DOI 10.18551/rjoas.2021-10.27

SELECTION TRAITS FOR CHILI PEPPER DROUGHT TOLERANCE AT GERMINATION STAGE USING POLYETHYLENE GLYCOL 6000 AND DIVERSITY AMONG 22 CHILI PEPPER GENOTYPES

Millah Zahratul^{1,4*}, Syukur Muhamad^{2,3}, Sobir^{2,3}, Ardie Sintho Wahyuning²

¹Plant Breeding and Biotechnology Program, Faculty of Agriculture,
Graduate School of Bogor Agriculture University, Bogor, West Java, Indonesia

²Department of Agronomy and Horticulture, Faculty of Agriculture,
Bogor Agriculture University, Bogor, West Java, Indonesia

³Center for Tropical Horticulture Studies, Bogor Agriculture University,
Bogor, West Java, Indonesia

⁴Department of Agroecotechnology, Faculty of Agriculture,
Sultan Ageng Tirtayasa University, Indonesia

*E-mail: muhsyukur@apps.ipb.ac.id

ABSTRACT

There is limited information regarding the tolerance of chili varieties to drought in Indonesia. Evaluation of chili drought tolerance at the germination stage using osmotic polyethylene glycol (PEG) is an alternative way to conduct rapid screening in order to assemble drought-tolerant varieties. Twenty two diverse genotypes of chili were evaluated for germination percentage, seed vigour index, root length, shoot length, root-to-shoot length ratio, number of lateral roots, seedling length, root dry weight, shoot dry weight, seedling dry weight, and drought sensitivity index for each variable under normal and drought conditions at laboratory. Considerable genotypic and phenotypic variation was observed for all the characters. Shoot length, root length, root to shoot length ratio, seedling length, shoot dry weight, and percentage of germination can be used as selection character to estimate the drought tolerance of chilli genotypes in the germination stage. The tolerance level of the 22 chilli genotypes varied against drought. Among the 22 genotypes, nine are tolerant, while eleven are moderate and two genotypes are sensitive to drought.

KEY WORDS

Capsicum annuum, drought tolerance, poly ethylene glycol, rapid screening.

Optimal land for cultivation of various agricultural commodities is decreasing from year to year due to competition in land use between various sectors, both agricultural and non-agricultural sectors. An effort to increase crop production in order to fulfill food needs is the utilization of marginal land, one of which is dry land. Dry land is an agroecosystem of land resources that has great potential for agricultural development, both food crops, horticulture (vegetables and fruits) and annual / plantation crops.

Constraints on seasonal horticultural crop cultivation on dry land are non-optimal plant growth and low crop productivity. One of the plant species that is sensitive to water shortages but very important and has a high economic value is chili plant. Drought is one of the abiotic stresses that contribute very significant to the decline of chilli plant's yield (Rosmaina *et al.*, 2018). Severe dryness in chili can reduce yield by 99% (Showemimo and olarewaju 2007).

Many varieties of chili grown in Indonesia but there is less information regarding the tolerance of chili varieties to drought. The part of breeding, which is very important to obtain tolerant varieties to drought is a fast and accurate screening technique (Boopathi *et al.*, 2013). One of the methods of rapid screening for drought tolerance is by simulating drought stress conditions in the laboratory using osmotic polyethylene glycol (PEG).

PEG is an osmoticum material which is able to control media water potential. This material cannot be absorbed by the plant, so it does not poisoning in plant (Verslues *et al.*, 1998). According to Michel and Kaufman (1973), the use of PEG solutions causes a

homogeneous reduction in water potential, and it can be used to mimic the potential of groundwater. Soil osmotic potential in conditions of field capacity is -0.33 bar, equivalent to 5% of PEG (MW 6000) solution, whereas in conditions of critical humidity points the soil osmotic potential reaches -15 Bar or equivalent to 32% PEG (Mexal *et al.*, 1975).

Germination phase and seedling growth are critical phases of drought stress (Li *et al.*, 2011; Channaoui *et al.*, 2017). Screening at this phase is expected to be an alternative to rapid screening and reduce the evaluation of genetic material in order to assembly drought-tolerant varieties. The use of PEG 6000 to identify drought tolerance in the germination phase has been carried out on various crops such as rice (Afa *et al.*, 2013), peanuts (Halimursyadah *et al.*, 2013), chili (Muddarsu and Manivannan, 2017) and cabbage (Channaoui *et al.*, 2017).

Sinaga *et al.*, (2015) stated that PEG concentrations of 10% (equivalent to osmotic potential -1.9 bar) and 15% (equivalent to osmotic potential -4.1 bars) can be used as selective agents for in vitro drought tolerance in eggplant. (Based on preliminary research, 15% PEG concentration or equivalent to osmotic potential -4.1 Bars can differentiate tolerance levels of chili genotypes against drought. This study aims to (1) identify variables that correlate with tolerance to drought stress in the chili germination phase; (2) selecting chili genotypes that are tolerant to drought stress through a rapid test using 15% PEG 6000 concentration at germination phase.

MATERIALS AND METHODS OF RESEARCH

The genetic material used was 22 chili genotypes collections of Genetic and Plant-Breeding-Education laboratory, IPB University. As drought tolerant comparative varieties we used Gada MK F1 which is known as a commercial drought tolerant variety (www.panahmerah.id) and Tit Super which is known to be quite good when planted in areas with moderate to severe drought (Sobir, 1994), while PBC 495 from AVRDC is used as drought sensitive comparative variety (base on preliminary study) (Table 1).

Table 1 – Genetic Material used for screening drought tolerant genotypes

No.	Genotypes	Annotation	Source
1	Cilibangi 3	Red chili, resistance to anthracnose, CVMV and phytophthora Ras 1	Malaysia
2	Jatilaba	high productivity, resistant to bacterial wilt	Panah Merah
3	ICPN 7#3	Bird pepper, resistant to CMV, CVMV, PVY	AVRDC
4	PBC 495	Bird pepper, resistant to CMV and gemini virus	AVRDC
5	VC 211a	resistant to CVMV	AVRDC
6	Tit Super	Red chili, high productivity, resistant to bacterial wilt	Panah Merah
7	Randu	Red chili, high productivity, resistant to bacterial wilt	Local east java
8	CA-MAZ	Ornamental chili, the fruit is purple, and slightly rounded shape	Indramayu
9	Tit segitiga	Red chili, high productivity	
10	Laris	Curly chili	Panah Merah
11	Kopay	Long curly chili (up to 30 cm long)	Payakumbuh IAC
12	Gelora	Red chili	Mutiara Bumi
13	Tombak	Red chili	BISI
14	Bara	Bird pepper	East West Seed Indonesia
15	Gada MK	Red chili hybrid, tolerant to drought	Panah Merah
16	Syakira	Bird pepper	IPB
17	Yuni	Curly chili	IPB
18	Jalapeno	Red chili	Mexico
19	Seloka	Red chili	IPB
20	SSP	Red chili	IPB
21	Bonita	Bird pepper	IPB
22	Anies	Red chili	IPB

The experiment was arranged in a factorial randomized completely block design (RCBD) with three replication. The first factor was drought stress which consisted of two levels, namely control (without the provision of PEG 6000 solution) and a 15% concentration of PEG 6000 solution (w/v) equivalent to -0.41 MPa (Mexal *et al.*, 1975), or equal to -4.1 bar. The second factor was the genotypes of chili (Table 1). Seeds were initially germinated until the radicle appears (± 1 mm). Ten seeds placed in a petri dish containing three layers of filter paper saturated with PEG 0 and 15% solution Three ml of distilled water was added to each

Petri dish under normal conditions every two days to compensate for losses through evaporation. At the same time, 3 ml of 15% PEG solution was added to each Petri dish under osmotic stress conditions. All the Petri dishes were placed in a growth chamber at room temperature (24 ± 2 C) with 80% relative humidity. Observations were carried out for 14 days. Data for germination percentage, vigor index, root length, number of lateral roots, shoot length, root to shoot length ratio, seedling length, root dry weight, shoot dry weight, and seedling dry weight were obtained from 10 seeds in each replication.

Data were analyzed using analysis of variance following Steel *et al.*, (1997). Components of variance, genetic variability, and heritability are estimated, according to Syukur *et al.*, (2011). Genetic variability is categorized as wide ($\sigma^2_g > 2\sigma^2_G$) or narrow ($\sigma^2_g \leq 2\sigma^2_G$) following Pinaria *et al.*, (1995).

Heritability values are classified as low (<20%), moderate (20-50%), and high (> 50%) following Syukur *et al.*, (2010). The principal component analysis (PCA) follows Abdi and Williams (2010), conducted to determine the traits that most contribute to determining differences. The selection of essential selection characters at the germination stage was carried out based on the results of ANOVA and PCA. The characters chosen were those who had significant genotypic value (G), environment (E) and interaction (GxE), and also wide genetic variability and high heritability. PCA confirmed the results of ANOVA.

Furthermore, by using selected variables from the analysis of variance and PCA, the calculation of drought sensitivity index (DSI) was carried out based on Fischer and Maurer (1978) to determine the tolerance level of the 22 genotypes tested. DSI based on Fischer and Maurer (1978) are as follows: $DSI = (1 - Y / Y_p) / (1 - X / X_p)$, where Y = response value of a genotype under drought stress conditions, Y_p = response value of a genotype under non-stress conditions, X = average response value of all genotypes in drought stress conditions, X_p = average response value of all genotypes in non-stress conditions.

The grouping criteria of each variable are as follows: $DSI \leq 0.5$ = tolerant, $0.5 < DSI \leq 1.0$ = moderate, $DSI > 1.0$ = sensitive. Genotype scoring refers to The Misnen *et al.*, (2012) method, to facilitate further evaluation by weighting the criteria of each variable by 2 for tolerant class, 1 for moderate tolerant, and 0 for the sensitive class. Next, genotypes that are tolerant against drought is classified according to the total score. Genotypes that have total scores more than the total number of characters are categorized as tolerant (able to maintain 51-100% growth in drought conditions). Genotypes that have total scores less than half of the number of variables are categorized sensitive (only able to maintain 0-25% growth in drought conditions). Genotypes that have total scores between tolerant and sensitive are categorized as moderate tolerant (able to maintain 26-50% growth in drought conditions).

RESULTS AND DISCUSSION

Analysis of variance revealed highly significant mean for all chilli germination characters except root dry weight among genotypes grown under laboratory condition (Table 2). This explained that genetic factors influence almost all of the characters observed. The coefficient of variation (C.V) was less than 20% for all character, this indicate the precision in the data recorded (Ritonga *et al.*, 2018).

Significant effect in all character was observed, except for the root dry weight and number of the lateral root as a result of 0 and 15% PEG 6000 application to simulate normal and drought condition. The significant effect of PEG showed that the PEG 6000 concentration treatment gave a different response. The use of PEG 6000 solution in germination media causes a decrease in water potential homogeneously so it can be used to mimic the potential of groundwater (Michael and Kauffman 1973). The use of media with 15% PEG 6000 in this study has the effect of drought stress on the seedling. Various studies report that the use of PEG 6000 can distinguish between tolerant and drought genotypes (Afa *et al.*, 2012; Halimursyadah *et al.*, 2013; Widyastuti *et al.*, 2016; Channaoui *et al.*, 2017).

There are significant to very significant interactions between the genotype and the growing environment (G x E) on the germination percentage, shoot length, root length, root to shoot length, seedling length, shoot dry weight, and the number of lateral roots (Table 2).

The G x E interaction is vital in the process of assembling genotypes that are tolerant to stress conditions.

Tolerant plants will grow and produce in extreme environments, such as drought, even though production decreases (Sikuku et al., 2010; Kapoor et al., 2020). Characters that show significant interactions (G x E) can be used as the basis of selection (Akbar et al., 2018). Significant interactions indicate that characters can respond differently to different environmental conditions (Eeuwijk et al., 2016). The mechanism of adaptation between genotypes against drought stress displays different character responses in different environments. Characters that can show differences in each environment among genotypes are essential (Akbar et al., 2018).

Significant differences among genotypes at control treatment (Table 3) were observed for shoot length, root length, root to shoot length, seedling length, shoot dry weight, root dry weight, seedling dry weight, and the number of the lateral root. While at drought condition (15% PEG), significant differences among genotypes were observed for all character, except vigor coefficient, root dry weight, and seedling dry weight.

The phenotypic variance and the phenotypic coefficient of variation (PCV) are higher than the genotypic variance and the genotypic coefficient of variation (GCV) for all traits (Table 4). These results imply the effect of the environment on their genetic expression, or in other words, performance is not only caused by genetic factors but also due to environmental conditions (Ritonga et al., 2018), in this study means there is PEG treatment effect on the performance of all character.

Wide genotypic variance and high heritability (Table 4) for germination percentage, vigor index, shoot length, root length, root to shoot length ratio, shoot dry weight, and the number of the lateral root in stress environment (E2) indicated that influence of genotypes are higher than the environment on the variability of these characters. Genetic diversity and heritability are beneficial in the selection process. Selection will be effective if the population has wide genetic diversity and high heritability (Syukur et al., 2011). According to Jhonson et al., (2009), characters with heritability value >50% are possible to use as selection characters. Principal component analysis (PCA) is used to see the characters that contributed most to shaping diversity. In statistics, PCA is a technique used to simplify data by transforming the data linearly so that a new coordinate system with maximum variation is formed (Miranda et al., 2008). Principal component analysis can be used to reduce the dimensions of a data without reducing the data significantly (Johnson, et al., 1998). The principal component analysis showed that the first principal component (PC 1) provides an explanation of germination variable diversity by 45.0%, while the PC2 explain 22.0% of diversity and PC3 explain 14.0% of diversity (Table 5).

The main component with an eigenvalue greater than one is selected. The principal component with an eigenvalue of less than one is not considered stable in the analysis because it explains less variability than single variables and is not maintained in the study (Girden 2001).

Table 2 – Mean, mean squares and coefficient variation of 22 chilli genotypes in normal (Control) and drought stress (15% PEG6000) environments for various characters recorded under laboratory conditions

Character	Mean		Mean Square			C.V
	Control	15% PEG6000	G	P	G x P	
Germination Percentage (%)	93.03 ± 9.76	83.18 ± 19.23	1.99**	11.25**	1.68**	6.14
Vigor index	2.56 ± 1.29	2.24 ± 0.60	0.6**	1.18**	0.03 ^{ns}	13.51
Vigor Coefficient	25.23 ± 2.17	24.52 ± 2.70	9.17**	16.58*	4.18 ^{ns}	8.18
Shoot length (cm)	3.06 ± 0.49	2.79 ± 0.82	1.05**	1.64*	0.50**	15.74
Root length (cm)	6.39 ± 1.64	5.00 ± 1.80	0.08**	0.57**	0.03*	6.78
Root to Shoot length Ratio (cm)	2.12 ± 0.57	1.86 ± 0.62	0.08**	0.24**	0.04**	7.82
Seedling length (cm)	9.45 ± 1.79	7.96 ± 2.34	0.01**	0.05**	0.002*	3.04
Shoot dry weight (g)	0.0071 ± 0.0020	0.0078 ± 0.0033	1 x 10 ⁻⁵ **	7 x 10 ⁻⁶ *	5 x 10 ⁻⁶ **	0.18
Root dry weight (g)	0.0070 ± 0.0027	0.0092 ± 0.0090	1 x 10 ⁻⁷ ^{ns}	3 x 10 ⁻⁷ ^{ns}	37x 10 ⁻⁸ ^{ns}	0.03
Seedling dry weight (g)	0.0141 ± 0.0043	0.0170 ± 0.0097	5 x 10 ⁻⁵ **	1 x 10 ⁻⁴ *	2 x 10 ⁻⁵ ^{ns}	0.64
Number of lateral roots	2.66 ± 1.29	2.64 ± 1.55	0.4**	0.01 ^{ns}	0.12*	13.78

* significant at 5% level of probability, ** significant at 1% level of probability, ns= not significant.

Eigenvalues are a special set of scalars associated with a linear system of equations (i.e., a matrix equation) that are sometimes also known as characteristic roots, characteristic values (Hoffman and Kunze 1971), proper values, or latent roots (Marcus and Minc 1988). In this sense, we can choose fewer factors than the number of original variables that is variables which contribute more to variability.

Table 3 – Range, mean, percentage decrease under osmotic stress (E2) compared with normal conditions (E1), mean squares and coefficient variation of various characters in 22 chilli genotypes

Character	Environment	Range	Mean	% decrease in E ₂ compared with E ₁	Mean square	C.V
Germination Percentage (%)	E1	60.00-100.00	0.93±0.10	10.59	0.013 ^{ns}	9.71
	E2	30.00-100.00	0.83±0.19		0.091 ^{**}	12.58
Vigor index	E1	1.29-3.03	2.56±0.34	12.54	0.113 ^{ns}	12.53
	E2	0.48-3.08	2.24±0.60		0.898 ^{**}	14.54
vigor coefficient	E1	20.69-30.43	25.23±2.17	2.81	4.929 ^{ns}	7.17
	E2	32.00-15.79	24.52±2.70		8.423 ^{ns}	9.23
Shoot length (cm)	E1	1.82-4.95	3.06±0.49	8.78	0.548 ^{**}	9.91
	E2	0.00-4.76	2.79±0.82		0.001 ^{**}	7.69
Root Length (cm)	E1	3.24-13.96	6.39±1.64	21.71	5.732 ^{**}	17.46
	E2	0.60-10.47	5.01±1.80		0.423 ^{**}	9.60
Root to Shoot Length Ratio	E1	1.03-3.36	2.12±0.57	12.47	0.682 ^{**}	19.02
	E2	0.64-3.73	1.86±0.62		0.097 ^{**}	10.41
Seedling Length (cm)	E1	5.44-18.91	9.45±1.79	17.53	6.849 ^{**}	13.04
	E2	0.600-14.443	7.80±2.34		0.009 ^{**}	7.30
Shoot Dry Weight (g)	E1	0.0029-0.0114	0.0071±0.0020	-9.77	9.75x10 ⁻⁶ ^{***}	14.48
	E2	0.0000-0.0161	0.0078±0.0033		1.02x10 ⁻⁵ ^{***}	0.22
Root Dry Weight (g)	E1	0.0005 -0.0136	0.0070±0.0027	-31.54	1.81x10 ⁻⁵ ^{***}	20.36
	E2	0.002-0.057	0.0092±0.0090		4.05x10 ⁻⁵ ^{ns}	0.87
Seedling Dry Weight (g)	E1	0.0059-0.0235	0.0141±0.0043	-20.60	4.85x10 ⁻⁵ ^{***}	14.37
	E2	0.002-0.0623	0.0170±0.0097		5.49x10 ⁻⁵ ^{ns}	0.89
Number of Lateral Root	E1	0.50-8.36	2.66±1.29	0.51	0.198 ^{**}	12.89
	E2	0.00 - 8.50	2.64±1.55		0.005 ^{**}	3.32

* significant at 5% level of probability, ** significant at 1% level of probability, ns= not significant.

Table 4 – Estimation of genotype coefficient of variation, phenotype coefficient of variation, genetic variability, phenotype variability and broad sense heritability of 22 chilli genotypes in normal (E₁) and drought stress (E₂) environments for various characters recorded under laboratory conditions

Character	Environment	GCV	PCV	Genetic Variability			Phenotypic Variability			Heritability	
				σ^2_g	$2\sigma^2_g$	Category	σ^2_p	$2\sigma^2_p$	Category	h ² (bs)	Category
Germination Percentage (%)	E1	4.31	7.07	0.0016	0.0035	narrow	0.0043	0.0026	wide	37.17	medium
	E2	19.63	20.93	0.0267	0.0182	wide	0.0303	0.0179	wide	87.97	High
Vigor index	E1	2.24	7.57	0.0033	0.0381	narrow	0.0376	0.0221	wide	8.75	Low
	E2	22.96	24.45	0.2641	0.1794	wide	0.2994	0.1766	wide	88.21	High
vigor coefficient	E1	2.95	5.08	0.5535	1.3821	narrow	1.6430	0.9690	wide	33.69	medium
	E2	4.28	6.83	1.1019	2.2634	narrow	2.8077	1.6559	wide	39.25	medium
Shoot length	E1	12.75	13.97	0.1522	0.1113	wide	0.1828	0.1078	wide	83.24	high
	E2	0.59	0.72	0.0003	0.0003	wide	0.0004	0.0002	wide	68.71	high
Root Length	E1	19.12	21.62	1.4950	1.1878	wide	1.9106	1.1268	wide	78.25	high
	E2	5.98	6.55	0.0895	0.0655	wide	0.1076	0.0634	wide	83.25	high
Root to Shoot Length Ratio	E1	19.58	22.45	0.1730	0.1428	wide	0.2273	0.1341	wide	76.08	high
	E2	9.54	10.49	0.0315	0.0232	wide	0.0380	0.0224	wide	82.82	high
Seedling Length	E1	14.10	15.98	1.7761	1.4222	wide	2.2829	1.3464	wide	77.80	high
	E2	1.63	1.91	0.0161	0.0143	wide	0.0223	0.0131	wide	72.25	high
Shoot Dry Weight	E1	24.09	25.51	2.9 x 10 ⁻⁶	1.9 x 10 ⁻⁶	wide	3.3x10 ⁻⁶	1.9x10 ⁻⁶	wide	89.23	high
	E2	20.80	23.77	2.6 x 10 ⁻⁶	2.1 x 10 ⁻⁶	wide	3.4x10 ⁻⁶	2.0x10 ⁻⁶	wide	76.57	high
Root Dry Weight	E1	33.05	35.08	5.4 x 10 ⁻⁶	3.6 x 10 ⁻⁶	wide	6.0x10 ⁻⁶	3.6x10 ⁻⁶	wide	88.77	high
	E2	8.03	39.91	5.5 x 10 ⁻⁷	1.4 x 10 ⁻⁵	narrow	1.3x10 ⁻⁵	8.0x10 ⁻⁶	wide	4.05	low
Seedling Dry Weight	E1	27.35	28.58	1.5 x 10 ⁻⁵	9.6 x 10 ⁻⁵	wide	1.6x10 ⁻⁵	9.5x10 ⁻⁵	wide	91.56	high
	E2	12.67	25.22	4.6 x 10 ⁻⁶	1.6 x 10 ⁻⁵	narrow	1.8x10 ⁻⁵	1.1x10 ⁻⁵	wide	25.26	medium
Number of Lateral Root	E1	8.10	9.67	0.0464	0.0428	wide	0.0660	0.0390	wide	70.19	high
	E2	1.31	1.54	0.0012	0.0011	wide	0.0016	0.0010	wide	73.10	high

Note: GCV = genotypic coefficient of variation, PCV = phenotypic coefficient of variation, σ^2_p = phenotypic variance, σ^2_g = genotypic variance, $\sigma^2_{e_g}$ = standard deviation of genotypic variance, $\sigma^2_{e_p}$ = standard deviation of phenotypic variance.

Table 5 – Principal component analysis of germination characters

Character	Komponen utama 1	Komponen utama 2	Komponen utama 3
Germination Percentage	0.34*	-0.03	-0.14
Vigor index	0.33*	-0.30*	0.37*
Vigor Coefficient	-0.07	-0.41*	0.53*
Shoot length	0.37*	-0.246	0.24*
Root length	0.37*	0.105	0.2
Root to Shoot length Ratio	0.20	-0.34*	-0.51*
Seedling length	0.33*	-0.244	-0.35*
Shoot dry weight	0.28	-0.206	-0.10
Root dry weight	0.30*	0.43*	0.14
Seedling dry weight	0.31*	0.30*	-0.17
Number of lateral roots	0.29	0.42*	0.19
Eigenvalue	4.95	2.43	1.48
Proportion	0.45	0.22	0.14
Cumulative	0.45	0.67	0.81

* component scores above the average score of each main component.

Table 6 – Drought sensitivity index of selected characters from 22 chili genotypes in drought stress conditions using 15% PEG 6000

Genotype	Shoot length	Root length	Root to shoot length ratio	Seedling length	Shoot dry weight	Germination percentage
C5	-0.28	1.24	2.44	1.10	5.00	0.63
C7	-0.30	-0.24	-0.26	-0.25	0.22	-1.05
C8	1.17	0.26	-0.52	0.44	2.85	1.35
C12	2.88	0.08	-3.86	0.63	-4.33	5.21
C10	1.04	1.53	2.17	1.42	2.72	0.00
C18	1.00	0.34	-0.15	0.44	4.24	-0.70
C19	-1.43	0.64	1.84	0.39	0.45	1.75
C20	1.10	1.60	2.24	1.53	4.02	-1.89
C37	-0.78	0.32	1.16	0.15	5.00	-0.33
C51	-2.84	1.54	3.91	1.01	7.13	0.65
C120	-0.58	-0.16	0.22	-0.22	-3.59	3.58
C142	1.71	0.92	0.29	1.04	-3.16	2.87
C143	8.05	2.91	-3.14	3.75	-6.85	4.56
C145	2.12	0.78	-0.42	1.01	0.37	0.31
Gada MK	1.60	0.84	0.43	0.98	2.07	0.31
Syakira	4.82	2.22	0.73	2.63	0.82	-0.36
Yuni	-2.54	1.79	4.33	1.17	-1.22	2.28
Jalapeno	0.38	0.23	-0.07	0.26	0.03	0.67
Seloka	2.38	0.94	-0.06	1.17	3.51	1.30
SSP	1.13	0.07	-0.91	0.27	3.24	0.00
Bonita	-0.53	2.58	4.92	2.05	4.94	0.00
Anis	-0.06	1.12	2.21	0.95	2.13	0.33

Table 7 – Characterization of selected traits based on drought sensitivity index from 22 chili genotypes in drought stress conditions using 15% PEG 6000

Genotype	Shoot length	Root length	Root to shoot length ratio	Seedling length	Shoot dry weight	Germination percentage
C5	tolerant	sensitive	sensitive	sensitive	sensitive	moderate
C7	tolerant	tolerant	tolerant	tolerant	tolerant	tolerant
C8	sensitive	tolerant	tolerant	tolerant	sensitive	sensitive
C12	sensitive	tolerant	tolerant	moderate	tolerant	sensitive
C10	sensitive	sensitive	sensitive	sensitive	sensitive	tolerant
C18	moderate	tolerant	tolerant	tolerant	sensitive	tolerant
C19	tolerant	moderate	sensitive	tolerant	tolerant	sensitive
C20	sensitive	sensitive	sensitive	sensitive	sensitive	tolerant
C37	tolerant	tolerant	sensitive	tolerant	sensitive	tolerant
C51	tolerant	sensitive	sensitive	sensitive	sensitive	moderate
C120	tolerant	tolerant	tolerant	tolerant	tolerant	sensitive
C142	sensitive	moderate	tolerant	sensitive	tolerant	sensitive
C143	sensitive	sensitive	tolerant	sensitive	tolerant	sensitive
C145	sensitive	moderate	tolerant	sensitive	tolerant	tolerant
Gada MK	sensitive	moderate	tolerant	moderate	sensitive	tolerant
Syakira	sensitive	sensitive	moderate	sensitive	moderate	tolerant
Yuni	tolerant	sensitive	sensitive	sensitive	tolerant	sensitive
Jalapeno	tolerant	tolerant	tolerant	tolerant	tolerant	moderate
Seloka	sensitive	moderate	tolerant	sensitive	sensitive	sensitive
SSP	sensitive	tolerant	tolerant	tolerant	sensitive	tolerant
Bonita	tolerant	sensitive	sensitive	sensitive	sensitive	tolerant
Anis	tolerant	sensitive	sensitive	moderate	sensitive	tolerant

Table 8 – Classification of drought tolerance from 22 chili genotypes by scoring drought sensitivity index of selected characters in drought stress conditions using PEG 6000 15%

No.	Genotype	Shoot length	Root length	Root to Shoot Length Ratio	Seedling length	Shoot dry weight	Percentage of germination	Total score	Classification of tolerance
1.	C5	2	0	0	0	0	1	3	moderate
2.	C7	2	2	2	2	2	2	12	tolerant
3.	C8	0	2	2	2	0	0	6	moderate
4.	C12	0	2	2	1	2	0	7	tolerant
5.	C10	0	0	0	0	0	2	2	sensitive
6.	C18	1	2	2	2	0	2	9	tolerant
7.	C19	2	1	0	2	2	0	7	tolerant
8.	C20	0	0	0	0	0	2	2	sensitive
9.	C37	2	2	0	2	0	2	8	tolerant
10.	C51	2	0	0	0	0	1	3	moderate
11.	C120	2	2	2	2	2	0	10	tolerant
12.	C142	0	1	2	0	2	0	5	moderate
13.	C143	0	0	2	0	2	0	4	moderate
14.	C145	0	1	2	0	2	2	7	tolerant
15.	Gada MK	0	1	2	1	0	2	6	moderate
16.	Syakira	0	0	1	0	1	2	4	moderate
17.	Yuni	2	0	0	0	2	0	4	moderate
18.	Jalapeno	2	2	2	2	2	1	11	tolerant
19.	Seloka	0	1	2	0	0	0	3	moderate
20.	SSP	0	2	2	2	0	2	8	tolerant
21.	Bonita	2	0	0	0	0	2	4	moderate
22.	Anis	2	0	0	1	0	2	5	moderate

Figure 1 – Biplot of 11 germination character on PC1, PC2 and PC3

The first principal component has the highest proportion value of 45%, which indicates that this main component can explain the diversity of data by up to 45%. The accumulation of diversity which can be explained by PC1, PC2, and PC3 is 81.0% Table 5; Figure 1). It means that the characters contained in these three principal components are characters that have a major contribution in shaping population diversity in a drought stress environment.

The characters chosen in each of the main components are characters which have component scores above the average score of each major component (Girden 2001). Based on the results of ANOVA and PC, six characters were chosen as selection characters, namely germination percentage, shoot length, root length, root to shoot length ratio, seedling length and shoot dry weight. Although number of lateral root interactions are significant and have broad genetic variability and high heritability, based on PC analysis the number of lateral roots does not contribute to shaping diversity in populations under drought conditions while the character of Vigor index, vigor coefficient, root dry weight, and seedling dry weight are not chosen as character selection because the interactions are not significant.

Drought Sensitivity Index. Drought sensitivity index (DSI) is one index that can be used to assess the decrease in yield due to the suboptimum environment compared to the optimum environment (Fisher and Maurer 1978). Low DSI values indicate that genotypes tested at sub-optimum conditions did not show a large decrease so that the genotype can be said to be tolerant.

The characters used for calculating DSI values were selected from the results of the analysis of variance, genetic parameter and the principal components analysis. These characters are shoot length, root length, root to shoot length ratio, seedling length, shoot dry weight, and percentage of germination. DSI values of the variables selected from 22 genotypes can be seen in table 6. Characterization of each drought-related characteristic is presented in Table 7. Scoring and classification of genotypes according to their ability to deal with drought stress, which refers to the method of Misnen *et al.*, (2012) are presented in Table 8. Scoring results indicate that two genotypes are sensitive, namely C10 and C20. There are 11 genotypes with moderate categories, namely C5, C8, C51, C142, C143, Gada, Syakira, Yuni, Seloka, Bonita, and Anis while the other nine genotypes are tolerant categories, namely C7, C12, C18, C19, C37, C120, C145, Jalapeno, and SSP.

Based on this study, the genotype C10 as the sensitive comparison genotype was in the same category, but the Gada MK genotype, as the tolerant genotype comparison, based on this study was in the moderate tolerant category. This difference category is possible because plants that experience drought will use more than one mechanism to defend themselves (Mitra 2001). The tolerance mechanism will be different in each growth phase and type. Therefore, these characteristics should be further examined in drought conditions at a later stage.

CONCLUSION

At the germination phase, 15% PEG 6000 solution can be used to detect drought tolerance in chili. Characteristics of shoot length, root length, root to shoot length ratio, seedling length, shoot dry weight, and percentage of germination can be used as character selection to detect the drought tolerance of chili genotypes in the germination phase.

Chili genotypes C7, C12, C18, C19, C37, C120, C145, Jalapeno, and SSP were drought tolerant. Genotypes C5, C8, C51, C142, C143, Gada, Syakira, Yuni, Seloka, Bonita, and Anis were moderately drought tolerant. While genotypes C10 and C20, belonged to sensitive groups.

REFERENCES

1. Abdi, H., L.J. Williams. 2010. Principal component analysis. WIREs Computational Statistics. 2:433-459.

2. Afa, L., B.S. Purwoko, A. Junaedi, O. Haridjaja, I.S. Dewi. 2013. Early Detection of Hybrid Rice Tolerance to Drought Using PEG 6000. *J. Agron. Indonesia*. 41:9-15.
3. Boopathi, N.M., G. Swapnashri, P. Kavitha, S. Sathish, R. Nithya, W. Ratnam, A. Kumar. 2013. Evaluation and bulked segregant analysis of major yield QTL qtl 12.1 introgressed into indigenous elite line for low water availability under water stress. *Rice Science*. 20:25-30.
4. Channaoui, S., R. El Kahkahi, J. Charafi, H. Mazouz, M. El Fechtali, A. Nabloussi. 2017. Germination and Seedling Growth of a Set of Rapeseed (*Brassica napus*) Varieties under Drought Stress Conditions. *Internat. J. Environ. Agric. Biotechnol.* 2:487-494.
5. Eeuwijk, F.A.V., D.V. Bustos-Korts, M. Malosetti. 2016. What should students in plant breeding know about the statistical aspects of genotype environment interactions. *Crop Sci.* 56:2119-2140. Fischer RA, Maurer R. 1978. Drought resistance in spring wheat cultivars. I. Grain yield response. *Aust. J. Agric. Res.* 29:897-907.
6. Girden, E.R. 2011. Evaluating research article from start to finish. Thousands Oaks. Calif., Sage Publ.
7. Halimursyadah, A.I. Hereri, A. Hafnizar. 2013. The Use of Polyethylene Glycole as Simulating Media of Drought Stress on Viability and Vigor of Seeds of Some Varieties of Peanut (*Arachis hypogaea* L.) on Germination Stadia. *J. Floratek.* 8:73-79
8. Hoffman, K., R. Kunze. 1971. Linear Algebra. 2nd ed. Englewood Cliffs, New Jersey: Prentice-Hall International Inc. p. 182.
9. Johnson, Richard A., Wichern, Dean W. 1998. Applied Multivariate Statistical Analysis. New Jersey: Prentice-Hall International Inc.
10. Kapoor, D., S. Bhardwaj, M. Landi, A. Sharma, M. Ramakrishnan, A. Sharma. 2020. The impact of drought in plant metabolism: how to exploit tolerance mechanisms to increase crop production. *Appl. Sci.* 10 (5692).
11. Kulkarni, M., U. Deshpande. 2007. In vitro screening of tomato genotypes for drought resistance using polyethylene glycol. *African J. Biotech.* 6:691-696.
12. Li, F.L., W.K. Bao, N. Wu. 2011. Morphological, anatomical and physiological responses of *Campylotropis polyantha* (French.) Schindl. seedlings to progressive water stress. *Sci HorticAmsterdam*. 127:436-43.
13. Marcus, M., H. Minc. 1988. Introduction to Linear Algebra. New York: Dover, p. 145.
14. Mexal, J., J.T. Fisher, J. Osteryoung, C.P. Reid. 1975. Oxygen availability in polyethylene glycol solutions and its implications in plant-water relations. *Plant Physiol.* 55:20-24.
15. Miranda, A.A., Y. A. Le Borgne, G. Bontempi. 2008. New Routes from Minimal Approximation Error to Principal Components. *Neural Processing Letters, Springer*. Volume 27 (3).
16. Misnen, E.R. Palupi, M. Syukur, Yudiwanti. 2012. Genotypes Screening of Physic Nut (*Jatropha curcas* L.) for Tolerance to Drought. 2012. *J. Agron. Indonesia*. 40: 232- 238.
17. Mitra J. 2001. Genetics and genetics Improvement of drought resistance in crop plants. *Curr. Sci.* 80(6): 758-763
18. Muddarsu, V.R., S. Manivannan. 2017. In Vitro Screening of Chilli (*Capsicum Annuum* L.) Cultivars for Drought Tolerance. *Chem Sci Rev Lett.* 6(24):2636-2644
19. Pinaria, A., A. Baihaki, R. Setiamihardja, A.A. Daradjat. 1995. Variabilitas genetik dan heritabilitas karakter-karakter biomassa 53 genotipe kedelai. *Zuriat* 6:88-92.
20. Ritonga, A.W., M.A. Chozin, M. Syukur, A. Maharijaya, Sobir. 2018. Short Communication: Genetic variability, heritability, correlation, and path analysis in tomato (*Solanum lycopersicum*) under shading condition. *Biodiversitas.* 19:1527-2531.
21. Rosmaina, Sobir, Parjanto, A. Yunus, 2018. Selection criterias development for chili pepper under different field water capacity at vegetative stage. *Bulg. J. Agric. Sci.* 24:80–90.
22. Showemimo, F.A., J.D. Olarewaju. 2007. Drought tolerance indices in sweet pepper (*Capsicum annum* L.). *Int. J. Plant Breed. Genet.* 1:29–33.
23. Sikuku, P.A., G.W. Netondo, D.M. Musyimi, J.C. Onyango. 2010. Effects of water deficit on days to maturity and yield of three NERICA rainfed rice varieties. *ARPN J. Agric. Biol. Sci.* 5:1-9.

24. Sinaga, E., M.S. Rahayu, A. Maharijaya. 2015. In Vitro Selection of Sixteen of Eggplant (*Solanum melongena* L.) Accessions to Drought with Tolerance Polyethylene Glycol (PEG). *J. Hort. Indonesia*. 6:20-28.
25. Steel, R.G.D., J.H. Torrie. 1981. *Principles and Procedures of Statistics: A Biometrical Approach*. 2nd ed. McGraw Hill Book, New York.
26. Syukur, M., S. Sujiprihati, R. Yuniarti, K. Nida. 2010. The estimation of varian component, heritability, and correlation to determine selection criteria in the F5 population of pepper (*Capsicum annuum* L.) populasi F5. *J. Hort. Indonesia*. 1:74-80.
27. Syukur, M., S. Sujiprihati, R. Yuniarti, D.A. Kusumah. 2011. Estimation of genetic variance and heritability for yield component characters in chili pepper genotypes. *J. Agrivigor*. 10:148-156.
28. Widyastuti, Y., B.S. Purwoko, M. Yunus. 2016. Identification of Drought Tolerance of Hybrid Rice Parental Lines (*Oryza sativa* L.) at Germination Stage Using Polyethylene Glycol (PEG) 6000. *J. Agron. Indonesia*. 44:235-241.