

## Agronomic Traits and Physiology of Maize at Various Levels of Water Application

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### Abstract

Maize cultivation in Gorontalo Regency is commonly done in dry land, and this has made water as the main limiting factor for the excellent growth of the plant, as the plant often experiences water stress. This study was conducted to examine the agronomy traits and physiology of hybrid and composite maize on various water application levels. The study was done using a Split Plot Design in each with three repetitions. The main plots were for four different levels of water application: without water ( $A_0$ ), once water application ( $A_1$ ), twice water application ( $A_2$ ), and three-time water application ( $A_3$ ). The sub-plots were for the maize varieties, which consisted of two levels namely hybrid maize variety Bisi-2 ( $V_1$ ) and composite maize variety Bisma ( $V_2$ ). The results of the study show that three-time water application has helped increasing the height of the plants, the dry weight of the plants, as well as the dry weight of grains. Plants without water supply showed a decrease in the dry weight of the grains as much as 36% compared to the plants with three-time water application. Three-time water application method on Bisi-2 variety showed the highest result and no water application on Bisma variety showed the lowest result. Water stress has caused production of Bisma variety 22% lower than Bisi-2 variety. The physiology analyses show that  $CO_2$  intercellular on Bisma variety is higher than Bisi-2 variety, yet the rate of photosynthesis, transpiration, and water efficiency on both Bisma and Bisi-2 variety are not different.

**Key words:** water application, variety, photosynthesis

### 1. Introduction

The government of Gorontalo province through their agropolitan Maize-Based Plan has established the program aiming at producing one million tons of maize (known as prosta tanjung), yet the target of that one million has not been achieved up to these days. Meanwhile, the demand of maize has kept increasing, both the domestic demand and the international demand. Gorontalo Regency as the pilot project area of maize cultivation in Gorontalo Province owns 77,577 ha of dry land potential for maize cultivation and approximately 39,133 ha of the land have not been cultivated (Anonymous, 2011). Maize cultivation in Gorontalo is usually done in rain-fed dry land, and is never done in wet land—and as such, the problem of lack water supply persists.

Drought is defined as insufficient amount of water supply causing the decrease in plant production (Hatta, 2013) or gaps between the amount of water supply and the amount of water needed by the plants (Blum, 2011). Drought is an abiotic stress which needs such great attention in farming (Jaleel *et al.* 2009, Cazares *et al.*, 2011) and is considered as a significant environmental factor affecting the growth and production of plants (Kramer dan Boyer, 1995; Arcoverde *et al.*, 2011). Drought, in addition, has also long become the most crucial problem hampering the growth and productivity levels of land ecosystem plants in many parts of the globe especially in arid and semi-arid areas (Xu, *et al.* 2010).

Water stress affects physiological, biochemical, and morphological activities of the plants, which finally results in hindrance in their growth and development. Plants respond quickly to water level changes by adapting their morphological, physiological, cellular, and molecular processes in parallel (Cazares *et al.*, 2011). Moreover, water stress is very harmful for plants since it can decrease their production and threaten their existence in the nature (Cazares *et al.*, 2011; Jaleel *et al.* 2009).

The effects of water stress depend on the phases of the stress (Ramesh and Mahadevaswamy, 2000). The accuracy of water supply and application in accordance with the rate of growth of maize is crucial. The periods of maize growth require suitable water supply level. The growth phases consist of initial growth phase (around 15-25 days), vegetative phase (25-40 days), flowering phase (15-25 days), grain filling phase (35-45 days), and maturity phase (10-25 days) (FAO, 2001). Maize is relatively tolerant to shortage of water during vegetative phase (phase 1) and maturity phase (phase 4), but is so vulnerable to water shortage during flowering phase and grain filling phase. Water shortage during the vegetative phase (phase 1) and maturity phase (phase 4) does not bring so much effect to productivity level (FAO, 2001). Dramatic decrease in productivity may happen

when maize experiences water shortage especially during flowering phase, that is the phase where tassels and silks appear and during fertilization (phase 2), that causes decreasing numbers of grains due to hindered seed filling as the result of desiccated silks or cobs; while water shortage during the seed filling or seed forming (phase 3) may cause diminution on the size of grains leading to low productivity (Kramer, 1972; FAO, 1979; Baneti and Wesgate, 1992).

Maize experiences water stress during the flowering phase of seed filling phase shows 30-60% decrease in its production compared to normal condition (no water shortage), while maize experiences water stress during the flowering phase until harvesting time shows 15-30% decrease in its production compared to normal condition (no water shortage) (Banziger *et al.*, 2000). Water stress happening to maize during days 50-60 will cause decrease in productivity as much as 70%, and if the water stress happens on days 60-70, the decrease in productivity is approximately 60%; these all are due to the fact that on days 50-70 maize undergoes flowering phase, cob forming, and seed filling (Kramer, 1995; Baneti and Wesgate, 1992).

Maize belongs to plants with medium level of water use. Maize needs 500-800 mm of water, depending on the climate, as to gain maximum productivity (Purseglove, 1972; Kramer, 1972). The levels of water application and rainfall bring such great influence to maize productivity. In one planting season, maize needs around 2-5 times of water application (Aqil, 2007). The water application on maize in irrigated farms with limited water supply and rain-fed farms is normally done four times, that is when maize is 15, 30, 45, and 60 days old, as to increase the productivity of the maize itself (Munandar, 2010).

Maize productivity is determined by the level of water supply, as well as its ability to conduct photosynthesis. Maize belongs to C4 category of plants, that is for plants having high photosynthesis efficiency dealing with the use of CO<sub>2</sub>, water, and sun light (Salisbury and Ross, 1995). Sun radiation at 400-700 nm wavelengths holds such crucial role in photosynthesis process. The radiation belongs to visible radiation or is widely known as PAR (photosynthetic active radiation)(Taiz and Zieger, 1998; Hall and Rao, 1999), and approximately 70% of PAR infiltrates into the mesophyll and is absorbed by chloroplast, that is chlorophyll and carotenoid (Larcher, 2003).

The ability of plants to conduct photosynthesis can be examined by measuring the exchange rate of carbon dioxide (CER) on canopy. The CER value is measured based on the rate of CO<sub>2</sub> entering the stomata. Stomatal closure due to increased stomatal resistance as the effect of water shortage affects CER; if water stress happens then mesophyll resistance will increase due to permanent damage in the photosynthetic apparatus(Gardner, 2008). The rate of photosynthesis and dry matter accumulation of plants generally show a linear relationship with the water content in the plant body (Ariffin, 2002).

The decrease in photosynthesis efficiency due to water stress is closely related to the biochemical process and enzyme activities in cytoplasm, since photosynthesis is a hydrolysis process that really depends on water (Harjadi and Yahya, 1988). Stomatal closure can hinder photosynthesis, and in the long run this can stop the process since the diffusion of CO<sub>2</sub> from the atmosphere to the plants is obstructed causing a decrease in the assimilate results and low plant weight (Salisbury and Ross 1995; Ariffin, 2003).

Based on the afore-presented explanation, this study was then aimed at examining the agronomic traits and physiology of hybrid maize variety Bisi-2 and composite maize variety Bisma at different levels of water application.

## 2. Research Method

The study was conducted in Bulila Village, Telaga Sub-District, Gorontalo Regency. This village is located ±10 m asl. The study was carried out from October 2011 until January 2012 using a Split Plot Design. The main plots were for four different levels of water application: without water (A<sub>0</sub>), once water application (A<sub>1</sub>), twice water application (A<sub>2</sub>), and three-time water application (A<sub>3</sub>). The sub-plots were for the maize varieties, which consisted of two levels namely hybrid maize variety Bisi-2 (V<sub>1</sub>) and composite maize variety Bisma (V<sub>2</sub>). Each combination was treated three times, which means that the experimental group consisted of 24 plots. The variables studied were the growth of the maize and the productivity, which consisted of:

1. The height of maize (cm): measured from the ground level up to the tip of leaves (leaves are straightened up parallel to stalk).
2. The number of leaves: the leaves counted were the green ones and the perfectly opened ones.
3. Dry weight of the plant (g), which was taken from the plant body (roots, stalks, and leaves without cobs or grains) put in the oven for 48 hours at 80<sup>0</sup> C until they reached constant weight.
4. The weight of dry grains (g)
5. The weight of 100 grains (g)

The data was analyzed using analysis of variance (F-test) with level of significance of 5%. If significant effect appeared to happen on treatment or real interaction, then Duncan's Multiple Range Test (DMRT) with  $\alpha$

5% would also be done to examine the difference in treatment (Hanafiah, 2011). Data analyses were all conducted using Microsoft Excel program.

Land cultivation was done a week before planting, the soil was cleaned from root remnants and weeds. Then, experimental plots sized 3.75 x 2.2 m were made as many as 24 plots. Maize was planted with spacing of 75 x 20 cm, with distance between replicates of 1 m, and with plot distance of 50 cm. Drainage was built around the plots with the depth of 30 cm. The seeds were planted at the depth of 5 cm from the ground surface, and each hole was planted with two seeds. A day before planting, the experimental plots were watered until it reached the saturation point. When the seeds had grown into sprout, one seed was taken from each hole and that each hole held only one sprout to grow.

Water was applied three times, when the plants were 15, 30, and 45 days old after the planting date. The amount of water applied to each plot was 41.50 liter, under the assumption that the minimum water needed by maize in is 5 l m<sup>-1</sup>day<sup>-1</sup>.

Urea (N), SP 36 (P), and KCl (K) fertilizers were given when the maize was planted, and were put in the lines on the right and left of the plots. The distance between the plots and the lines of fertilizers was around 10 cm, and the depth of the lines was 5 cm. There were six lines for each plot. The amount of fertilizers used was 30 kg ha<sup>-1</sup> N or 4.2 g urea for each line, 7.5 kg ha<sup>-1</sup> P or 3 g SP-36 for each line, and 50 kg ha<sup>-1</sup> or 6 g KCl for each line.

The seeds that failed to grow were substituted on day 7 after the planting date. Weeding was done three times, when the plants were 14, 28, and 42 days old. Control on pest attack was made suitable with the level of the attack. Harvesting time came when the cobs of the maize were physiologically matured, marked with the brown color of the cobs and the dryness, as well as the yellow color of the leaves.

The measurement on the photosynthesis rate was estimated with CER, transpiration rate was symbolized as E and intercellular CO<sub>2</sub> was symbolized with Ci. Measurement was done using *Portable Photosynthetic System Li-6400* (Licor Inc. Lincoln, Nebraska, USA) based on the manual of Li-6400 Photosynthetic System (Anonymous, 2007). CER, E and Ci were all measured at PAR 0, 50, 100, 200, 400, 800, 1500 and 2000 μmol.m<sup>-2</sup>.s<sup>-1</sup>. CER, E and Ci measurement was done during the tasseling phase by lamina the samples of the leaves (fully mature and healthy leaves, leaves number 3 or 4 from the tips of the rods). These leaves were clamped on the head censor of the Portable Photosynthetic System Li-6400. Observation was done from 8 AM until 2 PM. The data of CER, E and Ci was then recorded in the Portable Data Analyzer, a part of the Portable Photosynthetic System Li-6400. The relationship model between CER, E and Ci and PAR was estimated by using an approach proposed by Thornley (1976) employing Microsoft Excel software. The data was presented in graphs, showing relationship between CER and PAR, E and PAR, and Ci and PAR. All the data was analysed using regression analyses employing Microsoft Excel software.

### 3. Results and Discussion

#### 3.1 Agronomic Traits

The levels of water application influenced the height of the plants, the dry weight of the plants, and the weight of dried grains, but they did not influence the number of leaves as well as the weight of 100 grains. Two-time water application or three-time water application resulted in better growth and productivity of the plants than one-time water application. No water application caused plants to experience water stress leading to low productivity. The result of data analysis using DMRT on the height of plants, the number of leaves, the weight of dried plants without cobs, the weight of dried grains, and the weight of 100 grains on different water application is presented in Table 1.

Table 1. The average of the height of plants, the number of leaves, the weight of dried plants without cobs, the weight of dried grains, and the weight of 100 grains on different water application

Levels of Water Application	Height of plants (cm)	The number of leaves <sup>-1</sup> (in piece)	The weight of dried plants without cobs (g)	The weight of dried grains (g)	The weight of 100 grains (g)
No water	162.69 a	13.33 a	65.67 a	98.20 a	31.73 a
1 time	174.34 a	13.83 a	73.83 ab	126.01 b	31.85 a
2 times	190.57 b	13.87 a	82.39 bc	150.86 c	31.98 a
3 times	194.41 b	14.06 a	84.55 c	152.26 c	31.98 a

Note: figures followed by same letters on the same column did not differ significantly DMRT 5%

The decrease on the height of plants happened in line with the decrease in the amount of water applied. The decrease in water application from three times into two times and finally one time caused a decrease in productivity as much as 2%, 10%, and 16%. Water shortage results in impaired cell division and elongation

since the flow of water is obstructed—thus, water cannot run from xylems into the growing cells (Yang *et al.*, 2009). Water shortage can hinder the growth of the plants in form of disruption in the height of plants and the width of leaves, as well as the growth of the reproductive organs, compared to the normal growth of plants when water is not limited (Banziger, 2006).

The levels of water application did not affect the number of leaves (Table 1). Plants experiencing water stress and plants having normal supply of water do not show difference in the number of leaves. This is in accordance with the argument by Yang *et al.* (1993) stating that maize undergoing water stress (-10 bar) and maize without water stress (-3 bar) in their vegetative and reproductive phases in different seasons do not show any significant difference in the number of leaves but in the width of leaves. The dry weight of maize was affected by the levels of water application (Table 1). Maize with the three-time water application showed the highest dry weight and maize without any water application showed the lowest dry weight. Water stress decreased the dry weight of maize as much as 22% (in situation where water stress happens) compared to the normal condition (in situation where water application is done three times). Water stress causes the disclosure on stomata and makes a decrease on absorption rate of CO<sub>2</sub>. Hence, the process of photosynthesis is hindered because of a decrease on assimilate results and causes low weight of the plants (Salisbury dan Ross 1995; Tezara *et al.*, 2002, Ariffin, 2003).

The levels of water application affected the weight of the dried grains, but not the weight of 100 grains. The dried grains from the plots with three-time water application or two-time water application were heavier than those from the plots with onne-time water application or no water application. Water stress decreased the weight of dried grains as much as 36% (in situation where water stress happens). This result is supported by the findings of Yang *et al.* (1993) stating that water stress as much as -8 bar and -10 bar causes decreasing weigth of dried grains as much as 20% and 40%. Water stress can lead into low production since the assymilate translocation or photosyntates of plant organs, such as seed filling, is obstructed and influences on the plant results (Harjadi and Yahya, 1988).

Variety is also a factor determining the dry weight of plants and the weight of the dried grains, but it does not affect the height of plants, the number of leaves, and the weight of 100 grains (Table 2). The dry weight of Bisi-2 variety was significantly different from the dry weight of Bisma variety, in which the dry weight of Bisi-2 variety was 13% higher than the dry weight of Bisma variety. The same result also applied to the weight of the dried grains, in which Bisi-2 variety had higher weight of dried grains, as much as 26%, the weight of dried grains of Bisma variety. This was caused by genetic differences between both varieties. Hybrid maize Bisi-2 variety has genetic ability to produce two cobs of maize in a single plant (*prolific*) so it may result on higher production than Bisma composite variety with a single cob. It resonates well with Sudjana *et al.*, (1991) stating that generally the best hybrid maize produces higher results than free pollen maize variety. It occurs as the efficiency of photosynthesis from some plants has been increased through breeding (Salisbury and Ross, 1995).

Table 2. The average height of plants, number of leaves, dry weight of plants, weight of dried grains, and weight of 100 grains for Bisi-2 variety and Bisma variety

Variety	Height of plants (cm)	Number of leaves <sup>-1</sup> (piece)	Dry weight without cobs (g)	Weight of dried grains (g)	Weight of 100 grains (g)
Bisma	178.01 a	13.52 a	71.36 a	111.96 a	31.99 a
Bisi-2	182.99 a	14.03 a	81.86 b	151.70 b	32.06 a

Note: figures followed by same letters on the same column did not differ significantly DMRT 5%

There was an interaction between the levels of water application and varieties as shown by the the height of plants, number of leaves, dry weight, weight of the dried grains, and the weight of 100 grains (Table 3).

Table 3. The interaction of water application on the variables of plant height, number of leaves, dry weight, weight of dried grains, and weight of 100 grains.

Levels of Water Application	Variety	Height of plants (cm)	Number of leaves <sup>-1</sup> (piece)	Dry weight without cobs (g)	Weight of dried grains (g)	Weight of 100 grains (g)
No water	Bisi-2	161.98 a	13.53 ab	66.92 ab	110.31 b	31.74 ab
	Bisma	163.40 a	13.11 a	64.41 a	86.08 a	31.73 ab
1 time	Bisi-2	173.40 b	14.11 ab	77.03 c	137.56 c	31.54 a
	Bisma	175.26 bc	13.62 ab	70.63 b	114.46 b	32.16 ab
2 times	Bisi-2	188.43 d	14.44 b	85.19 d	174.51 d	32.36 ab
	Bisma	192.70 d	13.67 ab	70.78 c	117.293b	31.60 a
3 times	Bisi-2	208.14 e	14.00 ab	98.31 e	184.42 d	32.34 ab
	Bisma	208.14 e	13.67 ab	79.60 b	130.02 c	32.76 b

Note: figures followed by same letters on the same column did not differ significantly DMRT 5%  
 The highest dry weight was for Bisi-2 variety with three-time water application, whereas the lowest dry weight was for Bisma variety with no water application. The same matter also happened to the weight of the dried grains, in which the highest weight of dried grains was for Bisi-2 variety with three-time water application, whereas the lowest weight of dried grains was for Bisma variety with no water application. The water stress condition caused Bisma variety to have lower productivity, as much as 22%, compared to Bisi-2 variety. This was caused by hybrid maize Bisi-2 variety has a deep rooting for overcoming lack of water (Anonymous, 2010). The ability of plants in overcoming waters stress depends on the ability of plant organs and tissues to tolerate water stress (Fitter and Hay, 2002)

### 3.2 Physiological Traits

CER of the Bisma variety and Bisi-2 variety increased in line with the increase in PAR from 0 to 2000  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ . The increase in PAR was followed by the increase in CER with the relatively same patterns. The relationship between CER and PAR of Bisma variety and Bisi-2 variety can be seen in Figure 1.

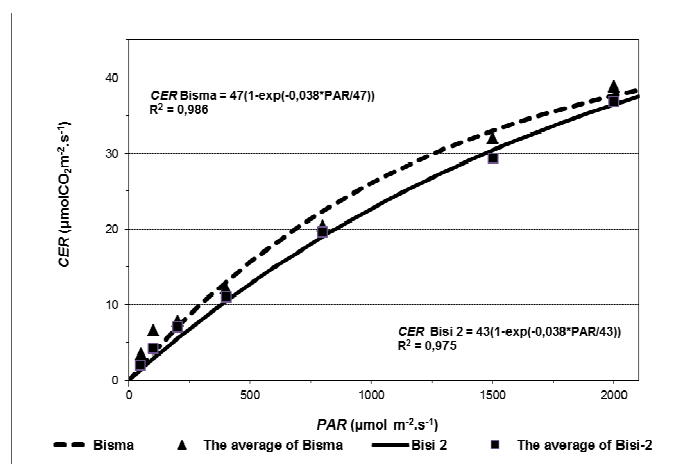


Figure1. The relationship between CER and PAR of Bisma variety and Bisi-2 variety on day 59. The lines in the figure show the estimation on the model of relationship between CER and PAR

Bisma variety resulted in CERmax 47  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ , while Bisi-2 variety resulted in CERmax 43  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ . The photosynthesis rate of Bisma and Bisi-2 variety did not show any significant difference, but Bisma variety tended to have higher photosynthesis rate than Bisi-2 variety. Gardner *et al.* (2008) state that many studies report different CER values among varieties in the same species of crops as many as two until three times of  $\text{CO}_2$  absorption between the lowest sample and the highest sample. The leaves of C-4 crops like maize do not show any saturated rates under or even over full sunlight, and they can have maximum photosynthesis rate twice more than the photosynthesis rate on C-3 species on each maximum temperature (Salisbury and Ross, 1995).

If the leaves absorb more radiation energy than what they need, the excess energy must be thrown away, one of which is through transpiration. Transpiration rate is determined by the width and the structure of the leaves, stomata, and environmental factors. Stomata play such an important role in the transpiration process;

many studies reveal that transpiration rate decreases when more stomata are closed (Salisbury and Ross, 1995). The relationship of the E and PAR of Bisma variety and Bisi-2 variety can be seen in Figure 2.

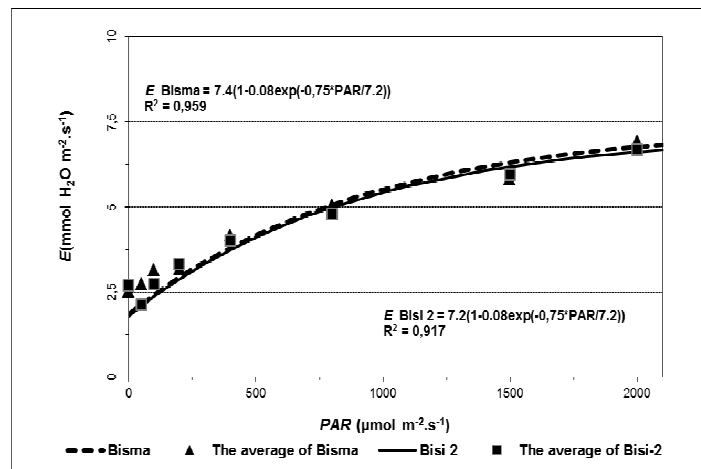


Figure 2. The relationship of the E and PAR of Bisma variety and Bisi-2 variety on day 59. The line in the figure show the estimation on the model of relationship between E and PAR.

Bisma and Bisi-2 variety showed similar E patterns. The maximum transpiration rate ( $E_{max}$ ) of Bisma variety was  $7.5 \text{ mmol H}_2\text{O m}^{-2}\text{s}^{-1}$ , while for Bisi-2 was  $7.2 \text{ mmol H}_2\text{O m}^{-2}\text{s}^{-1}$ ; yet, there was not any significant difference between these two values. E increased in line with the increase in PAR. This is supported by the results of previous studies showing that light has such great influence over stomata, apart from its role in the process of photosynthesis. It is assumed that light works in mesophyll cells and sends signals to guard cell. In addition, more intense light will increase temperature which will cause stomata to open even wider (although in some species the high temperature of  $30\text{-}35^\circ\text{C}$  simply causes stomata to close), and the disclosure on stomata will lead into increasing transpiration and it can expel heat (Salisbury and Ross, 1995) and makes it possible for roots to absorb water (Sugito, 2009). Many factors affecting the closure and disclosure of stomata, but the most important factor is light, as well as water. The closure and disclosure of stomata bring effect that is much more important to the total transpiration rate than the number and size of stomata (Gardner *et al*, 2008).

Photosynthesis can be expressed as a function of  $C_i$  (Salisbury and Ross, 1995).  $C_i$  increases in line with PAR, but to the certain extent, that is optimum PAR level, will not increase  $C_i$  concentration (Figure 3).  $C_i$  max of Bisma variety was significantly different from the  $C_i$  max of Bisi-2 variety.  $C_i$  max of Bisma variety was  $746 \text{ } \mu\text{mol CO}_2 \text{ mol}^{-1}$ , while  $C_i$  max of Bisi-2 variety was  $706 \text{ } \mu\text{mol CO}_2 \text{ mol}^{-1}$ .

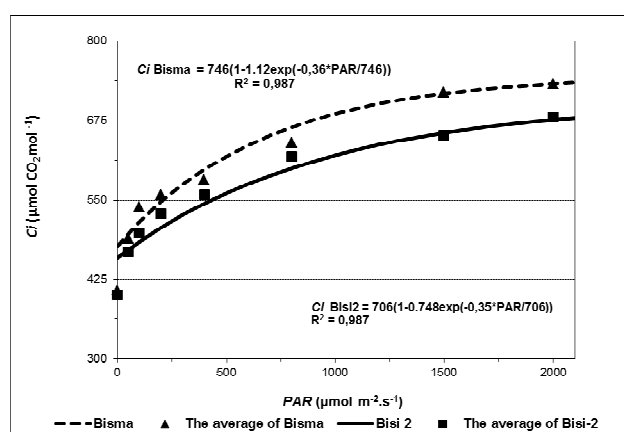


Figure 3. The relationship of  $C_i$  ( $\text{CO}_2$  intraseluler) and PAR on Bisi-2 and Bisma variety on day 59. The lines in the figure show the estimation on the model of relationship between PAR and  $C_i$ .

The efficiency on water usage on maize can be measured by comparing CER and E. The ratio of CER/E on Bisma variety was  $6.35 \text{ } \mu\text{mol CO}_2 / \text{mmol H}_2\text{O}$ , while on Bisi-2 variety was  $5.97 \text{ } \mu\text{mol CO}_2 / \text{mmol H}_2\text{O}$ ; this means that there was not any significant difference in the ratios of CER/E between those two varieties of maize on other words, the two varieties have the same water usage efficiency level.

The highest amount of dried grains was for Bisi-2 variety although Bisma variety had higher Ci than Bisi-2 variety. There many factors affecting the final product. The low dried grains produced may due to the low division of dried materials into the total amount of grains. Seed filling is also determined by the ratio of the original source of the plants, and also depends on the genotype and environment condition (Fischer and Palmer, 1996).

#### 4. Conclusion

1. Three-time water application on maize has improved the height of the plants, the dry weight of the plants, and the weight of the dried grains of the plants; yet, it does not improve the number of leaves and the weight of 100 grains. Water stress decreases the amount of dried grains as much as 36% compared to the normal situation (no water stress).
2. Difference in varieties affects the dry weight of maize and the weight of the dried grains; yet, it does not affect the height of the plants, the number of leaves, and the weight of 100 grains. Bisi-2 variety shows higher dry weight and weight of the dried grains, as much as 13% and 26% respectively, compared to Bisma variety.
3. Interaction of three-time water application on Bisi-2 variety shows the highest results on the height of the plants, the dry weight, and the weight of the dried grains, whereas the lowest results were for Bisma variety with no water application. Water stress caused a decrease on the dry weight of the plants and the dry weight of the grains respectively as much as 32% and 40% on Bisi-2 variety and a decrease as much as 20% and 33% on Bisma variety. Hence there was a variety tendency that Bisma variety was more tolerant than Bisi-2 variety.
4. The physiological traits such photosynthesis rate, transpiration and the efficiency of water usage on Bisi-2 variety and Bisma had no significant difference. But in terms of Ci variable, the Ci of Bisma variety was significantly higher than Bisi-2 variety. However, it did not increase the results of Bisma variety.

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