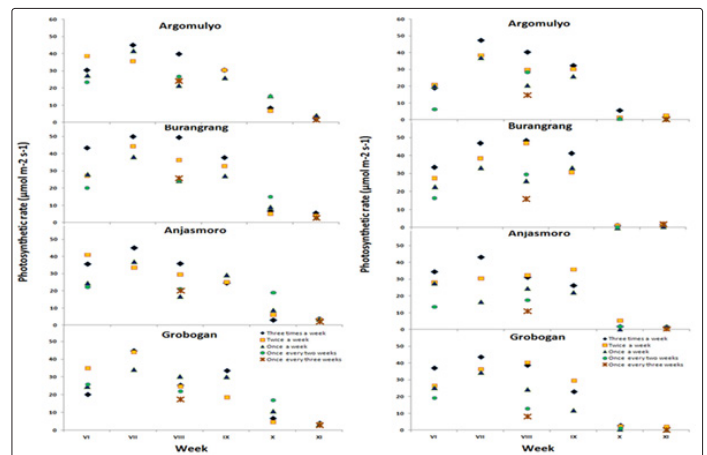


Figure 3: Photosynthetic rate each week (a) before and (b) after a watering event

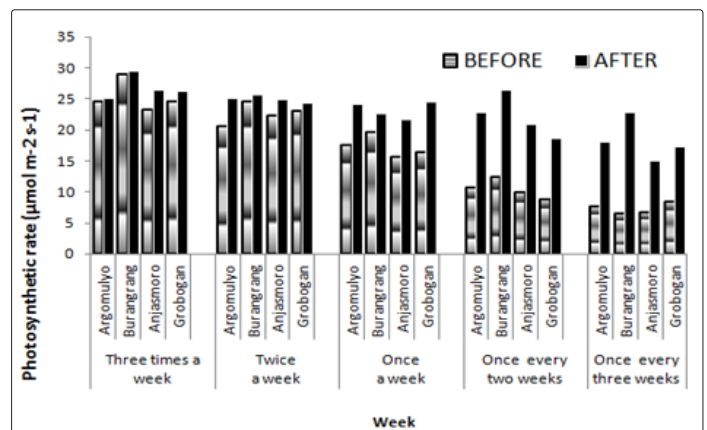


Figure 4: Photosynthetic rate during the experimental period

The correlation between stomatal conductance of CO_2 and photosynthetic rate is shown in Figure 5a, and the correlation between transpiration rate and photosynthetic rate is presented in Figure 5b. Those figures show that the correlation of both stomatal conductance and transpiration rate that has a close relationship with photosynthetic rate with successively r^2 values are 0.89 and 0.88 was obtained; it indicates that at photosynthetic rate was seemingly controlled by stomatal conductance and transpiration rate. Therefore it can be said clearly also, that all changes in photosynthesis were highly correlated with transpiration and stomatal conductance [17].

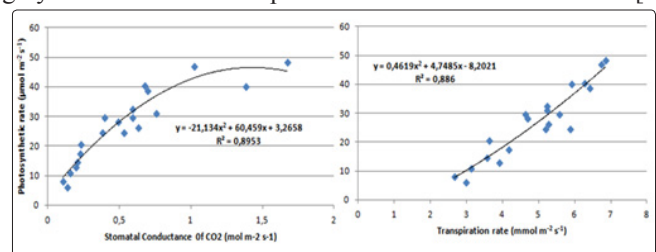


Figure 5: Correlation (a) between stomatal conductance of CO_2 and photosynthetic rate and (b) transpiration rate and photosynthetic rate

Furthermore, it is explained in Stomatal conductance of CO_2 before watering in the week eighth, the interaction between varieties with water supply significantly ($p < 0.001$) where a stomatal conductance will decrease due to lack of water and there is a significant effect

in each watered as well as varieties. The stomatal conductance of Burangrang variety had the higher value than other varieties. Just as with stomatal conductance, there was a significant interaction between varieties with the well watered in the transpiration rate ($p < 0.05$) and the photosynthetic rate ($p < 0.0001$) as can be seen in Figure 6. A high stomatal density has a role in increasing the conductivity of the leaves, especially under well watered conditions.

Transpiration Efficiency

The experiments firstly examined a transpiration efficiency of different varieties under well-watered. The varieties Burangrang and Argomulyo displayed significantly higher transpiration efficiencies than those of Anjasmoro and Grobogan varieties (Figure 7 and 8).

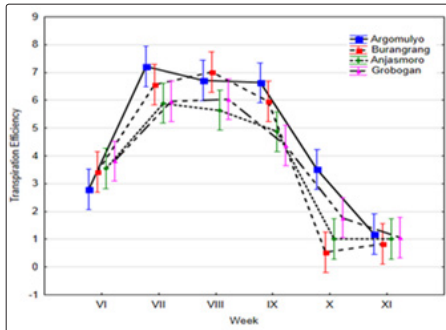


Figure 7: Transpiration Efficiency of four different varieties on well watered condition

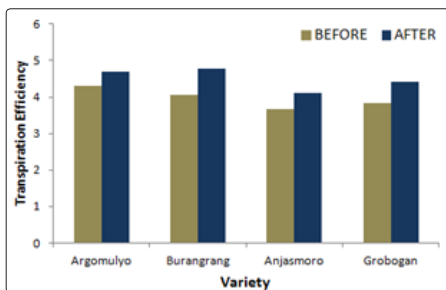


Figure 8: Transpiration Efficiency before and after watering event during sixth to eleventh week

The Argomulyo varieties had a higher value of transpiration efficiency and significantly different than the other three varieties (Figure 9 and 10a). Transpiration efficiency value will decrease if there is a deficit of water, this phenomenon is shown in Figure 8. The Transpiration Efficiency would significantly decline after watered once a week, ranging between 16-21%. It would decrease more than 30% when irrigated once every two weeks to three weeks (Figure 9 and 10b).

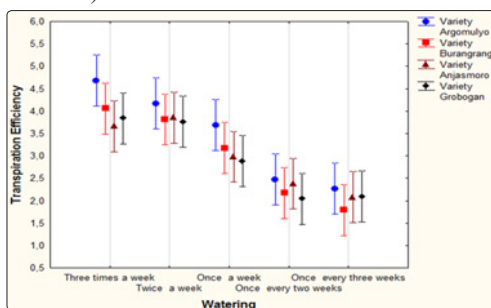


Figure 9: Transpiration Efficiency of four different varieties during the sixth to eleventh week

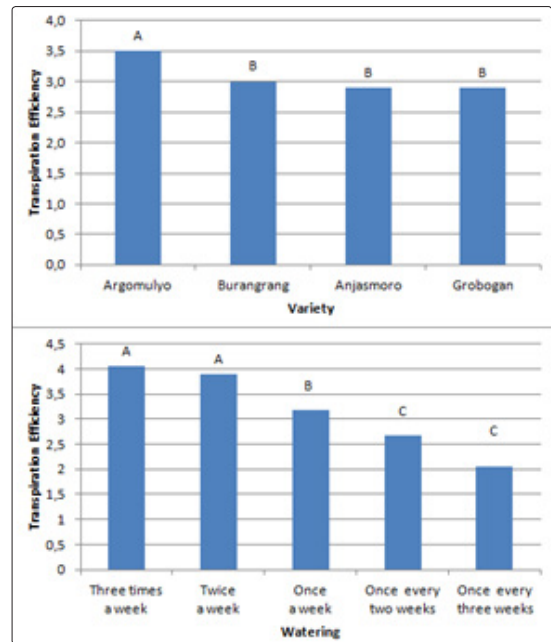


Figure 10: Transpiration Efficiency of different (a) varieties and (b) watering

TE fluctuations were observed on a weekly based on investigating with the photosynthetic rate and the transpiration rate before watering. The results showed that transpiration efficiency of Argomulyo and Burangrang varieties had a higher than the other two varieties. The TE values of Agromulyo variety were significantly higher than another varieties at weeks 7, 8 and 9; while the Burangrang varieties at weeks 7 and 8. The difference value of TE in watering differences began at week 7 (Table 1). During the experimental period, the TE values were significantly different at each treatments began entering week 7. Watered three and two times a week showed high values of TE whereas watered once a week showed a moderate TE value, and low value of TE occurred when the water supply once every two and three weeks (Figure 11).

Table 1: Transpiration Efficiency Throughout the Six Weeks of Investigate

| Transpiration Efficiency before watering | | | | | | |
|--|--------------------|--------------------|--------------------|--------------------|--------------------|------------------|
| Week | | | | | | |
| | VI | VII | VIII | IX | X | XI |
| Soybean Varieties means |means..... | | | | | |
| Argomulyo | 2,54 C | 6,64 A | 5,80 A | 6,50 A | 1,46 A | 0,74 A |
| Burangrang | 2,91 B | 5,98 B | 5,19 B | 5,45 B | 0,38 B | 0,71 A |
| Anjasmoro | 3,17 AB | 5,11 C | 4,60 C | 4,90 B | 1,29 A | 0,81 A |
| Grobogan | 3,26 A | 5,77 B | 4,74 C | 3,83 C | 1,24 A | 0,65 A |
| P value Soybean varieties effect | P<0,0001 | P<0,0001 | P<0,0001 | P<0,0001 | P<0,0001 | P<0,66 |
| Watering regime means |means..... | | | | | |
| Three times a week | 3,4 A | 6,41 A | 6,35 A | 5,46 A | 1,71 A | 1,02 A |
| Twice a week | 3,1 A | 5,9 B | 6,45 A | 5,51 A | 1,29 B | 1,13 A |
| Once a week | 3,2 A | 5,31 C | 4,85 BC | 4,55 B | 0,52 C | 0,65 B |

| | | | | | | |
|---------------------------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|
| Once every two weeks | 2,1 B | - | 4,71 C | - | 0,86 C | - |
| Once every three weeks | - | - | 3,04 D | - | - | 0,26 C |
| P value Watering regime effect | P<0,0001 | P<0,0001 | P<0,0001 | P<0,0001 | P<0,0001 | P<0,0001 |

Grain Weight Per Plant

In well watered condition (watering three times a week), grain weight per plant for Burangrang and Grobogan Varieties significantly different ($P \leq 0.05$) with Argomulyo and Anjasmoro varieties. Watering three times and two times a week showed no significant difference in all varieties. Grain weight would drop significantly when irrigation conducted once a week and two weeks. The stress of soil kept between saturation and field capacity was evidenced by the grain weight, and was proportionally more important in soybean. Decreasing seed weight significantly on watering once a week to two weeks against three times a week is highest for Argomulyo varieties ranged from 59.3 to 94.3%, furthermore Grobogan and Anjasmoro varieties respectively between 30.3 to 92.7 % and 40, 3 and 91.2%. While the lowest for Burangrang variety between 32.3 to 86.5%.

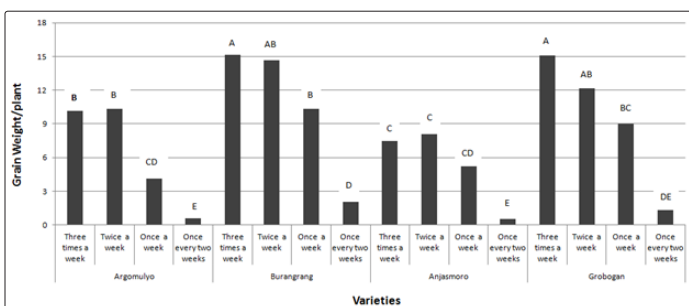


Figure 11: Grain weight per plant from different varieties and levels of watering. Histogram bars with the same letter are not significantly different at $P \leq 0.05$. Watering once every three weeks is not shown, it was harvested on flowering stage

Discussion

This study identified several common soybean varieties in Indonesia that have potential to adapt drought condition. These traits describe a water consumption use of several watering level and also identified differences in gas exchange parameters among varieties related with the physiological mechanism of different soybean varieties.

Initially this study was done to investigate the soil water content described by volumetric water content (VWC). Soil water content greatly affect the photosynthesis process. Along the period of stress occurred, obstacles to chloroplast metabolism substantially lead to photosynthetic activity dropped precipitously [18,19]. This study identified differences in correlation between VWC with photosynthetic among four varieties of soybean. Burangrang and Grobogan varieties maintained higher VWC in well condition (watered two and three times a week) however VWC value decreased accordance with decreased level of watered treatment in stress condition (once a week, once every two weeks and once every three weeks). This product indicated that maintained photosynthesis might be used to maintain the VWC.

The physiological mechanisms of different soybean varieties that may be involved in the control of photosynthetic of soybean

during progressive soil drying. Mechanism of opening-closure stomata played a role in the process of photosynthesis such as carbon assimilation and water consumption. At the top of plant where metabolism is accelerated especially in flowering stage, increasing photosynthesis, the stomatal conductance as well as transpiration rate is observed. Finally, it was clear, that the stomatal conductance (g_s), Transpiration rate (E) and Photosynthetic rate (A) were the most sensitive processes to biological and environmental variable such as soil drying in soybeans [20-22]. The similarity in the sensitivity of g_s , E and A in response to soil drying indicated that a decrease of A during soil drying was due mainly to a decrease of g_s and E in line with earlier findings. Burangrang have better value of photosynthesis rate and transpiration efficiency than the varieties Argomulyo, Anjasmoro and Grobogan have. In sugarcane, varietal variation were significant interaction effect with water regime on stomatal conductance and instantaneous transpiration rate in term of both individual leaves in the top leaf layer and the whole canopy [23].

Indeed, these findings are very helpful when determining how local conditions should be manipulated to adapt the survival and to improve the growth of soybean in arid and semiarid regions. A maintenance of turgor in leaf during reduction water status due to drought, is considered the means by which plants maintain metabolic processes and sustain the growth and survival [24]. Increased transpiration efficiency is often suggested as a critical opportunity for genetic improvement for increased crop yields in water-limited environment [25].

In the area of agricultural land, in the dry climates, where crop production as a primary consideration, watering has a main goal to keep the leaves turgiditas by giving water to the plant and then the plant transports water from the soil to the leaves thereby enabling continuous leaf gas exchange and leaf death delay. This is shown by the application of water regimes ranging from once a week, once every two weeks, once every two weeks; and the rate of photosynthesis will decrease progressively from well watered in these four varieties. The soil water deficit is considered to be the main environmental factor limiting global plant photosynthesis [26].

When the different varieties of plant water status are compared, they should be done in a reasonable short period of time and when the plant is in a relatively stable condition under a relatively stable environment as well. As a stress condition, stomatal closure becomes the main controls of leaf resistance. A stomatal response to humidity is of special interest with respect to plant water use in unfavorable environments. A stomata can open and close as hydraulically and chemically driven valves in the leaf surface. It opens to allow CO_2 uptake and closes to prevent excessive loss of water. Movement of these valves is regulated by environmental cues, mainly light, CO_2 and atmospheric humidity [27,28].

Transpiration in a physiological meaning is indispensable phenomenon in plants as the ratio of above ground biomass accumulation to the amount of water transpired, it can be influenced by cultivar and drought [29,30]. Transpiration efficiency differences happened between varieties, in this case Burangrang and Argomulyo varieties showed higher transpiration efficiency compared with other varieties. This is partly due to differences in morphological characteristics closely related to canopy-level stomatal control and canopy structure in which stomatal conductance and stomatal response to water deficits are potential traits for selection in a range of cultivars are

very strong control of transpiration. Transpiration efficiency in maize was found also with the gas-exchange measurements or gravimetric determination of transpiration in green-house studies [31].

Besides that transpiration is strongly controlled by stomatal conductance at the leaf level and environmental condition in this case water availability, on the fourth varieties The transpiration efficiency in treatment would decline after significantly watered once a week, ranging between 16-21%. It would decrease more than 30% when irrigated once every two weeks to three weeks.

Soil water conditions were significantly different to dry weight seeds/plant and pods containing occurred on generative phase especially on pod filling. Lack of availability of water on that condition would yield decreased caused inhibit the synthesis of chlorophyll in the leaves due to decreased photosynthetic rate and an increase in temperature and transpiration [32-35]. Stomatal conductance generally correlated with transpiration process to influence rate of decomposition of the nutrient from soil and to the entering into metabolism process.

Conclusion

In soil-plant parameters, there was a close relationship between the Volumetric Water Content and Photosynthetic rate on Burangrang and Grobogan varieties.

A leaf gas exchange of soybean was followed in well-watered and drought-stressed plants with the change of global environment, the impact of drought stress on crop yield becomes more significant. When the water supply was applied once a week, once every two weeks, and once every two weeks; the rate of photosynthesis would decrease progressively from well watered in these four varieties.

Furthermore, the correlation of stomatal conductance and transpiration rate had a close relationship with photosynthetic rate, where stomatal conductance of Burangrang variety had a higher value than other varieties. There was significant interaction between varieties with the well watered in transpiration rate and Photosynthetic rate. Burangrang and Argomulyo appeared to be well adapted than those of Anjasmoro and Grobogan varieties. The transpiration efficiency significantly declined after watered once a week, once every two weeks and three weeks. The transpiration efficiency of Argomulyo and Burangrang varieties were significantly higher than another varieties.

In well watered condition, the grain yield of Burangrang and Grobogan were better than Argomulyo and Anjasmoro varieties. Watering three times and two times a week showed no significant difference in all varieties. Decreasing grain weight would drop on watering once a week to two weeks.

Acknowledgement

The financial support of IAARD (Indonesian Agency for Agriculture Research and Development) through SMART D Project. Georg-August-Universität Göttingen, Department for Crop Sciences, Crop Production Systems in the Tropics for the use their facilities. Mrs Birgit Bode for her professional technical assistance.

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