

**Research Article**

**Morphological characteristic of purple long yard bean cultivars and their tolerance to drought stress**

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**Abstract:** The cultivation of purple long yard bean which tolerance to drought stress and have high productivity can improve farming in arid area. The purpose of this study was to evaluate the mechanism of the tolerance purple long yard beans to drought stress based on morphologic characters, to get the hypothesis method of tolerance and to obtain tolerance cultivars to the drought stress. Eight cultivars of purple long yard beans, i.e. UBPHU1-41, UBPHU1-130, UBPU3-153, UBPU1-202, UBPU2-222, UBPU1-365, Brawijaya 4 and Bagong 2, were tested in two environmental conditions, 100% field capacity and 50% field capacity. The results showed that drought stress in purple long yard bean affected all morphological characters observed, except for root length and flowering time. Estimation of tolerance to drought stress using the Principles Component Analysis (PCA) showed that the shoot fresh weight could be an indicator of purple pod bean tolerance to drought stress. However, the test using Stress Susceptibility Index (SSI) was not able to classify the purple long yard bean tolerance to drought stress. The results of analysis using PCA followed by discriminant analysis and clustering dendrogram showed that the UBPU1-41, UBPU1-130, UBPU2-222, UBPU1-365, UB4 and Bagong 2 cultivars were medium cultivars that are tolerant to drought stress. Therefore, they can be planted in semiarid regions.

**Keywords:** *drought stress, morphological characters, purple long yard bean*

**Introduction**

Indonesia has arid area about 148 million ha (78%) and wet lands (wet lands) area with 40.20 million ha (22%) from 188.20 million ha of the total land area. Of the total 148 million ha of dry land, only 76.22 million ha (52%) is suitable for agricultural cultivation (Rachman and Sutono, 2005). Water scarcity in arid and semi-arid regions has become a major world's concern (Ghamarnia and Sepehri, 2010; Shen et al., 2011).

The cultivation of purple long yard bean cultivars that are tolerant to drought stress and that produce high yield offer chance to develop the cultivation of the beans on dry land. According to Hidayat et al. (2000), dry lands have a strategic value in order to support the development of sustainable agriculture, particularly to achieve national food security and endurance. However, the yield per unit area of some agricultural crops decreased significantly

around 50-80% due to drought stress (Bray, 2002). Generally, green, light green or green and white long yard beans, all have their advantages. However, there are types of purple long yard beans that are not known by many people. The aphid pests do not favour the advantages of purple long yard beans because their skin pods, leaves and stems are covered with feathers along the surface (Hardinaningsih, 2012). Protein, fiber, and anthocyanin contents in the beans are very beneficial substances to the plant to live longer, to have longer production period and yard longer shelf life, as well as more resistant to drought conditions compared with beans in general.

To survive in drought stress, plants have adaptation characters such as mechanism of escape, avoidance, tolerance, and recovery (Levitt, 1980; Fukai and Cooper, 1995). The response of plants to drought stress is a complex mechanism that includes molecular changes and extends to the whole plant metabolism, then

affects to the morphology and phenology of plants (Blum, 1996; Chaves et al., 2003; Condon et al., 2004; Molnar et al., 2004). Morphological characters change due to drought stress may occur in both the vegetative and generative phase. In soybean, drought stress at vegetative phase can reduce plant height, number of nodes, root length, root dry weight, and shoot (Sunaryo, 2002). Rice productivity in drought conditions have lower productivity and longer harvest period (Boyer 1992; Richards, 1996; Nguyen et al., 1997; Blum, 2005) compared with normal conditions. The level of losses suffered by plants due to drought depends on some factors i.e. when plants experience lacks of water, water intensity and water duration (Nio and Kandou, 2000).

Drought stress of purple long yard beans are the problems faced in their development efforts on dry land. Therefore, it is necessary to find purple long yard bean varieties that tolerant and suitable in dry land. Naturally, plant potentially adapt to drought stress particularly concerned with the control of transpiration. However, information on the adaptation of plant morphology purple long yard beans to drought stress has not been widely published. This study aims were to observe the tolerance mechanism of purple long yard beans based on its morphological character; to achieve the tolerance hypothesis method of some purple long yard bean cultivars toward drought stress, and to get tolerance cultivars to drought stress.

## **Materials and Methods**

Each of eight purple long yard bean cultivars (UBPHU1-41, UBPHU1-130, UBPU3-153, UBPU1-202, UBPU2-222, UBPU1-365, Brawijaya 4 and Bagong 2) was grown under two drought conditions (100% of field capacity and 50% of field capacity) for 30 days in a 15-cm diameter plastic pot containing 10 kg of soil. Sixteen treatments were arranged in a factorial randomized block design with three replicates.

The experiment was in a glasshouse house of the Faculty of Agriculture, Malang Islamic University from March to June 2014. During the experiment, soil water content was adjusted and maintained 100% field capacity and 50% field capacity by regularly adding water. At harvest (30 days), the morphological characters observed were leaf area, root dry weight, root fresh weight, root length, shoot dry weight, shoot fresh weight, early flowering, number of flowers, number of yard, yard length, yard fresh weight. The degree of tolerance of the purple long bean cultivars toward drought stress was estimated using Stress Susceptibility Index (SSI) that was calculated

based on the formula proposed by Fischer and Maurer 1978):

$$(SSI) = \frac{1 - (Y_s/Y_p)}{1 - (\bar{Y}_s/\bar{Y}_p)}$$

where,  $Y_s$  and  $Y_p$  are the result of a cultivar that was received in a state of drought stress and control conditions, whereas  $\bar{Y}_s$  and  $\bar{Y}_p$  is the average result of all cultivars that gets drought in a state of control conditions.

The criteria to determine the level of drought tolerance are as follows: SSI value < 0.5 for tolerant genotypes, 0.5 < SSI < 1.0 for medium-tolerant genotypes, and SSI > 1.0 for or sensitive genotype.

The selection of variables having large diversity of the grouping tolerance and as a character selection for purple long yard bean genotypes to drought stress was made after a Principle Component Analysis (PCA) and Discriminated Analysis. The PCA was used to reduce the amount of original variable ( $p$ ) which was correlated into a new variable  $q$  which was uncorrelated ( $q < p$ ) without many reducing information of the original variables, so the selection of cultivars can be done by using fewer variables. The number of principal components used was the main component having a root characteristic  $\geq 1$ , because it has a large contribution to diversity. The variable that had been reduced was then further tested by discriminant analysis. This test was aimed to determine the variables that can distinguish groups of tolerant, medium or sensitive cultivars to drought stress.

Quantitative data of morphological observations were statistically analyzed using ANOVA with SPSS Release 15. If the treatment showed a significant effect, it was then continued with Duncan's test at 95% confidence interval.

## **Results and Discussion**

### ***Morphological characters of several cultivars purple long yard bean due to drought***

Drought stress purple long yard bean plants effect on all morphological characters were observed both growth and results except root length and early flowering (Table 1). Drought stress caused all cultivars of purple long yard beans plant flowering faster and showed an increase in root length.

Table 1. Average growth and yield of some cultivars of purple long yard beans due to drought stress

Variable	Treatment	Cultivar							
		UBPU1-41	UBPU1-130	UBPU3-153	UBPU1-202	UBPU2-222	UBPU1-365	Brawijaya 4	Bagong 2
Leaf area (cm <sup>2</sup> )	Control	399.33a/B	538.67a/B	445.00a/B	566.33a/B	517.00a/B	432.33a/B	483.00a/B	650.67b/B
	Stress	238.33b/A	216.00a/A	219.00a/A	90.33a/A	196.33a/A	209.67a/A	81.67a/A	201.67a/A
Root dry weigh (g)	Control	0.37ab/B	0.42abc/B	0.49bc/B	0.68 d/B	0.44bc/B	0.60c/B	0.21a/B	0.47bc/B
	Stress	0.17a/A	0.08c/A	0.12a/A	0.25b/A	0.25b/A	0.19a/A	0.05a/A	0.18a/A
Root fresh weight (g)	Control	0.89a/B	0.72a/B	0.96a/B	0.92a/B	0.81a/B	1.84b/B	0.71a/B	1.15a/B
	Stress	0.43a/A	0.18a/A	0.29a/A	0.33a/A	0.44a/A	0.62ab/A	0.66b/A	0.46a/A
Root length (cm)	Control	22.80b/A	10.15a/A	18.96ab/A	11.89a/A	16.13a/A	18.15ab/A	16.54ab/A	16.37ab/A
	Stress	33.37b/B	23.51a/B	27.96ab/B	20.38a/B	23.13ab/B	23.79a/B	21.65a/B	19.45a/B
Shoot dry weight (g)	Control	4.27c/B	2.18a/B	3.67bc/B	3.35bc/B	3.39bc/B	3.05ab/B	2.56ab/B	3.34b/B
	Stress	1.45a/A	0.81a/A	1.35a/A	1.05a/A	1.88a/A	1.68a/A	1.03a/A	1.52a/A
Shoot fresh weight (g)	Control	14.29cd/B	5.75a/B	16.75d/B	11.06bc/B	14.92cd/B	11.84bc/B	8.57ab/B	10.95bc/B
	Stress	6.78b/A	2.30a/A	3.16a/A	2.58a/A	4.45a/A	5.81a/A	2.51a/A	5.10a/A
Early flowering(day)	Control	52.70b/B	48.27a/B	48.70ab/B	52.00ab/B	50.03ab/B	50.47ab/B	50.53ab/B	51.80ab/B
	Stress	49.78b/A	44.95a/A	46.60ab/A	46.68ab/A	46.70ab/A	46.00ab/A	45.87ab/A	50.13b/A
Number of flower	Control	6.43 b/B	6.85 b/B	6.60 b/B	5.00 a/B	5.25 a/B	4.83 a/B	5.17 a/B	5.33 a/B
	Stress	4.68 c/A	5.00 c/A	5.77 d/A	3.60 a/A	4.67 c/A	4.08 b/A	4.50 c/A	4.42 c/A
Number of yard	Control	6.33 a/B	7.00 a/B	6.67a/B	6.67 a/B	7.67 a/B	6.33 a/B	8.00 a/B	8.33 b/B
	Stress	2.00 a/ A	4.33 b/A	3.33 ab/A	4.33 b/A	3.00 ab/ A	2.33 ab/ A	1.67 a/A	1.33 a/ A
Yard length(cm)	Control	25.30 a/B	28.71 a/ B	27.94 a/ B	27.89 a/ B	28.98 a/ B	22.76 a/B	32.62 b/ B	25.51 a/ B
	Stress	19.98 a/A	19.03 a/ A	15.05 a/ A	15.07 a/ A	18.30 a/ A	15.62 a/ A	24.91 b/A	15.78 a/ A
Yard fresh weight (g)	Control	22.32 a/B	34.41 ab/B	33.56 ab/B	28.66 ab/ B	37.72 ab/ B	25.17 a/B	40.53 b/B	35.19 ab/ B
	Stress	15.38 a/ A	13.24 a/A	10.21 a/ A	9.16 a/A	16.97 a/ A	12.87 a/ A	11.09 a/A	13.61 a/A

Remarks: from the table above, the same capital letter in the vertical case or the same small font in the horizontal case showed that no significant difference at 5%.Duncan's test

Drought stress that started from the vegetative phase also negatively affects the reproductive phase. Drought stress that began in the vegetative phase produces pod, pod length and fresh weight of pod per plant is less than the plants in the control condition. The reduced water availability during the period of yard formation causes a decrease in the results of purple long yard beans. Drought stress is characterized by a decrease in water content, withered, stomata closure and reduction of expansion and also the growth of cells (Prevete et al., 2000; Delfine et al., 2001), which in turn can reduce production.

The magnitude of decline in growth variables and the results of each cultivar purple long yard beans length varied (Figures 1 and 2), except the long of root that increase drought stress. Brawijaya 4 cultivars showed the highest leaf area reduction compared to other cultivars. Plant leaf area decreased with the increase in drought stress. Reduction of leaf area is one of the reactions of plants that can be detected from the lack of moisture in the early vegetative growth.

The declining of the leaf area is one of plant responses toward of lacking water. It is the avoidance mechanisms to suppress the plant from

lacking of water by reducing the occurrence of transpiration. The Much smaller leaf area, it will reduce the sun light, which causes extensive surface area undergo the photosynthesis process narrows. This causes inhibition of plant growth (Wijana, 2001).

The highest fresh weight shoot decline occurred in cultivar UBPU3-153 and UBPU1-202. In general, drought stress will hamper the plant growth that will reduce the amount of biomass produced. This is due to drought stress will inhibit the plant growth and development of plan that cause of the inhibit cells division process. Cell enlargement is highly sensitive to drought stress. It depends on the inhibition of biochemical and molecular processes that occur due to reduced water content in plants. Inhibition of growth is partly due to inhibition of various important processes such as cell wall and membrane biosynthesis, cell division and protein synthesis. Each gram of organic matter making up the average plant requires 500g of water absorbed by the roots are transported to all parts of the plant (Taiz and Zeiger 1991). UBPU1-202 cultivars also decreased highest shoot dry weight.

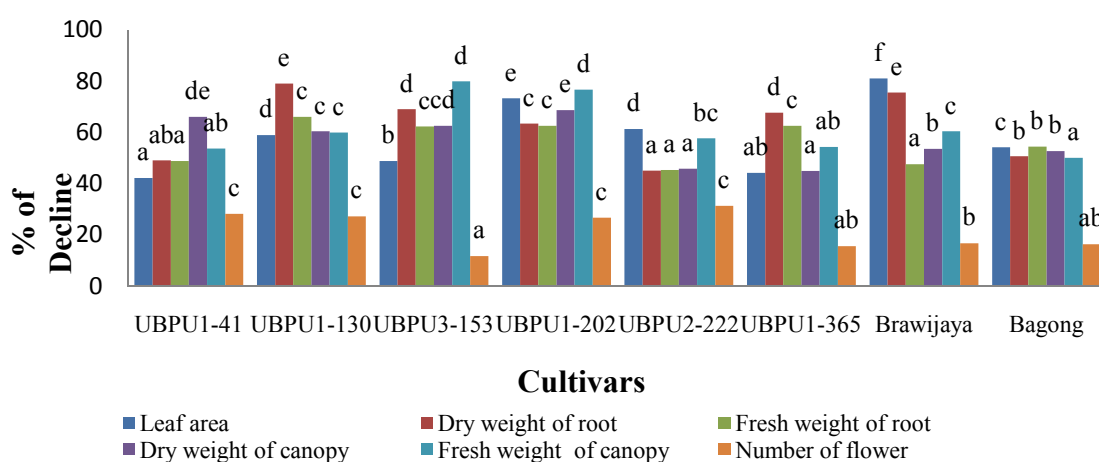


Figure 1. The decrease amount in the growth of some cultivars of purple long yard bean plants due to drought stress. Description: The same letters in the same growth variables showed that no significant difference at the 5% Duncan's tes

The increase of the highest root length was at UBPU1-130 cultivar (Figure 2). Increasing the length root is one of the plants mechanisms to cope with drought stress. The plants ability to respond the condition of drought stress are include of making root extension is indispensable, in order to increase the absorption of water from the soil particles to the bottom layer which still holds water. Root elongation in drought stress

conditions may occur because of the plants have regulatory mechanisms shoots root growth ratio (Taiz and Zeiger, 1991). Root elongation process will also able to reach a larger volume of the soil, so there will be much more to absorb water (Levitt, 1980).

Table 2. Stress Susceptibility Index (SSI) of some cultivars purple long yard beans.

Cultivar	Leaf area		Root dry weight		Root fresh weight		Root length		Shoot dry weight		Shoot fresh weight		Number of flower		Number of yard		Yard length		Early flowering		Yard fresh weight	
	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2	1	2
UBPU1-41	0.62	M	0.81	M	0.90	M	1.01	S	1.13	S	0.81	M	1.41	S	1.13	S	0.61	M	0.81	M	0.63	M
UBPU1-130	0.93	M	1.23	S	1.31	S	1.82	S	1.08	S	0.92	M	1.40	S	0.63	M	0.98	M	1.00	S	1.24	S
UBPU3-153	0.79	M	1.14	S	1.22	S	1.14	S	1.08	S	1.24	S	0.66	M	0.82	M	1.33	S	0.63	M	1.41	S
UBPU1-202	1.33	S	0.94	M	1.12	S	1.33	S	1.18	S	1.18	S	1.45	S	0.58	M	1.33	S	1.49	S	1.38	S
UBPU2-222	0.96	M	0.92	M	0.79	M	0.93	M	0.77	M	1.07	S	0.58	M	1.00	S	1.07	S	0.97	M	1.11	S
UBPU1-365	0.80	M	1.01	S	1.16	S	0.76	M	0.77	M	0.78	M	0.81	M	1.04	S	0.91	M	1.29	S	0.99	M
Brawijaya-4	1.29	S	1.14	S	0.14	T	0.76	M	1.03	S	1.08	S	0.67	M	1.30	S	0.68	M	1.34	S	1.47	S
Bagong-2	1.07	S	0.92	M	1.04		0.51	M	0.93	S	0.82	M	0.89	M	1.38	S	1.10	S	0.47	T	1.24	S

Remarks: S :Sensitive, M : Medium, T :Tolerance, 1: SSI, 2: note

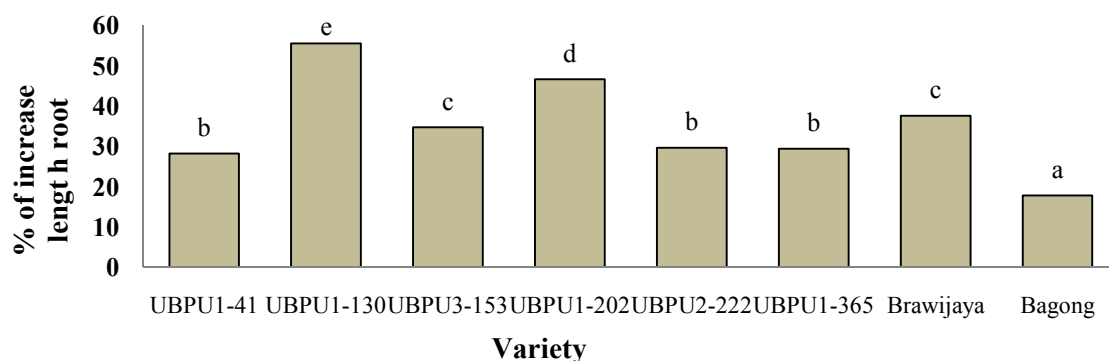


Figure 2. The magnitude of the root length increase in several cultivars of purple long yard bean plants due to drought stress. Description: The same letters in the same growth variables showed that no significant difference at the 5% Duncan's test

The lowest decrease of the long yard beans in cultivar of UBPU1-41 and Brawijaya, the highest decrease occurred in cultivar UBPU3-153 and UBPU1-202. The lowest decrease at the number variable of yard occurred in cultivar UBPU1-202 and UBPU1-130, in the other hand, the highest decrease is in cultivar Bagong 2 (Figure 3). The

lowest decrease of weight variable cultivar occurred in cultivar UBPU1-41 and the highest decrease is in cultivar Brawijaya 4. Jaleel et al. (2008) reported that the cultivars tolerant respond to the stress better with a lower percentage reduction in vegetative and generative growth in drought stress conditions.

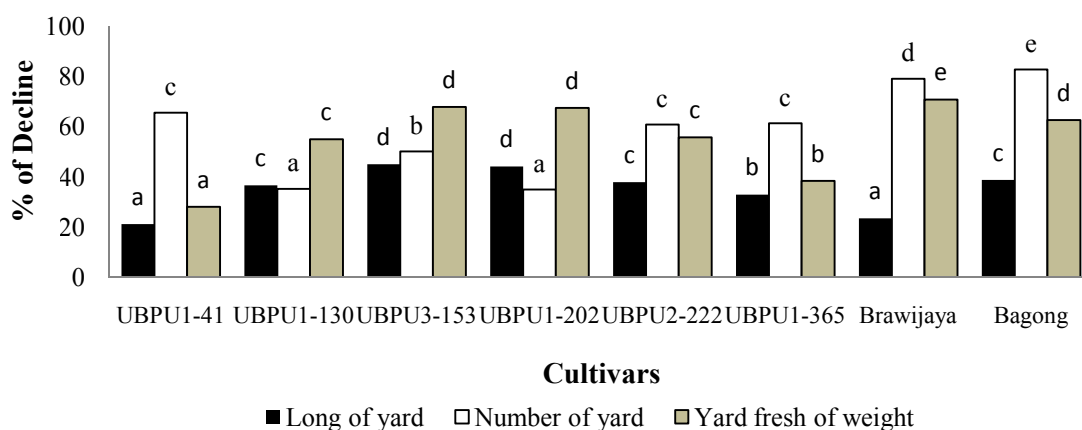


Figure 3. The amount of purple long yard bean crop yield reduction due to drought stress. Description: The same letters in the same outcome variables showed that no significant difference at the 5% Duncan's test

### Tolerance sensitivity test drought stress

Estimation tolerance of purple long yard bean cultivar by using the IS value which is calculated based on the growth and yield variables showed values Sensitivity Index (IS) which is different for each variable, so it is difficult to classify the cultivar tolerance of purple long yard beans to drought stress (Table 2). Determination of the variables that have a major influence on the diversity tolerance of cultivars purple long yard bean to drought stress can be seen from the results of Principle Component Analysis (PCA).

The results of first principal component analysis showed that the variables greatly affect to

the diversity of purple long pods bean cultivars toward drought stress were leaf area, root dry weight, root fresh weight, root length, shoot dry weight, shoot fresh weight, yard length and yard fresh weight. The proportion of diversity was of 50.34%. In the second principal component variables that greatly affected the diversity of cultivars of long yard bean tolerance to drought stress purple was early flowering variety with the proportion of 11.68%. The third major component indicated that the variable of root length greatly affected the tolerance diversity of purple long yard bean to drought stress with diversity proportion of 9.28%. The cumulative major components 1 to 3 were 71.31% (Table 3). To

determine elected components of most influential to drought stress is used discriminant analysis (Table 4). Discriminant analysis (Table 4) shows the value of Wilks' Lambda approaching 0 is the fresh weight of the shoot, it means that the only variable fresh shoot tend to have differences among purple long yard bean cultivars, so that it can be used as the best predictor to predict the cultivar tolerance of chick pea purple yard to drought stress.

Table 3. The main component values of selected variable

Variable	Main Component		
	1	2	3
Leafarea	<b>0.736*</b>	-0.300	0.001
Dry weight of root	<b>0.765*</b>	0.329	-0.015
Fresh weight of root	<b>0.669*</b>	0.226	0.156
length of Root	<b>0.582*</b>	0.119	<b>0.636*</b>
Dry weight of canopy	<b>0.906*</b>	0.252	0.108
Fresh weight of canopy	<b>0.824*</b>	0.353	0.001
Early flower	0.452	<b>0.519*</b>	-0.442
Number of flower	0.464	-0.409	0.422
Yardlength	<b>0.681*</b>	-0.320	-0.351
Fresh weight of yard	<b>0.770*</b>	-0.429	-0.227
Number of yard	<b>0.804*</b>	-0.331	-0.180
Characteristic of root	5.538	1.285	1.021
Proportion diversity (%)	50.343	11.684	9.281
Cumulative diversity (%)	50.343	62.027	71.307

Remarks: \*selected variables that most influence on the tolerance grouping purple long yard beans cultivar to drought stress on components 1, 2, and 3.

Table 4. Correlation between growth variables and the result with the formed discriminant function (Tests of Equality of Group Means)

Variable	Wilks' Lambda	F	df1	df2	Sig.
Leaf Area	0.98	0.12	1	6	0.75
Dry weight of root	0.97	0.21	1	6	0.66
Fresh weight of root	0.78	1.71	1	6	0.24
Length of root	0.91	0.59	1	6	0.47
Dry weight of canopy	0.63	3.47	1	6	0.11
Fresh weight of canopy	<b>0.10</b>	51.67	1	6	0.00
Early flower	0.99	0.03	1	6	0.87
Length of canopy	0.54	5.01	1	6	0.68
Number of canopy	0.69	2.73	1	6	0.15
Fresh weight of canopy	0.77	1.83	1	6	0.23

Further grouping tolerance test cultivars purple yard beans to drought stress based on the results of discriminant analysis result is in accordance with the grouping tolerance using a dendrogram (Figure 4). Sensitive cultivars such as UBPU3-153 and UBPU1-202 characterized as having the character of a decrease in fresh weight of the shoot that is higher than the other cultivars.

Dendrogram using Average Linkage (Between Groups)

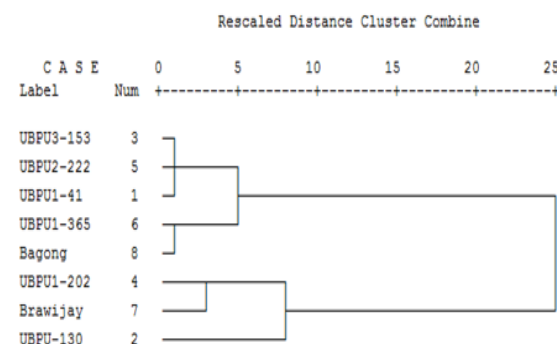


Figure 4. Grouping dendrogram cultivar tolerance purple long yard beans to drought stress by morphological characters

## Conclusion

Drought stress affected the plant morphologies of purple long yard bean that indicated by the decline all of observation variables both growth and results. Morphological responses were also indicated by the faster flowering time and increased root length of all lines. Tolerance Estimation to drought stress using PCA analysis showed that the variables fresh weight shoot could be used as a tolerance predictor of purple long yard bean to drought stress. Tolerance Estimation to drought stress using calculation of the sensitivity index (IS) was not able to classify the tolerance of purple long yard beans to drought stress. The results of PCA analysis followed by discriminant analysis and dendrogram clustering showed that the cultivar UBPU1-202 and UBPU3-153 were cultivars that sensitive to drought stress.

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