

Tolerance Some Soybean Cultivars to Stress Drought at Vegetative to Generative Phase

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Abstract: Soybean is one of crop which many conducting in Jambi province, where this area is one of sentra soybean producer in Indonesia. Main constraint in expansion of soybean in Jambi province is ability of adaptation of low crop to condition of area that is partly are consisted of area of marjinal with level of low fertility and has water supply internal issue. Plant breeding, especially repair of genetic quality applies strains or cultivar indication by drought tolerant to earn is one of alternative of trouble-shooting. Examination of soybean cultivar tolerance to stres drought at vegetative to generative phase (15 - 45 Day After Plants/DAP) done in glasshouse by using Split Plout Design. Treatment of stres drought as main check and soybean cultivar as child of check. From result of observation, treatment of stres dryness at vegetative phase and generative can reduce and pursues growth and result of soybean cultivar tested. But for soybean cultivar Derap-1, Dena-2, Deja-2, Dering-1, Grobogan, Detam-1 and Dega-1, treatment of stres drought exactly causes improvement of root length to range from 7,14 - 60,57%. Based on variable dry weigh seed, there are seven soybean cultivars tested able to be classified in group of tolerant and tolerant medium to stres drought at vegetative to generative phase that is var. Argo Mulyo, Dena-1, Derap-1, Dena-2, Deja-2, Dering-1, Grobogan dan Detam-1. Stres drought causes improvement can of leaf proline content at some soybean cultivars tested with different improvement pattern. Therefore mechanism of soybean tolerance to stres drought can through improvement of root length as mechanism of tolerance in morphology and or through improvement of proline content as mechanism of tolerance physiologically.

Keywords: Soybean Cultivar; Stres drought; Vegetative to Generative phase; Content proline.

Introduction

Soybean (*Glycine max* (L) Merrill) strategic food commodity can be one of the main sources of vegetable protein with high nutritional value. Soybean seeds contain 42-45% protein. In the last 5 years, domestic soybean production has tended to decline from year to year, where in 2015 soybean production was at 0.96 million tons and tended to decrease to only 0.32 million tons in 2020 (Figure 1).

Domestic soybeans are still the center of attention today, because their production is still unable to meet the demand for soybeans in Indonesia. Indonesia's soybean production on average can only meet approximately 24 percent of the total national soybean

demand. The average demand for Indonesian soybeans is currently around 2.8 million tons per year.



Figure 1. Indonesian soybean production 2015-2020 (million tons) Source: Kementerian Pertanian, 2021

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In Jambi Province, soybean plants planted on PMK soil are easily stressed by water shortages, especially during the dry season. This is because the soil has properties that are not able to hold water properly and is exacerbated by the condition of the soybean root type which is shallow, so that it is easy to experience stress from lack of water so that it affects plant growth and production.

Most plant species such as soybean (*Glycine max*), during vegetative and reproductive growth are very sensitive to limited water conditions. Soybean is a dehydration-sensitive species that requires an optimum amount of water for seed germination, seedling growth and plant development (Chen et al, 2006; Patriyawaty, 2020). In soybean plants, stress from lack of water will inhibit the uptake of water by the roots. The inability of plant roots to absorb water to compensate for water loss by transpiration can cause plants to wilt. In general, plants will close their stomata to reduce water loss. Closed stomata can help plants to avoid water shortages quickly. However, the closed stomata pores also inhibit the absorption of carbon dioxide and oxygen from the air by the internal tissues of the plant. This condition will actually stop the flow of water through the plants so that it can also reduce nutrient absorption. All the factors described above are the causes of soybean plants reducing their metabolism in order to sustain their life during times of water shortage (drought) (Borges and Pinto, 2008; Manavalan et al, 2009). In soybean, drought stress during the vegetative phase can reduce plant height, number of nodes, root length, root dry weight and crown (Pratiwi et al, 2019).

Genotypes tolerant to water stress have lower transpiration, higher photosynthesis, use water more efficiently and are able to produce higher pod yields than genotypes sensitive to water stress. In peanut plants, the tolerance of the Singa genotype is better, because this genotype has a lower transpiration rate, higher photosynthesis and is more efficient in water use, which is due to differences in root spread, canopy closure and leaf development (Singh. G, 2010). According to Hapsah, et al. 2006, soybean plants have the most important physiological mechanism in adapting to water stress, namely by maintaining turgor through a decrease in osmotic potential and accumulation of dissolved compounds, namely proline and an increase in ABA and IAA content.

There are various approaches to tolerant selection of environmental stresses, and they can be grouped into two, namely (i) direct (empirical) selection and (ii) indirect (physiological) selection. Direct selection implies selection for absolute performance (growth rate and yield) under actual stress conditions or selection for plants that experience only a slight decrease in growth/yield under environmental stress conditions. Indirect selection implies screening for morphological or

physiological characteristics that may correlate with resistance to a particular stress.

The aim of this experiment was to determine the effect of drought stress that occurred since the vegetative and generative phases on soybean growth and yield, as well as evaluate tolerance, leaf proline content of fourteen soybean cultivars under optimal environmental conditions (without stress) and under stress conditions (drought stress) in the vegetative and generative phases.

Method

Evaluation of the response of high yielding soybean cultivars and those indicated to be drought tolerant to drought stress under controlled conditions. The stages of the experiment to be carried out include:

Preparation of pots for planting in the greenhouse

For testing in the greenhouse, the seeds were planted in one-liter plastic pots containing a mixture of soil, sand, manure in the ratio of 2: 1: 1. The planting medium was sterilized by pouring formalin solution (30%), wrapped in airtight plastic, and incubated for 14 days. After 14 days of incubation. The media is put into polybags with a size of 35 x 40 cm as much as + 8 Kg per polybag. Each polybag was given 0.4 gram of urea fertilizer; SP36 0.8 grams and 0.8 grams KCL and doused with 1000 ml of water or until saturated. After these various treatments, the polybags with planting media were ready to be planted with the tested soybean seeds.

Research design

This study used a Split Plot Design in Completely Randomized Design (CRD) which consisted of 2 factors, namely the main factor of drought stress (C) and subplots of variety (V). The main factor is the treatment of plant drought stress in the vegetative to generative phases (from 15 to 45 days after planting) which consists of 2 levels, namely: C0 = without drought stress and C1 = drought stress in the vegetative to generative phases (15 - 45 DAP) Subplots are a Variety (V), which consists of 14 levels namely:

V1 (Anjasmoro)	V8 (Defon-1)
V2 (Agromulyo)	V9 (Detap-1)
V3 (Dena-1)	V10 (Dering-1)
V4 (Derap-1)	V11 (Grobogan)
V5 (Dena-2)	V12 (Gepak kuning)
V6 (Deja-1)	V13 (Detam-1)
V7 (Deja-2)	V14 (Dega-1)

Each treatment level was replicated 2 times, resulting in $2 \times 14 \times 2 = 56$ experimental units consisting of 6 plants per experimental unit, resulting in 168 experimental units for the main factor without drought stress, 168 experimental units for the main factor of drought stress, so that the total In total, there were 336

plants and 3 plants as samples for each experimental plot.

Planting seeds in a greenhouse

For each variety of soybean plant, a minimum of five seeds were planted for each reduction of water treatment with one seed per pot (a total of 10 seeds for each genotype of soybean plant) and repeated 3 times. The plant pots are arranged with the distance between the pots used being 0.2 m in the row and 0.4 m between the rows. The pots in which the tested seeds were grown were grown in a greenhouse.

For genotypic treatment of soybean plants, they were placed in subplots. As a control, genotypes identified as sensitive to drought stress were planted. Control plants were planted in rows between genotypes of drought tolerant soybean plants with a ratio of four rows of drought tolerant plants and one row of control plants. Control plants were grown in sufficient quantities to anticipate various experimental errors.

The drought stress treatment was placed as the main plot with the treatment of reducing the application of water, namely the plants were watered up to field capacity and left without watering until the plants began to show symptoms of wilting. Once wilting symptoms are visible, the plants are watered again the next morning to field capacity. The period of reducing water supply which causes the plants to start to wilt usually ranges from 4-7 days after the start of watering. Field capacity is determined by pouring water on the growing medium until it is saturated and dripping from the aeration hole at the bottom of the pot.

Observations were made on the age of flowering, plant height at flowering and at harvest, root length and PC, total number and PC of pods, number and PC of filled pods, and number and PC of seeds. Except for the age of flowering and plant height at the time of flowering, observations were made after harvest.

Analysis of Leaf Proline Content

The physiological response of the tested plants to drought stress was observed by analyzing the proline content of the leaves. Proline levels were analyzed based on the method of Abraham et al, (2010). The second leaf from the shoot is harvested and dried in an incubator filled with silica gel. The dried leaves (0.2 g) were crushed and homogenized with 10 ml of sulfosalicylic acid (3%). After centrifugation at 5000 rpm for 15 minutes, 2 ml of the supernatant obtained was reacted with 2 ml of ninhydrin acid solution (1.25 g ninhydrin, 30 ml of glacial acetic acid, and 20 ml of 6 M H₃PO₄) and 2 ml of glacial acetic acid and heated over a bath. water to 100°C for 60 minutes. The reaction was terminated by cooling the solution in ice water for 5 minutes. The reaction product was extracted with 4 ml of toluene to form chromoform and the absorbance of the

chromoform was measured with a spectrophotometer at a wavelength of 520 nm. DL-Proline (Sigma) dissolved in sulfosalicylic acid (3%) was used as standard.

Sensitivity index to drought stress

The stress sensitivity index (S) is calculated using the formula developed by Dababat et al, (2016), namely: $S = (1 - [Y/Y_p]) / (1 - [X/X_p])$; Y and Y_p are the average observed values for a particular genotype under drought stress and non-stress conditions, while X and X_p are the observed mean values for all genotypes under drought stress and non-stress conditions. The S value was calculated using the seed dry weight variable. Based on the S value obtained, the soybean plants tested were categorized as tolerant if $S < 0.5$, medium tolerant if $0.5 < S < 1$, and sensitive if $S > 1$.

Result and Discussion

Result

Effect of drought stress treatment in the vegetative to generative phases on soybean growth and yield

From the results of observations on the growth and yield variables of soybean plants, it was found that drought stress treatment in the vegetative to generative phase significantly reduced the growth component variables (plant height, root length, flowering age and harvest age) and yield components (total number of pods), total pod dry weight, number of filled pods, filled pods, seed BK and 100 seed BK) on the tested soybean varieties (Tables 1 and 2). Particularly for the variables of flowering age and harvesting age, the effect of drought stress actually shortens flowering age and harvesting age. Meanwhile, root length variables, and drought stress treatment in the vegetative to generative phase (from 15 days after planting (MST) to 45 WAP) were not significantly different from conditions without stress (optimum) or drought stress treatment in the vegetative to generative phases. effect on root length variables in the tested soybean cultivars (Table 1).

From Table 2, it can be seen that stress treatment in the vegetative to generative phase (from 15 to 45 HST) can cause a decrease in the growth and yield of soybean plants, it is suspected that stress treatment in the vegetative to generative phase is classified as severe stress and has a greater impact on physiological processes of plants, especially in the process of photosynthesis. Conditions of severe stress with a very minimal level of water availability can disrupt plant metabolic processes which really need sufficient water availability for their growth so it mainly affects the process of forming flowers, pods, and the period of filling the pods or forming seeds. The drought stress treatment period can be seen in Table 3.

Table 1. Effect of drought stress on plant growth component variables of several soybean varieties tested

Treatment	Plant Height (cm)	Root Length (cm)	Flowering Age (days)	Harvest Age (days)
No drought stress	96.3 a	44.4 a	36.7 a	90.1 a
Drought stress from vegetative to generative phases	88.2 b	42.1 a	34.9 b	86.9 b

Note: The mean data in columns with the same lowercase letters are not significantly different based on Duncan's multiple range test at $\alpha=0.05$.

Table 2. The effect of drought stress on the variable components of plant yields for several soybean varieties tested

Perlakuan	Jumlah Polong Total	BK Polong Total (gr)	Jumlah Polong Isi	BK Polong Isi (gr)	BK Biji (gr)	BK 100 biji (gr)
Tanpa stres kekeringan	92.2 a	30.5 a	90.7 a	30.5 a	22.4 a	13.5 a
Stres kekeringan fase vegetatif sampai generatif	62.9 b	17.0 b	59.7 b	16.8 b	11.8 b	11.6 b

Note: The mean data in columns with the same lowercase letters are not significantly different based on Duncan's multiple range test at $\alpha=0.05$.

Table 3. Drought stress treatment period of several soybean varieties in the vegetative and generative phases in the greenhouse

Stress Period *)	Length of stress from vegetative to generative phases (days)	Information
I	6	Beginning of the
II	8	stress period in
III	7	the vegetative to
IV	5	generative phase
V	4	starting at 15 HST
VI	3	until 45 HST

Note: *) one period of stress is calculated starting from the time the plants were treated with drought stress (no watering) until

the plants showed wilting symptoms of 75% of the population of the tested soybean cultivars/varieties.

Response of several soybean cultivars tested to drought stress treatment in the vegetative and generative phases

Treatment of drought stress in the vegetative and generative phases significantly reduced the growth and yield of all tested soybean varieties by showing different responses, both in the growth component variables and the yield component variables. From the results of further tests, it was found that each tested soybean variety showed a different response pattern (Tables 4 to 12).

Table 4. Response of several soybean varieties on growth component variables of plant height to drought stress treatment from vegetative to generative phases

Soybean Varieties/Cultivars	Average Plant Height (cm)			Percentage of decline	
	No Stress	Vegetative to Generative phase stress			
Anjasmoro	99.00	a	98.33	a	-0.68
Argo Mulyo	104.11	a	88.22	b	-15.26
Dena-1	89.67	a	85.83	b	-4.28
Derap-1	87.33	a	80.33	b	-8.02
Dena-2	85.44	a	73.78	b	-13.65
Deja-1	103.67	a	88.67	b	-14.47
Deja-2	99.00	a	91.67	b	-7.40
Defon-1	84.63	a	75.33	b	-10.99
Detap-1	87.89	b	90.11	a	2.53
Dering-1	107.33	a	96.67	b	-9.93
Grobogan	100.89	a	95.78	b	-5.06
Gepak kuning	102.33	a	84.78	b	-17.15
Detam-1	95.78	a	93.67	b	-2.20
Dega-1	99.89	a	90.89	b	-9.01

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Based on the growth component variables, stress treatment in the vegetative to generative phase significantly reduced plant height in all tested soybean varieties with a reduction rate ranging from 2.20 to 17.15% compared to no stress except for Anjasmoro and

Detap-1 varieties (Table 4). For the age of flowering variable, stress treatment in the vegetative to generative phase significantly accelerated the flowering time for all soybean varieties tested in Table 5. However, for the harvest age variable, stress treatment in the vegetative to

generative phase also significantly accelerated the harvest time compared to no stress, except for varieties Dena-1 and Deja-2 (Table 6).

Drought stress treatment in the vegetative to generative phases generally reduced root length in almost all tested soybean varieties. However, for several soybean varieties such as cultivars Derap-1, Dena-2,

Deja-2, Dering-1, Grobogan, Detam-1 and Dega-1, drought stress treatment in the vegetative to generative phases actually caused an increase in root length ranging from 7.14 – 60.57%. The highest increase in root length due to drought stress in the vegetative phase was owned by the Detam-1 variety, which was 60.57%, followed by the Dena-2 cultivar, which was 44.94% (Table 7).

Table 5. Response of several soybean varieties to the growth component variables of flowering age against drought stress treatments from the vegetative to generative phases.

Soybean Varieties/Cultivars	Average Age of Flowering (days)				
	No Stress	Vegetative to Generative phase stress		Percentage of decline	
Anjasmoro	33.00	a	28.78	b	-12.79
Argo Mulyo	31.11	a	29.22	b	-6.08
Dena-1	40.33	a	28.67	b	-28.91
Derap-1	31.00	a	28.56	b	-7.87
Dena-2	35.89	a	28.22	b	-21.37
Deja-1	40.89	a	31.67	b	-22.55
Deja-2	39.78	a	33.44	b	-15.94
Defon-1	45.89	a	29.96	b	-34.71
Detap-1	38.67	a	28.22	b	-27.02
Dering-1	36.11	a	28.11	b	-22.15
Grobogan	32.44	a	28.00	b	-13.69
Gepak kuning	39.00	a	30.00	b	-23.08
Detam-1	35.67	a	28.89	b	-19.01
Dega-1	35.56	a	32.22	b	-9.39

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Table 6. Response of several soybean varieties to the growth component variables of harvesting age against drought stress treatment from the vegetative to generative phases

Soybean Varieties/Cultivars	Average Age of Harvest (days)				
	No Stress	Vegetative to Generative phase stress		Percentage of decline	
Anjasmoro	91.33	a	81.56	b	-10.70
Argo Mulyo	86.33	a	80.78	b	-6.43
Dena-1	83.33	a	83.33	a	0.00
Derap-1	87.44	a	83.33	b	-4.70
Dena-2	91.67	a	80.00	b	-12.73
Deja-1	82.33	a	80.67	b	-2.02
Deja-2	85.67	a	85.67	a	0.00
Defon-1	86.19	a	81.67	b	-5.24
Detap-1	88.22	a	80.00	b	-9.32
Dering-1	85.11	a	80.00	b	-6.00
Grobogan	83.67	a	80.33	b	-3.99
Gepak kuning	91.22	a	86.00	b	-5.72
Detam-1	82.00	a	80.00	b	-2.44
Dega-1	92.00	a	84.00	b	-8.70

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Based on the yield component variables, stress treatment in the vegetative to generative phase significantly reduced the number of filled pods compared to no stress on all soybean varieties tested

with reduction rates ranging from 15.5 – 82.4% compared to no stress, except for Agro Mulyo, Dena-1, Dena-2 and Deja-2 (Table 8).

Table 7. Response of several soybean varieties on the Root Length variable to drought stress treatment from the vegetative to generative phases

Soybean Varieties/Cultivars	Average Root Length (cm)				
	No Stress	Vegetative to Generative phase stress		Percentage decrease or increase	
Anjasmoro	44.89	a	35.67	b	-20.54
Argo Mulyo	44.78	a	39.89	b	-10.92
Dena-1	45.56	a	45.13	a	-0.94
Derap-1	39.00	b	47.78	a	22.51
Dena-2	37.56	b	54.44	a	44.94
Deja-1	46.33	a	31.11	b	-32.85
Deja-2	43.56	b	46.67	a	7.14
Defon-1	45.17	a	41.89	b	-7.26
Detap-1	43.22	a	42.56	a	-1.53
Dering-1	47.67	b	51.44	a	7.91
Grobogan	41.78	b	47.11	a	12.76
Gepak kuning	39.22	a	40.22	a	2.55
Detam-1	34.67	b	55.67	a	60.57
Dega-1	36.22	b	42.00	a	15.96

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Table 8. Response of several soybean varieties to yield component variables Number of filled pods against drought stress treatment from vegetative to generative phases.

Soybean Varieties/Cultivars	Average Number of Filled Pods				
	No Stress	Vegetative to Generative phase stress		Percentage decrease or increase	
Anjasmoro	64.67	a	38.33	b	-40.7
Argo Mulyo	7.33	b	14.22	a	94.0
Dena-1	118.11	a	118.89	a	0.7
Derap-1	48.00	a	32.89	b	-31.5
Dena-2	46.22	a	42.56	a	-7.9
Deja-1	104.67	a	78.67	b	-24.8
Deja-2	102.67	a	102.78	a	0.1
Defon-1	132.84	a	66.00	b	-50.3
Detap-1	89.67	a	57.11	b	-36.3
Dering-1	58.56	a	49.11	b	-16.1
Grobogan	52.67	a	33.67	b	-36.1
Gepak kuning	182.56	a	59.00	b	-67.7
Detam-1	139.11	a	97.11	b	-30.2
Dega-1	127.56	a	52.78	b	-58.6

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Table 9. Response of several soybean varieties to the yield components of filled pod pods against drought stress treatments from the vegetative to generative phases

Soybean Varieties/Cultivars	Average BK of filled pods (gr)				
	No Stress	Vegetative to Generative phase stress		Percentage of decrease/increase	
Anjasmoro	34.43	a	16.09	b	-53.3
Argo Mulyo	3.28	b	7.26	a	121.3
Dena-1	35.44	a	28.71	b	-19.0
Derap-1	22.77	a	14.73	b	-35.3
Dena-2	15.48	a	14.22	a	-8.1
Deja-1	29.12	a	14.22	b	-51.2
Deja-2	27.13	a	24.00	b	-11.5
Defon-1	44.40	a	21.18	b	-52.3
Detap-1	36.56	a	17.02	b	-53.4
Dering-1	18.93	a	14.41	b	-23.9
Grobogan	17.00	a	11.28	b	-33.6
Gepak kuning	54.02	a	11.76	b	-78.2
Detam-1	40.54	a	26.03	b	-35.8
Dega-1	49.19	a	16.01	b	-67.5

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Stress treatment in the generative phase reduced the BK of Filled Pods and Seeds compared to no stress on all tested soybean varieties with reduction rates ranging from 19.0 - 78.2%, except for the Agro Mulyo variety, Dena-1 (Tables 9 and 10). Specifically, for the

Argomulyo and Dena-2 varieties, stress treatment in the vegetative to generative phases had no significant effect on reducing yield components (Number of filled pods, filled pods and seeds) (Tables 8, 9, 10).

Table 10. Responses of several soybean varieties to the variable components of yield BK Seeds to treatment of drought stress from the vegetative to generative phases.

Soybean Varieties/Cultivars	Average BK Seeds (gr)				
	No Stress		Vegetative to Generative phase stress		Percentage of decrease/increase
Anjasmoro	24.94	a	11.32	b	-54.6
Argo Mulyo	2.37	b	5.10	a	115.2
Dena-1	26.74	a	20.54	b	-23.2
Derap-1	16.67	a	10.92	b	-34.5
Dena-2	11.34	a	10.32	a	-9.0
Deja-1	21.12	a	9.49	b	-55.1
Deja-2	19.87	a	16.90	b	-14.9
Defon-1	32.38	a	14.73	b	-54.5
Detap-1	26.32	a	11.69	b	-55.6
Dering-1	13.14	a	10.04	b	-23.6
Grobogan	12.28	a	7.89	b	-35.7
Gepak kuning	39.16	a	7.96	b	-79.7
Detam-1	30.01	a	18.23	b	-39.3
Dega-1	37.80	a	11.26	b	-70.2

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

In the 100-seed DM variable, drought stress treatment in the vegetative to generative phase significantly reduced seed size in almost all varieties

tested, except for cultivars Dena-2, Defon-1, Dering-1, Grobogan and Detam-1, the drought stress treatment did not reduce seed size (Table 11).

Table 11. Responses of several soybean cultivars/varieties on the variable component yield of 100 seed DM to drought stress treatment from the vegetative to generative phases

Soybean Varieties/Cultivars	Average BK 100 seeds (gr)				
	No Stress		Vegetative phase stress		Percentage of decline
Anjasmoro	18.80	a	13.87	b	-26.22
Argo Mulyo	21.67	a	18.27	b	-15.69
Dena-1	11.50	a	9.64	b	-16.17
Derap-1	17.50	a	15.90	b	-9.14
Dena-2	13.53	a	13.50	a	-0.22
Deja-1	9.53	a	6.23	b	-34.63
Deja-2	9.60	a	7.90	b	-17.71
Defon-1	14.95	a	13.90	a	-7.02
Detap-1	14.23	a	11.57	b	-18.69
Dering-1	11.37	a	10.97	a	-3.52
Grobogan	12.03	a	11.87	a	-1.33
Gepak kuning	9.50	a	8.00	b	-15.79
Detam-1	10.83	a	10.20	a	-5.82
Dega-1	13.50	a	10.13	b	-24.96

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Effect of Drought Stress on Leaf Proline Content.

Under stress-free conditions at 47 HST, stress treatment in the vegetative to generative phases markedly increased leaf proline content compared to no

stress in all soybean varieties tested with levels of increase ranging from 158.60 to 596.73%, except Dena-1, Dena-2, Defon-1 and yellow gepak. A very high increase in leaf proline content occurred in the Agromulyo

variety with a proline content of 4101 ug proline/g leaf (Table 13).

Table 13. Analysis of Proline Content of Drought Stress in the Vegetative to Generative Phases at the age of 47 HST

Soybean Varieties/Cultivars	ug proline/g leaf		Percentage Increase or decrease
	No Stress	Vegetative to Generative phase stress	
Anjasmoro	1433 a	3705 b	158.60
Argo Mulyo	721 a	4101 b	468.59
Dena-1	672 a	655 a	-2.49
Derap-1	1052 a	3727 b	254.32
Dena-2	3459 a	4149 a	19.95
Deja-1	1941 a	4149 b	113.79
Deja-2	1043 a	2425 b	132.54
Defon-1	784 a	897 a	14.43
Detap-1	548 a	3820 b	596.73
Dering-1	856 a	2484 b	190.24
Grobogan	1215 a	3977 b	227.30
Gepak kuning	1020 a	885 a	-13.22
Detam-1	1107 a	3977 b	259.15
Dega-1	1591 a	4158 b	161.40

Note: The mean data in the same row with the same letter are not significantly different based on Duncan's multiple range test at $\alpha = 0.05$.

Effect of drought stress in the vegetative to generative phases on the sensitivity index (S) of the tested soybean cultivars.

The index of sensitivity to drought stress in the vegetative to generative (S) phases from the number and BK of filled pods and BK of soybean seeds indicated that only eight varieties of the 14 tested soybean cultivars had levels of tolerance and medium tolerance to drought stress from the vegetative to generative phases. Plant response to drought stress was estimated using the S value based on the number and BK of pod content per plant and could not classify the tested cultivars into groups that were consistent with stress from the

vegetative to generative phases (Table 14). On the other hand, the S value calculated based on the seed BK variable was able to classify the 8 (eight) tested soybean varieties into tolerant and medium tolerant groups in both the vegetative and generative phases, while the other six varieties had a tolerance level that was classified as sensitive to drought stress (Table 14). Eight soybean varieties classified as tolerant and medium tolerant to stress in the vegetative and generative phases, namely varieties Argo Mulyo, Dena-1, Derap-1, Dena-2, Deja-2, Dering-1, Grobogan and Detam-1.

Table 14. Sensitivity index to stress (S) calculated based on the variables number and dry weight (BK) of filled pods and BK of seeds harvested and the tolerance of 14 soybean varieties to drought stress in the vegetative to generative phases.

Soybean Varieties/Cultivars	Sensitivity index according to yield component variables and drought stress treatment		
	Number of pods Fill	BK Filled pods	BK Seeds
Anjasmoro	1.34 (P)*	1.41 (P)	1.35 (P)
Argo Mulyo	-0.95 (T)	-0.68 (T)	-0.60 (T)
Dena-1	-0.01 (T)	0.29 (T)	0.34 (T)
Derap-1	0.90 (M)	0.68 (M)	0.59 (M)
Dena-2	0.17 (T)	0.11 (T)	0.11 (T)
Deja-1	0.65 (M)	1.30 (P)	1.38 (P)
Deja-2	0.00 (T)	0.16 (T)	0.20 (T)
Defon-1	1.98 (P)	1.36 (P)	1.35 (P)
Detap-1	1.11 (P)	1.42 (P)	1.41 (P)
Dering-1	0.38 (T)	0.39 (T)	0.35 (T)
Grobogan	1.10 (P)	0.63 (M)	0.63 (M)
Gepak kuning	4.09 (P)	4.45 (P)	4.42 (P)
Detam-1	0.85 (M)	0.69 (M)	0.73 (M)
Dega-1	2.77 (P)	2.57 (P)	2.66 (P)

Description: *) x(y) - Sensitivity index value to drought stress (S) and tolerance grouping of the tested soybeans into tolerant (T) if $S < 0.5$, medium tolerant (M) if $0.5 < S < 1$, or sensitive (P) if $S > 1$.

Discussion

In this experiment, drought stress was carried out by reducing the application of water in the vegetative to generative growth phase (15–45 DAP). During this growth period, the plant groups that were given the stress treatment were watered once every 3 – 8 days, while the control plant group was watered every day. After 3-8 days without watering, the plants begin to show symptoms of wilting. Although the plants spring back fresh after being watered, the periods of reduced water provided have led to drought stress.

The effect of drought stress on the vegetative to generative growth phases reduced plant height at harvest and root length of soybean plants compared to the no-stress treatment. Although plant height, root length, flowering and harvest time decreased compared to plants without stress. However, for several cultivars Derap-1, Dena-2, Deja-2, Dering-1, Grobogan, Detam-1 and Dega-1, drought stress treatment in the vegetative phase actually caused an increase in root length ranging from 7.14 – 60.57 %. Var. Detam-1 is 60.57% and followed by var. Dena-2 of 44.94% (Table 10). This is presumably because under stress conditions, soybean plants use the photosynthate produced to maintain root growth, causing a decrease in soybean plant height growth but does not affect root development (Yanli Du, 2020). Decreasing plant height, flowering and harvesting ages to reduce transpiration rates and increasing root growth to increase water absorption is one of the existing mechanisms in plants to deal with drought stress (Saxena. 2002) and can be used as an indicator of the nature of plants that are tolerant of drought stress.

After the age of 47 DAP (after the stress of the vegetative to generative phase) all plants are given an adequate amount of water every day. However, the negative impact of drought stress that occurs in the vegetative to generative phases still affects the yield of harvested soybeans. Soybean plants that experienced drought stress in the vegetative and generative growth phases produced lower soybean pods and seeds than those without stress.

The age period of 15 – 45 DAP is reported to be a period of rapid growth of soybean plants, which requires the availability of sufficient amounts of water. The reduced availability of water during this period has disrupted the growth of soybean plants, which is a growth period in which the formation and filling of soybean pods requires the availability of sufficient water (Anne et al, 2018). The reduced availability of water during this period may have disrupted the formation and filling of pods as shown in this experiment. Drought stress was also reported to inhibit photosynthetic activity and photosynthate translocation thereby reducing harvested yields (Riduan et al, 2005). In

soybean, drought stress causes the loss of flowers and pods and reduces seed yield (Dogan et al, 2007).

Drought stress in the vegetative phase also caused a decrease in the vegetative growth of soybean plants, except that root length was not affected by drought stress for some of the cultivars tested. Previous research revealed that increasing root volume and length is one of the mechanisms by which plants can cope with drought stress (Kitbanmroong and Chatachume, 1993).

Based on the variable GC of the seeds, there were seven soybean cultivars tested that could be classified into groups tolerant and medium tolerant to stress in the vegetative to generative phases, namely var. Argo Mulyo, Dena-1, Derap-1, Dena-2, Deja-2, Dering-1, Grobogan and Detam-1. In plants experiencing stress from the vegetative to generative phases aged 15 - 45 DAP, plant growth and development had a significant effect on increasing leaf proline content in almost all tested soybean cultivars, except for Var. Dena-1, Dena-2, Defon-1 and Gepak kuning. This can be ascertained as the form of gene expression that codes for proline formation is influenced by the induction of drought stress treatment as part of the mechanism of plant tolerance to drought stress. However, specifically for var Dena-1, an increase in proline accumulation has occurred, without having to be induced by drought stress treatment.

Plants that are tolerant and medium tolerant to drought stress have been reported to be more able to increase leaf proline content as a response to drought stress compared to plants that are sensitive (Kavi Kishor et al, 2005). A positive correlation between leaf proline content and plant ability to adapt to drought stress and high salt stress has also been reported (Ozturk et al, 2021; Sacita et al, 2018).

However, under drought stress conditions in the vegetative and generative growth phases, soybean plants that are classified as tolerant and medium tolerant have different patterns of increasing proline content as a form of tolerance mechanism to overcome drought stress.

1. Var. Argo Mulyo, Derap-1, Deja-2, Dering-1, Grobogan and Detam-1, there was a significant increase in proline content ranging from 132.54 to 468.59% in drought stress conditions of the vegetative to generative phases at 47 DAP.
2. Var. Dena-1 did not increase the proline content in drought stress conditions from the vegetative to generative phases at 47 DAP.
3. Var. Dena-2 increases in proline content have actually occurred in the conditions before and after drought stress from the vegetative to generative phases at the age of (3459 – 4149 ug proline/g leaf).

Thus, there are two mechanisms of tolerance of soybean plants to drought stress, namely through

increasing root length as a morphological tolerance mechanism and through increasing proline content as a physiological tolerance mechanism (Buezo et al, 2019; Dong et al, 2019). For var. Derap-1, Dena-2, Deja-2, Dering-1, Grobogan and Detam-1 have two mechanisms of tolerance to drought stress by increasing root length and proline content, while var. Argo Mulyo only has one tolerance mechanism, namely through an increase in proline content. Therefore, for further research on marginal land conditions (FMD soil) with mycorrhizal treatment, 8 soybean varieties were used (var. Argo Mulyo, Dena-1, Derap-1, Dena-2, Deja-2, Dering-1, Grobogan and Detam-1) which are classified as tolerant and medium tolerant to drought stress.

Conclusion

From the results of the observations and carried out above, it can be concluded that drought stress treatment in the vegetative to generative phases (15 - 45 HST) significantly reduced the growth and yield components of the soybean plants tested. For several soybean cultivars, namely var. Derap-1, Dena-2, Deja-2, Dering-1, Grobogan, Detam-1, and Dega-1, treatment of drought stress in the vegetative to generative phases actually caused an increase in root length ranging from 7.14 - 60.57%. Based on the variable BK of the seeds, there were seven soybean cultivars tested that could be classified into groups tolerant and medium stress-tolerant to the generative to vegetative phases, namely var. Argo Mulyo, Dena-1, Derap-1, Dena-2, Deja-2, Dering-1, Grobogan and Detam-1. Under drought stress conditions in the vegetative to generative growth phases, soybean plants that were classified as tolerant and medium tolerant had different patterns of increasing proline content. There are two mechanisms of tolerance of soybean plants to drought stress, namely through increasing root length as a morphological tolerance mechanism and through increasing proline content as a physiological tolerance mechanism.

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