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Assessment of Soybean Genotypes for PEG Induced Drought Tolerance at Germination and Seedling Level

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Crop plants are often exposed to drought and high temperature stresses that decrease crop yield worldwide and the problem is expected to further accentuate in future. Drought is one of the most important environmental stresses affect the growth and development thereby reducing the yield to greater extent in many crops. Among the oilseeds, soybean plays key role as it contains high protein content and it is highly sensitive to drought stress. Developing soybean (*Glycine max*) cultivars that can perform well in drought and other abiotic stress is considered as important to attain global stability. Germination is one of the vital growth stages for seedling establishment and success in this stage is highly dependent on moisture availability in the soil. A laboratory experiment was conducted to assess the germination associated traits of forty seven genotypes of soybean under Polyethylene Glycol (PEG) induced drought. The optimum osmotic stress levels were maintained from 0 to -5 bars by altering the concentration of PEG. This investigation was performed in factorial experiment using Completely Randomized Design (CRD) with three replications. Germination and early growth were affected by drought stress. Decreasing water potential or increasing moisture stress adversely affected germination and all seedling growth related characters with every attribute reducing significantly with level of decreasing water potentials. The extent of reduction varied with the genotypes and water potential. The variation among genotypes for germination percentage, vigour index, stress tolerance index (STI), was found to be significant and reliable indicators to screen the drought tolerant genotypes at germination and seedling stage in soybean.

Key words: Drought, Germination percentage, Tolerance index, Vigour index.

Worldwide agricultural productivity is subject to increasing environmental constraints in the form of abiotic stresses that adversely influence plants growth and development causing crop failure and decreasing average yields more than 50% (Mittler 2006; Wu *et al.,* 2011). Water is the most important and widely operative limiting factor for crop production. Responses of plants to water deficit condition have been employed to make a physiological evaluation of drought resistance. Breeding for drought resistance has been accomplished by selecting for seed yield under field conditions (Brown *et al.,* 1985; Specht *et al.,* 1986), but since such procedures require full season field data, this is not always an efficient approach, especially in dry locations (Sammons *et al.,* 1978). Another alternative has been to screen material under laboratory or greenhouse conditions using seeds or seedlings as test material.

Among the various traits which help assess drought tolerance, seed germination percentage and root traits are more reliable on account of their high correlation with drought tolerance mechanism (Chang *et al.,* 1982). Plant breeding techniques has proved very handy for the identification of stress-tolerant genetic traits in various crops and cultivars (Ashraf and Mehmood, 1990). Different workers have used several methods to evaluate genetic differences in drought tolerance, but most of them have limited application. The main methods for screening for drought tolerance in crop plants is the observations in seed germination influenced by osmoticum substances and further growth or survival of young seedlings subjected to soil or simulated water stress (Sullivan, 1979). Tolerance indices might be useful to screen the genotypes as dry weight of a plant at particular age is universally considered as a more stable character than other morphological parameters (Dutta and Bera, 2008). In the present investigation attempts have been made to identify tolerant genotypes based on germination percentage, vigour index, and tolerance index under PEG induced drought.

Material and Methods

The experiment was conducted in the laboratory of Crop Physiology, Tamil Nadu Agricultural University, Coimbatore, during kharif season 2017. Mature seeds of soybean variety was used for standardization of PEG concentration based on which forty seven genotypes were screened. Various parameters such as germination percentage, vigour index, percent reduction of shoot root growth over control, stress tolerance index and tolerance index were calculated.

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The study was performed using petriplates with filter paper as medium for germination. Seeds were selected for size, homogeneity and were surface sterilized for three min in 1% (v/v) sodium hypochlorite and then rinsed twice with distilled water. Ten seeds of each genotype were placed in the petridishes with two treatments *viz*, control and -3 bar PEG concentration and kept in an incubator (60% relative humidity) at 28°C. Respective PEG solution was applied on daily basis after draining out the previously applied solution. Number of seeds germinated was manually counted on 7th day and the seed germination associated traits were recorded. After seven days, seedling vigour index was measured using the protocol given by Abdul-Baki and Anderson (1973). The treatment was replicated thrice and germination count was taken seven days after sowing and is expressed in percentage.

The length of shoot and root of ten seedlings from the seed to the tip of the root to the tip of the leaf were recorded and expressed in centimeters. The percent reduction of shoot and root growth over control was calculated using the formula suggested by Senthil Kumar (2001). Fresh weights of different plant parts were taken immediately, and then dried in a hot air oven at 80 ± 2°C for 72 hours. Tolerance index (TI) based on dry weight was calculated using formula given by Maiti *et al.,* 1994 and stress tolerance index was calculated using formula given by Dhopte and Livera (1989).

Number of germinated seedlings

Germination percentage = -------------------x 100

Number of seeds taken for germination

Vigour index = (Average shoot length + Average root length) x Germination percentage

Vigour index of the treated seedling

Stress tolerance index = -------------------- x 100

Vigour index of the control seedling

Percent reduction shoot and root growth over control = [(C-R)/C] x 100

Where,

C – Shoot and root length of control sedling

R – Shoot and root length of stressed seedling

Dry weight of seedlings in stress (g)

Tolerance Index (TI) = ---------------------------------

Dry weight of seedlings in control (g)

Statistical analysis was carried out as per Gomez and Gomez (1984).

Results and Discussion

Germination percentage

Seed germination determines the standard establishment and final yield of the crop. It is one of the most important traits in early stage of growth under moisture stress conditions. It is an essential process in plant development to obtain optimal seedling number that result in high seed yield. Lower water potential created by PEG 6000 significantly reduced the seed germination percentage (Wafa'a *et al*., 2010). Osmotic stress caused by PEG decreased the germination percentage, seedling survival and growth of plants Aniat-ul-Haq *et al.,* (2010). Responses to PEG concentration varied among genotypes indicating the genetic variability for germination associated traits. Among the genotypes higher germination percentage was recorded in Cat 1260 (93.33) and lower in Cat 1847B, Cat 411B, Cat 2034B, Cat 292, Cat 1826, Cat 1878, Cat 2070B,Cat 2057, Cat 2065, PS 1343,Cat 2090, Cat 2049, Cat 2086B, Cat 313A, Cat 1293B, Cat 2005, Cat 1995, Cat 2024, Cat 2058, Cat 1969 (0.00) (Table 1). It is evident from the study that there is a significant difference in germination percentage among the genotypes and in the interaction of genotypes also. Germination of seeds involve the activation of enzyme systems as well as mobilization of reserve foods and these process are may adversely affected by PEG induced drought stress. Higher germination percentage of genotype showed high correlation with drought tolerance mechanism (Chang *et al.,* 1982), which support the present study information.

Slow and poor germination under water stress is obviously due to decreased water potential of the germination medium, which restricted the water availability to the seeds (Soltani *et al.*, 2002). The germination percentage decreased with increasing salt concentration. Among the experimental genotypes, higher germination reduction over control was observed in Cat 1847B, Cat 411B, Cat 2034B, Cat 292, Cat 1826, Cat 1878, Cat 2070B, Cat 2057, Cat 2065, PS 1343, Cat 2090, Cat 2049, Cat 2086B, Cat 313A, Cat 1293B, Cat 2005, Cat 1995, Cat 2024, Cat 2058, Cat 1969 (100%) and the lower in Cat 1260 and Cat 1847 ie., 7.33 percentage and 13.33 percentage respectively (Table 1). Seed germination and seedling emergence were significantly affected by decreasing water potential. Reduction in germination at higher level of moisture stress may be attributed to the moisture deficit in seeds below the threshold, which may lead to degradation and inactivation of the essential hydrolytic and other group of enzymes as suggested by Wilson (1971). Similar type of reduction in germination of different crops under moisture stress has been already reported in wheat (Jong and Best, 1979; Winter *et al.*, 1988).

Vigour index

Tolerant rice cultivar recorded higher vigour index than sensitive cultivar under stress as indicated by Sadasivam *et al.,* (2000). The interaction between genotypes and PEG concentration was highly significant in this study. Among the genotypes, higher vigour index was observed in PS 1343 (1980.00), Cat 1260 (1727.00), while lowest in Cat 411B (148.86), Cat 2090 (172.89) in control treatment (Table 1). In PEG treatment higher vigour index was observed

Table 1. Effect PEG 6000 (-3 bar) on germination (%), % reduction of germination over control, Vigour index on soybean genotypes

Genotypes	Germination (%)			% reduction of germination over control	Vigour index		
	Control	Treatment			Control		Treatment
Cat 1987B	93.33	0.00		100.00	633.71		0.00
Cat 1809	100.00	46.67		53.33	1161.40		113.18
Cat 140A	100.00	53.33		46.67	902.00		143.99
Cat 259B	80.00	23.33		70.84	520.00		51.33
Cat 1157	100.00	0.00		100.00	502.50		0.00
Cat 2047	90.00	46.67		70.84	665.10		99.55
Cat 1942	93.33	40.00		57.14	1119.03		102.00
Cat 411B	73.33	0.00		100.00	148.86		0.00
Cat 1937	100.00	13.33		86.67	948.00		26.66
Cat 1912	100.00	20.00		80.00	1086.00		55.00
Cat 2034B	100.00	0.00		100.00	952.50		0.00
Cat 2036	90.00	30.00		60.00	1298.70		51.00
Cat 292	53.33	20.00		33.33	287.98		20.00
Cat 1826	100.00	0.00		100.00	1096.00		0.00
Cat 1878	70.00	0.00		100.00	630.00		0.00
Cat 2070B	100.00	0.00		100.00	770.00		0.00
Cat 2057	100.00	0.00		100.00	1460.00		0.00
Cat 1328	100.00	0.00		86.67	1160.00		0.00
Cat 2065	90.00	0.00		100.00	568.30		0.00
PS 1343	100.00	0.00		100.00	1980.00		0.00
Cat 2090	63.33	0.00		100.00	172.89		0.00
Cat 2049	80.00	0.00		100.00	320.00		0.00
Cat 2067	86.67	13.33		73.34	572.02		8.00
Cat 1290	73.33	10.00		63.33	463.45		20.00
Cat 2086B	100.00	0.00		100.00	1183.00		0.00
Cat 313A	100.00	0.00		100.00	1223.00		0.00
Cat 1293B	53.33	0.00		100.00	377.58		0.00
Cat 1091	63.33	33.33		30.00	360.98		77.66
Cat 2005	86.67	0.00		100.00	542.81		0.00
Cat 1995	73.33	0.00		100.00	637.97		0.00
Cat 1811	93.33	43.33		50.00	989.31		112.66
Cat 1847	100.00	86.67		13.33	1155.00		249.61
Cat 2024	90.00	0.00		100.00	554.40		0.00
Cat 1260	100.00	93.33		7.33	1727.00		476.92
Cat 2058	73.33	0.00		100.00	481.78		0.00
Cat 2722	100.00	53.33		46.67	855.00		180.26
Cat 19934	63.33	13.33		50.00	658.63		17.33
UPSL 229	100.00	53.33		46.67	1702.00		117.33
Cat 120	100.00	46.67		53.33	548.00		101.74
Cat 1267B	100.00	23.33		76.67	582.50		31.51
Cat 1969	100.00	0.00		100.00	527.10		0.00
Cat 2086A	100.00	13.33		86.67	662.50		8.00
Cat 2084	100.00	10.00		90.00	1058.00		1.10
Cat 1839	93.33	23.33		70.00	776.51		28.00
Cat 2050	96.67	33.33		63.34	979.71		30.00
Cat 2026	100.00	20.00		80.00	1282.00		25.00
Cat 2083A	100.00	23.33		76.67	633.71		0.00
Mean	87.94	18.86		77.08	818.00		14.00
Factors	G**	T**	G×T**	G**	G**	T**	G×T**
SEd	2.37	0.49	3.35	2.66	10.20	2.10	14.42
CD (0.05)	4.68	0.96	6.61	5.28	20.16	4.16	28.51

G : Genotype
T : Treatment
G x T: Genotype and treatment interaction
Cat: Catalog number

* : Significance level at P<0.05
** : Significance level at P<0.01
NS : Non-Significant

in Cat 1260 (476.92), Cat 1847 (249.61) and lower in Cat 1847B, Cat 411B, Cat 2034B, Cat 292, Cat 1826, Cat 1878, Cat 2070B, Cat 2057, Cat 2065, PS 1343, Cat 2090, Cat 2049, Cat 2086B, Cat 313A, Cat 1293B, Cat 2005, Cat 1995, Cat 2024, Cat 2058, Cat 1969 (0.00). This reduced growth under stress

Table 2. Effect PEG 6000 (-3 bar) on stress tolerance index, % reduction of shoot and root length over control on soybean genotypes

Genotypes	Stress tolerance index	% reduction of shoot and root length over control	Tolerance index
Cat 1987B	0.00	100.00	0.00
Cat 1809	9.75	79.12	1.00
Cat 140A	15.96	70.06	0.86
Cat 259B	9.87	66.15	1.00
Cat 1157	0.00	100.00	0.00
Cat 2047	14.97	71.18	0.86
Cat 1942	9.11	78.57	0.62
Cat 411B	0.00	100.00	0.00
Cat 1937	2.81	78.90	0.85
Cat 1912	5.06	74.68	1.00
Cat 2034B	0.00	100.00	0.00
Cat 2036	3.93	88.23	1.22
Cat 292	6.95	81.48	0.93
Cat 1826	0.00	100.00	0.00
Cat 1878	0.00	100.00	0.00
Cat 2070B	0.00	100.00	0.00
Cat 2057	0.00	100.00	0.00
Cat 1328	0.00	100.00	1.25
Cat 2065	0.00	100.00	0.00
PS 1343	0.00	100.00	0.00
Cat 2090	0.00	100.00	0.00
Cat 2049	0.00	100.00	0.00
Cat 2067	1.39	90.91	1.00
Cat 1290	4.32	68.35	0.00
Cat 2086B	0.00	100.00	0.00
Cat 313A	0.00	100.00	1.09
Cat 1293B	0.00	100.00	0.78
Cat 1091	21.51	59.12	0.00
Cat 2005	0.00	100.00	2.00
Cat 1995	0.00	100.00	0.00
Cat 1811	11.39	75.47	1.14
Cat 1847	21.61	75.06	1.00
Cat 2024	0.00	100.00	1.00
Cat 1260	27.62	70.44	1.29
Cat 2058	0.00	100.00	0.83
Cat 2722	21.08	60.46	0.00
Cat 19934	2.63	87.50	0.73
UPSL 229	6.89	87.02	1.10
Cat 120	18.57	60.22	0.92
Cat 1267B	5.41	76.82	1.00
Cat 1969	0.00	100.00	0.82
Cat 2086A	1.21	90.94	1.08
Cat 2084	0.10	98.96	0.60
Cat 1839	3.60	85.58	0.00
Cat 2050	3.41	90.11	0.00
Cat 2026	1.95	90.25	0.00
Cat 2083A	0.00	100.00	1.25
Mean	1.71	88.25	0.83
Factors	G**	G**	G**
SEd	0.20	0.36	0.019
CD (0.05)	0.40	0.71	0.038

G : Genotype * : Significance level at P<0.05 T : Treatment ** : Significance level at P<0.01
G x T: Genotype and treatment interaction NS : Non-Significant Cat: Catalog number

has been described either due to osmotic or ionic effects; inhibition of cell division and cell elongation processes associated with the growth of seedling and the decrease in plastic extensibility of the growing cell walls. Similar type of observation was given by Anaytullah *et al.,* (2008) were cumulative seedling growth (root and shoot length) decreased significantly with increasing intensity of stress irrespective of rice cultivars tested by using PEG 6000 and lead to reduction of vigour index.

Per cent reduction shoot and root growth over control

Tolerant cultivar recorded lower percent reduction of shoot and root growth over control in soybean as reported by Ange *et al.,* 2016. The interaction between genotypes and PEG concentration was highly significant. Among the genotypes lower percent reduction of shoot and root growth was observed in Cat 1091 (59.1%), Cat 1260 (70.4%), Cat 2722 (60.5%), Cat 120 (60.2%) and higher in Cat 1847B, Cat 411B, Cat 2034B, Cat 292, Cat 1826, Cat 1878, Cat 2070B, Cat 2057, Cat 2065, PS 1343,Cat 2090, Cat 2049, Cat 2086B, Cat 313A, Cat 1293B, Cat 2005, Cat 1995, Cat 2024, Cat 2058, Cat 1969 (100.0%). Reduction of shoot and root length was higher in susceptible genotypes, whereas reduction was less in the tolerant variety (Table 2). These results corroborate with earlier study of Dutta and Bera (2008) in mung bean. The reason behind that reduction of shoot length may be due to reduction of cell elongation by low water potential created by PEG. Cell elongation is mainly based on turgidity of the cell which is reduced under PEG treatment causing reduction of shoot length.

Stress tolerance index

Stress tolerance index (STI) is a more stable character and can be considered as a useful tool to screen drought tolerant genotypes (Dutta and Bera, 2008). Among the genotypes under investigation, higher stress tolerance index was recorded in Cat 1260 (27.62) and Cat 1847 (21.61) while lower in Cat 1847B, Cat 411B, Cat 2034B, Cat 292, Cat 1826, Cat 1878, Cat 2070B, Cat 2057, Cat 2065, PS 1343, Cat 2090, Cat 2049, Cat 2086B, Cat 313A, Cat 1293B, Cat 2005, Cat 1995, Cat 2024, Cat 2058, Cat 1969 (0.00) (Table 2). The high stress tolerance index of tolerant genotype might be due to higher germination percentage with higher root and shoot length in both control and stress. Reduced stress tolerance index in susceptible genotypes might be due to PEG induced osmotic effect which was deleterious and alters the plants from maintaining their proper nutritional contents necessary for their healthy growth (Krishnamurthy *et al.*, 2007).

Tolerance index

Genotypes having higher tolerance index and lower tolerance index shows tolerance and susceptibility, respectively (Dutta and Bera, 2008). The interaction between genotypes and PEG concentration was highly significant. Among the genotypes, higher tolerance index was recorded in Cat 1328 (1.25), Cat 2067 (1.25), Cat 1260 (2.00) and while lower in Cat 1847B, Cat 411B, Cat 2034B, Cat 292, Cat 1826, Cat 1878, Cat 2070B, Cat 2057, Cat 2065, PS 1343, Cat 2090, Cat 2049, Cat 2086B, Cat 313A, Cat 1293B, Cat 2005, Cat 1995, Cat 2024, Cat 2058, Cat 1969 genotypes (Table 2). Water restriction acted slowing physiological and biochemical processes and seedlings at low water deficit revealed a weak growing leading to a lower accumulation of dry matter (Marur *et al.,* 1994). Minimum reduction of dry weight of the stress seedlings result in the higher tolerance index. The results indicated tolerant genotypes maintain higher tolerance index compare to susceptible genotypes.

Conclusion

It is concluded from the experimental results that, genotypes Cat 1260 and Cat 1847 showed better performance in terms of germination percentage, vigour index, Percent reduction shoot and root growth over control, stress tolerance index, tolerance index and based on which grouped as tolerant while Cat 2084, Cat 1847B, Cat 411B, Cat 2034B, Cat 292, Cat 1826, Cat 1878, Cat 2070B, Cat 2057, Cat 2065, PS 1343, Cat 2090, Cat 2049, Cat 2086B, Cat 313A, Cat 1293B, Cat 2005, Cat 1995, Cat 2024, Cat 2058, Cat 1969 was selected as sensitive genotypes.

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