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## Morphological and physiological characterization of four soybean varieties with different drought stress

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**Abstract** The experiment reported the morphological and physiological response of soybean varieties grown under different field capacity in sandy soil. Results indicated there was an interaction between soybean varieties and field capacity on days to flowering. Soybean varieties affect on plant height (2 and 4 WAP), number of leaves (6 WAP), number of flowers, days to flowering, leaf greenness, leaf area index, number of stomata, number of opened stomata and stomata density. Meanwhile, field capacity influenced the number of leaves (4 and 6 WAP), number of branches (4 and 6 WAP), leaf area, number of flowers, days to flowering, leaf area index and seed weight per plant. Grobogan variety had the fastest flowering time (26 days after planting). In addition, there were no significant differences among the soybean varieties in terms of seed weight per plant. Soybean grown in 100, 75 and 50 % FC had the highest seed weight per plant, while soybean grown in 25% FC had the lowest seed weight per plant. In conclusion, morphologically effects of drought stress of soybean took place when soybean grown at 50% FC and lower. Meanwhile, physiological responses of soybean to different in FC were resulted from changes in stomata density.

**Keywords:** Drought, Sandy soil, Soybean, Stress

### Introduction

Soybean (*Glycine max* L.) is one of the most important leguminosae plants in Indonesia since it is widely used for industrial purposes as well as for food and feed. Soybean is an excellent source of protein compare to other food in which it protein content reaches 40 % (Pambudi, 2013). Data from Indonesian Central Bureau of Statistics indicated national soybean production in 2014 and 2015 were amounted to 954.993 and 963.183 tons, respectively, with a land productivity of 1.55 ton/ha and 1.56 ton/ha . This productivity was

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considered as low productivity compared to soybean productivity in other countries, *e.g.* Thailand has 1.68 tons/ha in 2015 (AFSIS, 2016), Meanwhile, the productivity in China in 2015 reached 45.8 k ha/yr (Zang and Lu, 2020).

An attempt to improve soybean production can be done by empowerment of marginal land and the use of improved varieties. According to Adisarwanto (2006) the use of proper varieties plays an important role in increasing soybean production since high yield is also largely determined by their genetic potential. Yield performances of soybean in the field are influenced by the interaction between genetic factors and good management of environmental conditions. Furthermore, Wirnas *et al.* (2006) reported that the development of high-yielding varieties and adaptation to strained environmental conditions can increase its productivity. Hartman *et al.* (2011) also emphasized that high yields is largely determined by genetic potential and the growing environment.

During the courses of crop development, the drought stress that occurs at each growth phase might have resulted on decreasing yields. However, the greatest is during the critical plant period, namely the flowering phase, seed formation, and pod filling. According to Suyanto and Soegiyanto (2002), in the field, drought stress during the pod filling phase reduced soybean yields by 55%. During the vegetative phase, drought stress might cause the reduction of leaves area and stem diameter, over all plant size and shoot dry weight. Whereas, drought stress during the flowering process bring about the reduction of the number of flowers and the number of pods.

The use of resistant soybean variety to drought stress is necessary to avoid the risk of yield reductions. Drought stress is an important factor influencing symbiosis and reducing nodule formation, size, and N fixation (Seversike, 2011). According to Kusvuran (2012) plant response to drought stress varies depending on the duration, stress intensity, plant species, and growth stage. Furthermore, soybean plants are subjected to drought stress during the vegetative phase which experienced a large decline in growth and development, such as decreased in height, root length, root dry weight, and canopy (Aboyami, 2008). The coastal land is a vulnerable to drought stress. Drought stress influences the soybean growth such as the number of pod filled out and 100 seeds weight (Asyura *et al.*, 2018). Therefore, this study aimed to determine the morphological and physiological response of soybean varieties grown under different field capacity in sandy soil.

## **Materials and methods**

### ***Research design***

The experiment used a completely randomized design with two factors. The first was soybean varieties consisting of Devatra 1, Devatra 2, Dering, and Grobogan. The second factor was the soil moisture content of the percent of field capacity (FC) was 100, 75, 50, and 25 % of FC.

### ***Population and samples***

After combining two factors, there were 16 treatment combinations and replicated five times. The total numbers of experimental units were 80. The soybean plants were planted at the polybags. The arrangement of polybags were done as follows: the distance between polybags was 20 cm x 20 cm and the distance between replications was 50 cm.

### ***Research stage***

#### **Preparation of planting media**

The samples were collected from coastal environment in Kuala Alam. Gading Cempaka sub-district, Bengkulu City. The land was dominated with sandy soil. Before sampling, the land was cleared from weeds. Soil was sampled at depth of 0 – 20 cm. the soil was air-dried and homogenously mixed.

#### **Measurement of moisture content (MC)**

Water content (MC) was determined using gravimetric method. A soil sample and chicken manure at volume ratio of 1:1 was incorporated homogeneously. Ten g of the mixture was weighed and put into a copper bottle. and oven-dried in 105 °C. After 24 hours. The sample was weighed and the moisture content was calculated.

#### **Filling polybags**

The growing media were incorporated by air-dried sandy soil and chicken manure at the volume ratio of 1:1. Five kilograms of the media were put into a polybag with the of 17 cm x40 cm.

#### **Determination of field capacity treatments**

Field capacity (FC) measurement was carried out by soaking the polybag with soil for 48 hours, then draining the water from the polybag with soil to the last drop to be 100% FC. Then, the soil water content was measured. After getting the 100% FC, then measuring 75, 50 and 25 % water content of FC were conducted accordingly.

### Data collection and analysis

Data were collected in terms of plant height (cm) at 2, 4, and 6 weeks after planting (WAP), number of leaves at 2, 4, and 6 WAP, number of branches at 4 and 6 WAP, leaf area (cm<sup>2</sup>) 6 WAP, number of flowers, days to flowering (days), leaf greenness level (SPAD index), leaf area index 6 WAP, number of stomata, number of opened stomata, stomata density, and seed weight per plant (g). Data were analyzed using analysis of variance (ANOVA) at 5% of probability level. The treatment means were separated using Least Significance Different (LSD) 5%.

### Results

Analysis of variance indicated that there was a significant interaction effect between soybean varieties and field capacity on days to flowering (Table 1). The treatment of soybean varieties had a significant effect on the planta height (2 and 4 WAP), the number of leaves (6 WAP), number of flowers, days to flowering, leaf greenness, leaf area index, number of stomata, number of opened stomata and stomata density. Meanwhile, the field capacity significantly effect number of leaves (4 and 6 WAP), number of branches (4 and 6 WAP), leaf area, number of flowers, days to flowering, leaf area index and seed weight per plant.

**Table 1.** Analysis of variance for each observed variable

Variabel	F-Value			Coefficient of diversity
	Varieties	Field Capacity	Interaction	
Plant height 2 WAP	13.35 *	0.36	0.33	21.3
Plant height 4 WAP	11.04 *	1.39	1.35	25.82
Plant height 6 WAP	2.65	2.53	0.54	21.92
Number of leaves 2 WAP	2.3	0.7	0.94	26.49
Number of leaves 4 WAP	2.36	8.41 *	0.41	22.28
Number of leaves 6 WAP	3.48 *	18.55 *	1.32	12.05
Number of branches 4 WAP	2.21	8.18 *	0.21	24.38
Number of branches 6 WAP	1.71	6.58 *	0.71	29.42
Leaf area	2.77	14.23 *	1.21	21.69
Number of flowers	9.41 *	3.68 *	0.51	14.91
Flowering period	210.75 *	3.41 *	2.36 *	1.97
Leaf greenness level	15.39 *	0.38	0.47	6.61
Leaf area index	2.94 *	18.47 *	0.72	17.69
Number of stomata	3.29 *	0.48	1.78	28.56
Number of opened Stomata	3.88 *	0.5	0.97	21.94
Stomata density	3.29 *	0.48	1.78	28.56
Seed weight per plant	2.45	3.71 *	0.67	29.71

\* = significant difference at 5 % significance level according to Least Significant Different. WAP=weeks after planting

***Effect of varieties on morphological traits of soybean***

Morphological responses of soybean were measured by plant height, number of leaves and number of branches. At 2 WAP, Devatra1 had the highest plant height compared to other varieties, followed by Grobogan. Plant height of both Devatra2 and Dering varieties were not significantly different (Table 2). However, at 4 WAP, Grobogan variety had the highest plant height, followed by Devatra 1, Devatra2 and Dering varieties. All soybean varieties had similar plant height at 6 WAP (Table 2). In addition, leaf numbers of all soybean varieties had similar responses at 2 and 4 WAP, but at 6 WAP Dering variety had the highest leaf numbers, though it was similar to Devatra1 and Grobogan. Number leaves of Devatra2 had the lowest among treatments, though it was similar to those of Devatra1 and Grobogan varieties. Results also indicated that all soybean varieties had similar number of branches at 4 and 6 WAP (Table 2).

**Table 2.** Effect of soybean varieties on plant height, number of leaves and number of branches at 2, 4 and 6 WAP

Varieties	Variable		
	Plant height	Number of leaves	Number of branches
<i>2 weeks after plant</i>			
Devatra 1	14.53 a	1.53	-
Devatra 2	9.82 c	1.33	-
Dering	8.84 c	1.16	-
Grobogan	12.25 b	1.45	-
<i>4 weeks after plant</i>			
Devatra 1	31.55 b	7.25	2.14
Devatra 2	29.07 bc	6.08	1.87
Dering	23.53 c	6.00	1.73
Grobogan	45.20 a	7.08	2.14
<i>6 weeks after plant</i>			
Devatra 1	43.58	26.83 ab	8.50
Devatra 2	42.83	24.33 b	8.16
Dering	51.58	28.33 a	8.91
Grobogan	51.75	25.66 ab	6.83

\* = significant difference at 5 % significance level according to Least Significant Different.

WAP=weeks after planting

### *Morphological responses of soybean to drought stress*

Field capacity (FC) of crop growing media reflects the level of available water for a plant. Results indicated that FC did not affect plant height, but did affect the number of leaves and the number of branches at 4 and 6 WAP. It appeared that both 100% and 75% FC had the highest number of leaves and the number of branches at 4 and 6 WAP, compared to 0% and 25% FC (Table 3).

**Table 3.** Effect Field Capacity on plant height, number of leaves and number of branches of soybean

Field Capacity (%)	Variable		
	Plant height	Number of leaves	Number of branches
<i>2 weeks after plant</i>			
100	11.81	0.91	-
75	11.45	1.00	-
50	11.40	0.91	-
25	10.79	0,75	-
<i>4 weeks after plant</i>			
100	34.68	7.91 a	2.32 a
75	29.85	7.33 a	2.28 a
50	33.12	5.91 b	1.79 b
25	28.7	5.25 b	1.5 b
<i>6 weeks after plant</i>			
100	54.58	30.16 a	8.58 a
75	45.58	27.83 ab	10.00 a
50	44.91	26.25 b	8.08 b
25	44.66	20.91. c	5.75 c

\* = significant difference at 5 % significance level according to Least Significant Different.

### *Effect of varieties on physiological traits of soybean*

Results indicated that all soybean varieties had similar responses in terms of leaf area (Table 4). However, Devatra1 had the highest leaf area index (LAI) of Devatra1, though it was not significantly different with Devatra 2 and Dering varieties. Although it was not significantly different with Devatra 2 and Dering varieties, Grobogan variety had the lowest LAI. In terms of leaf greenness level, Grobogan variety had the highest greenness level, followed by Devatra1

and Dering varieties. Devatra 2 had the lowest leaf greenness level, though it was similar to that of Dering variety. In addition, Devatra 2 had the highest number of stomata, followed by Grobogan variety. Both Devatra1 and Dering varieties had the lowest number of stomata, though it was similar to that of Grobogan variety. With respect to the number of opened stomata, both Devatra 2 and Grobogan varieties had the highest number, followed by Devatra 1 and Dering varieties. Although Dering had the lowest number of opened stomata, it was similar to Devatra 1 variety. In addition, Devatra 2 had the highest stomata density, followed by Grobongan variety. The stomata density of both Devatra1 and Dering varieties were among the lowest, though it was similar to that of Grobogan variety. Lastly, there were no significant differences among the soybean varieties in terms of seed weight per plant.

**Table 4.** Effect of soybean varieties on variable leaf area, leaf area index, leaf greenness level, number of stomata, number of opened stomata, and stomata density

Varieties	Leaf area (cm <sup>2</sup> )	Leaf Area Index (LAI)	Leaf greenness (SPAD index)	Number of stomata	Number of Opened Stomata	Stomata density	Seed weight per plant (g)
Devatra 1	169.07	21.08 a	39.09 b	18.91 b	3.73 ab	124.45 b	20.30
Devatra 2	194.98	19.22 ab	36.52 c	26.08 a	4.47 a	171.60 a	14.58
Dering	182.91	19.76 ab	37.45 bc	19.58 b	3.36 b	128.83 b	18.43
Grobogan	152.61	17.03 b	43.15 a	22.16 ab	4.19 a	145.83 ab	17.64

\* = significant difference at 5 % significance level according to Least Significant Different.

#### ***Effect of field capacity on physiological traits of soybean***

Field capacity (FC) significantly affected physiological traits of soybean, *i.e.* leaf area and leaf area index (LAI), but not leaf greenness, number of stomata, number of opened and stomata density (Table 1). Results indicated that soybeans grown with 100% and 75% FC had the highest leaf area and LAI, followed by those of Dering and Grobogan varieties (Table 5). Nevertheless, FC treatments had similar effects on leaf greenness, number of stomata, number of opened and stomata density of soybeans. Lastly, soybean grown in 100, 75 and 50 % FC had the highest seed weight per plant, while soybean grown in 25% FC had the lowest seed weight per plant.

**Table 5.** Effect of field capacity on variable leaf area, leaf greenness level, number of stomata, number of opened stomata and stomata density on drought stress

Field Capacity (%)	Leaf area (cm <sup>2</sup> )	Leaf Area Index (LAI)	Leaf greenness (SPAD index)	Number of stomata	Number of Opened Stomata	Stomata density	Seed weight per plant (g)
100	212.39 a	23.09 a	39.27	22.58	3.89	148.57	19.15 a
75	202.51 a	22.00 a	38.43	21.50	3.91	141.44	19.94 a
50	162.78 b	18.28 b	39.51	22.66	4.18	149.12	18.42 a
25	121.88 c	13.72 c	38.43	20.00	3.76	131.58	13.43 b

\* = significant difference at 5 % significance level according to Least Significant Different.

## Discussion

Results indicated that a significant effect on the treatment of soybean varieties on the number of leaves 6 WAP. Dering variety had the highest number of leaves in 2 WAP (28.33 leaves). However, there was no significantly affected the variety on the number of branches. There were significant difference of all soybean varieties in terms of plant height up to week 4 WAP. In 4 WAP, Grobogan variety had a higher plant height than the Devatra1, Devatra2 and Dering varieties. However, 6 WAP there were no significant differences between the four varieties. These plant heights were almost similar to its genetic potential. According to its variety descriptions, Grobogan variety has a height of 50-60 cm (Balitkabi, 2016), Devatra1 61.20 cm (Kep.Kementan No.852, 2020a), Devatra2 68.39 cm (Kep. Kementan No. 853, 2020b) and the height of Dering soybean variety is 57 cm (Balitkabi, 2013). Although the decrease in plant height gain in Grobogan variety had the smallest percentage, it can not be concluded that this variety was quite tolerant drought stress.

Various field capacity conditions might inhibit vegetative growth in soybean plants. Results showed that various field capacity conditions had a significant effect on the number of leaves and number of branches. It appeared that the number of leaves and the number of branches (6 WAP) with a field capacity (FC) of 25% was lower than 50% of a field capacity of 20.91 and 26.25 (number of leaves) and 5.75 and 8.08 (number of branches), respectively. The number of leaves and the number of branches formed at 75% field capacity likely did not experience drought stress because they were not significantly different from 100% FC. According to Chun *et al.* (2021), soybeans have sensitivity to drought in the early vegetative stages. One of the morphologically plant responses in the face of drought stress is a decrease in

leaf growth rate (Banziger *et al.*, 2000; Hamim, 2007). The low number of leaves formed at 25% and 50% FC indicated that soybean plants had experienced drought stress disorders. According to Purwanto and Agustono (2010) low FC brought about drought stress that inhibited plant growth which resulted in plants become more stunted. Inhibition of plant growth might be caused by disruption of the photosynthesis process and loss of cell turgidity which can inhibit cell division and development due to lack of water. Taiz and Zeiger (2006) suggested that plant growth is very sensitive to water deficit because it is associated with turgor and loss of turgidity which eventually produced smaller plants due to reduction of cell division and enlargement.

Soybean varieties had a significant effect on the leaf area index and leaf greenness. It appeared that Devatra1 was significantly different from Grobongan, which accounted 21.08 and 17.03, respectively. However, it is not significantly different from Devatra 2 and Dering varieties. Differences in leaf area of soybean varieties might have influenced by physiological factors of soybean leaves of four varieties. Such things might have resulted from decreased in leaf surface area and leaf curl occurred due to the shrinkage process of bulliform cells which functions to protect the underlying tissue from being damaged due to greater water loss. According to Srivastava (2002), leaf rolling is a mechanism of plant adaptation to drought stress due to the shrinkage process of bulliform cells which serves to protect leaf tissue due to loss of water in large quantities, resulting in a small leaf surface area. According to Sinay (2015) plants experiencing drought stress would have leaf growth inhibition by reducing leaf surface area and reducing the number of stomata to prevent evaporation.

Soybean varieties had a significant effect on stomata number, opened stomata and stomatal density. Devatra2 had significantly affected with Devatra1, Dering and Grobongan varieties. Based on changes in the physiology of soybean varieties facing drought stress, Devatra 2 and Grobongan varieties have good adaptability or drought tolerance in coastal areas compared to Devatra1 and Dering varieties as indicated by the observed variables, *i.e.*, the number of stomata, the number of open stomata and the density of stomata. Based on the varietal description, Devatra1 and Dering varieties have acid-dry resistance. However, the study experienced drought stress conditions due to land characteristic of sandy land. The number of opened stomata indicated the plant's effort to survive drought stress conditions. This is in accordance with Taiz and Zeiger (2006) that plants withstand drought stress by closing stomata to reduce transpiration. Purwanto and Agustono (2010) also concluded that the reduction in density and the number of stomata openings are some of the

tolerance mechanisms of plants against stress to reduce water loss in large quantities.

Results indicated that FC had significant effects on the variable leaf area and leaf area index, where 100% FC was significantly different from 50% and 25% FC, but not significantly different from 75% FC (Table 5). It appeared that leaf area of soybean at 100% FC was higher than those of 50% and 25% FC. These results are in line with Chun *et al.* (2021) who concluded soybeans required soil moisture content of more than 70% FC. Similar responses were obtained in high leaf area and LAI at 75% FC, which were not significantly different from those of grown in 100% FC. However, the number of stomata, number of opened stomata and stomatal density at 50% and 25% FC were not significantly different from 75% and 100% FC. These results were in line with Sulistyono *et al.* (2011), who concluded the decrease in leaf area and size are considered as the main physiological process influenced by drought stress, which eventually causes a decrease in stomatal conductivity.

It can be concluded that the morphology performances of soybean in dry conditions on sandy soil is influenced by the varieties. Meanwhile, the physiological changes of the four varieties showed non significantly differed in each of the observed variables. Devatra 2 and Grobogan varieties were more tolerant than Devatra1 and Dering varieties to drought in sandy soil as indicated by stomata variables. Morphologically effects of drought stress of soybean as measured by FC took place when soybean grown at 50% FC and lower. Physiological responses of soybean grown under different in FC affected the LAI of soybean due to changes in stomata density.

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