

**Original Research Paper**

## ***In vitro* screening of chilli (*Capsicum annuum* L.) genotypes for drought tolerance**

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### **ABSTRACT**

Chilli drought tolerance testing at the germination stage using osmotic polyethylene glycol (PEG) is an alternative method for conducting rapid screening in order to assemble drought tolerant varieties. Under normal and drought conditions, 16 chilli genotypes and 3 levels PEG 6000 @ 5% (-0.3 Mpa), 10% (-0.6 Mpa), 15% (-0.9 Mpa) and control were screened under *in vitro* for drought tolerance at the seeding stage in completely randomized factorial design with three replications. Among the genotypes tested, UARChH 42, UARChH 43, and Arka Swetha were found as drought-tolerant, indicating their utilization in breeding drought-tolerant genotypes and enhance cultivation in drought-prone regions.

**Keyword:** Chilli, drought tolerance, PEG treatment, screening

### **INTRODUCTION**

Chilli (*Capsicum annuum* L.) is a commercial vegetable crop belongs to the family solanaceae. Crops are typically subjected to an increased number of abiotic and biotic stress combinations as a result of global warming and potential climate abnormalities, which have a negative impact on their growth and yield (Ramegowda & Senthil-Kumar, 2015). Drought and heat have been demonstrated to be more detrimental to crop production when they occur at the same time rather than at different stages of crop growth (Prasad et al., 2011).

Drought stress significantly hampers germination and seedling growth by disrupting water uptake and metabolic processes. Recently, Singh et al. (2020) and Reed et al. (2022), highlight reduced water availability leading to delayed germination, decreased seedling vigor, and altered gene expression patterns. Additionally, findings from Ahmad et al. (2021) indicate that drought-induced oxidative stress negatively impacts seedling development. Understanding these effects is crucial for developing resilient crop varieties and sustainable agricultural practices in the face of climate change. While, withholding irrigation is a common selection method, challenges arise in consistently imposing drought conditions, especially in variable field plots. Stress bed screening, using a shared container for uniform soil

moisture, addresses this, but may not distinguish subtle genotype responses.

Another method of imparting drought is to manipulate soil osmotic potential and, as a result, plant water potential was also manipulated using solutions of known osmoticum concentration (Sikurajapathy et al., 1983). Even though there are many compounds which are effective osmotica, polyethylene glycol (PEG) has been favored by researchers. Polyethylene glycol (PEG) has the primary advantage of allowing accurate control of soil and plant water (An et al., 2006). Therefore, an experiment was carried out on *in vitro* screening of chilli genotypes for drought tolerance at the seeding stage using an osmotic solution polyethylene glycol.

### **MATERIAL AND METHODS**

The experiment comprised of 16 chilli genotypes (ACB 1, BVC 42, DKC 8, JNA 1, JNB 1, PBC 80, RAWE UP, UARChH 42, UARChH 43, Arka Kyati, Arka Haritha, Arka Meghana, Arka Swetha, Byadgi Dabbi, Raichur Local and Raichur Bullet) and 3 levels of PEG 6000 concentration @ 5% (-0.3 Mpa), 10% (-0.6 Mpa), 15% (-0.9 Mpa) and control, was laid out in completely randomized factorial design with three replications, for *in vitro* screening of chilli genotypes for drought tolerance at the seedling stage. Before germination test, seeds were sterilized with Bavistin (0.1%) using autoclaved distilled water for three hours



and washed with autoclaved distilled water three to four times for disinfection. Then seeds were immersed in autoclaved (0.1% HgCl<sub>2</sub>) for 8-10 minutes and washed with autoclaved distilled water three to four times for surface sterilization (Agogbua & Okoli, 2022). Ten seeds of each variety were transferred into sterilized petri plate with germination paper. To assess water stress tolerance during germination, 2 mL of PEG 6000 of different concentration was added to each petri dish every 3<sup>rd</sup> day and a control was maintained using distilled water for drought screening at the seedling stage. Data for germination percentage, vigor index, seedling length, root dry weight, shoot dry weight and dry seedling weight were obtained from 10 seeds in each replication. Statistical significance of means was tested by SPSS package.

## RESULTS AND DISCUSSION

Data presented in Table 1 summarizes the germination percentages of genotypes under different PEG concentrations. The genotype Arka Swetha exhibited the highest germination per cent across different concentrations, whereas, genotype PBC 80 consistently displayed lower germination per cent compared to other genotypes. At 5% PEG concentration, Arka Swetha, UARChH 42, UARChH 43 and Arka Meghana showed similar high germination per cent. Under 10% PEG concentration, UARChH 43, Arka Swetha, Arka Meghana, Arka Haritha, UARChH 42, and Arka Kyati had relatively higher germination per cent, while, PBC 80 showed the lowest. At 15% PEG concentration, UARChH 43 exhibited the highest germination, which was on par

**Table 1 : Effect of polyethylene glycol (PEG) on germination (%) of chilli genotypes**

Genotype		Germination (%)			
		PEG concentration			
		Control	5%	10%	15%
G <sub>1</sub>	ACB 1	90.00	80.00	73.33	56.67
G <sub>2</sub>	BVC 42	96.67	93.33	90.00	76.67
G <sub>3</sub>	DKC 8	93.33	76.67	73.33	46.67
G <sub>4</sub>	JNA 1	96.67	86.67	76.67	36.67
G <sub>5</sub>	JNB 1	90.00	73.33	60.00	56.67
G <sub>6</sub>	PBC 80	76.67	63.33	43.33	40.00
G <sub>7</sub>	RAWE UP	93.33	83.33	76.67	53.33
G <sub>8</sub>	UARChH 42	96.67	93.33	86.67	83.33
G <sub>9</sub>	UARChH 43	96.67	93.33	90.00	86.67
G <sub>10</sub>	Arka Kyati	96.67	90.00	86.67	73.33
G <sub>11</sub>	Arka Haritha	96.67	90.00	90.00	46.67
G <sub>12</sub>	Arka Meghana	96.67	93.33	90.00	83.33
G <sub>13</sub>	Arka Swetha	96.67	96.67	90.00	83.33
G <sub>14</sub>	Byadgi Dabbi	96.67	76.67	56.67	43.33
G <sub>15</sub>	Raichur Local	76.67	66.67	53.33	36.67
G <sub>16</sub>	Raichur Bullet	96.67	83.33	63.33	53.33
Mean		92.92	83.75	75.00	60.00
		S.E.m±		CD (0.01)	
PEG Concentration (A)		0.91		3.38	
Genotype (G)		1.83		6.75	
A x G		3.65		13.51	

with UARChH42, Arka Swetha, and Arka Meghana, while, JNA 1, Raichur Local, and PBC 80 showed the lowest germination. Increased PEG-6000 concentration led to a significant decline in germination per cent due to its low hydraulic conductivity, impeding seed water absorption. Elevated PEG levels depleted seed reserves, reducing hydrolysis and germination percentage (Aazami et al., 2010). Drought-tolerant genotypes exhibited faster germination rates, consistent with recent studies on various plants (Kumar et al., 2017; Poobalan et al., 2020).

Table 2 highlights the significant interaction between genotype and osmotic stress levels on seedling length and seedling dry weight. The genotype UARChH 42 consistently displayed the highest overall seedling length, while, JNA 1 exhibited the lowest. Maximum seedling length occurred under control conditions (5.78 cm), with UARChH 42, UARChH 43, and Arka

Swetha showing comparable lengths. Conversely, JNA 1 had the shortest seedlings under control (3.16 cm). At 5% PEG, UARChH 42 again showed the longest seedlings (6.19 cm), paralleled by UARChH 43 and Arka Swetha. At 10% PEG, UARChH 42 maintained its lead (4.36 cm), however, PBC 80 exhibited the shortest seedlings (1.46 cm). Similar trend was followed in 15% PEG where highest seedling length was observed in UARChH 43. Reduced seedling length under stress is attributed to decreased cell elongation from low water potential induced by PEG, consistent with recent chilli and tomato studies (Garg et al., 2019; Kumar et al., 2017; Poobalan et al., 2020).

Significant interaction effects between genotypes and moisture stress levels influenced seedling biomass. Under control, UARChH 42, UARChH 43, and Arka Swetha displayed similar high biomass, contrasting with JNA 1, which had the lowest. At 5% PEG, similar

**Table 2 : Effect of polyethylene glycol on seedling length and dry weight of chilli genotypes**

Genotype		Seedling length (cm)				Five seedlings dry weight (mg)			
		PEG concentration				PEG concentration			
		Control	5%	10%	15%	Control	5%	10%	15%
G <sub>1</sub>	ACB 1	4.12	3.19	2.41	1.28	24.25	20.30	16.15	8.15
G <sub>2</sub>	BVC 42	5.76	4.49	3.58	2.45	37.67	29.99	22.60	13.89
G <sub>3</sub>	DKC 8	4.87	3.49	2.78	1.26	20.79	15.43	11.70	7.11
G <sub>4</sub>	JNA 1	3.16	2.14	1.61	0.54	18.53	11.92	9.55	3.71
G <sub>5</sub>	JNB 1	3.88	3.05	2.51	1.25	22.60	19.10	17.01	9.86
G <sub>6</sub>	PBC 80	3.29	2.40	1.46	0.83	19.04	13.17	9.17	5.71
G <sub>7</sub>	RAWA UP	5.57	4.70	2.88	1.07	33.99	30.10	19.11	6.85
G <sub>8</sub>	UARChH 42	7.96	6.19	4.36	3.18	47.57	38.49	24.37	16.85
G <sub>9</sub>	UARChH 43	7.93	6.15	4.34	3.15	47.53	38.32	24.36	16.84
G <sub>10</sub>	Arka Kyati	6.47	5.18	3.84	1.99	41.13	35.15	22.80	9.91
G <sub>11</sub>	Arka Haritha	6.48	4.36	3.17	2.36	40.44	22.90	16.71	9.59
G <sub>12</sub>	Arka Meghana	6.78	5.07	4.17	2.27	37.30	28.09	23.45	11.32
G <sub>13</sub>	Arka Swetha	7.91	6.14	4.33	3.15	47.45	38.28	24.35	16.83
G <sub>14</sub>	Byadgi Dabbi	5.66	3.87	3.14	2.60	31.49	23.84	19.59	13.82
G <sub>15</sub>	Raichur Local	5.79	2.79	2.25	1.34	41.26	18.04	15.55	9.01
G <sub>16</sub>	Raichur Bullet	6.96	4.78	3.29	1.78	43.92	32.20	21.24	10.15
Mean		5.78	4.25	3.13	1.91	34.68	25.54	18.61	10.60
		S.E.m±		CD (0.01)		S.E.m±		CD (0.01)	
PEG Concentration (A)		0.018		0.069		0.127		0.235	
Genotypes (G)		0.037		0.139		0.127		0.471	
A x G		0.076		0.281		0.254		0.942	

trends persisted. UARChH 42 consistently exhibited maximum biomass across stress levels, paralleled by UARChH 43 and Arka Swetha. Conversely, JNA 1 consistently displayed the lowest biomass across all stress levels. At 10% PEG, UARChH 42, UARChH 43, and Arka Swetha again showed maximum biomass, while, PBC 80 exhibited the minimum. At 15% PEG, similar patterns persisted.

Data presented in Table 3 indicated decrease in seedling length and seedling biomass across osmotic stress and genotypes. Under 5% PEG, UARChH 42, Arka Swetha, and UARChH 43 showed the least seedling length reduction (20.26%, 20.39%, and 21.04%, respectively), while, Raichur Local experienced the highest (51.87%). At 10% PEG, Arka Swetha, UARChH 43, and UARChH 42 exhibited minimal reduction (43.34%, 43.65%, and 44.05%,

respectively), contrasting with Raichur Local with maximum reduction (61.08%). At 15% PEG, Arka Swetha, UARChH 43, and UARChH 42 again had the least reduction (58.83%, 58.92%, and 59.17%, respectively), whereas PBC 80 and Raichur Local had the highest (76.24% and 76.80%, respectively). Root and shoot lengths serve as key indicators for drought resistance screening (Bayoumi et al., 2008). Reduced lengths may result from obstacles to cell division and elongation, allowing plants to cope with stress (Anjum et al., 2011). Findings align with recent studies on tomato and chilli (Mollah et al., 2021).

Under 5% PEG, UARChH 42, Arka Swetha, and UARChH 43 had the least seedling biomass reduction (16.55%, 17.02%, and 17.76%, respectively), while, Raichur Local showed the highest (56.29%). At 10% PEG, UARChH 42, UARChH 43, and RAWE UP had

**Table 3 : Effect of polyethylene glycol concentrations on percentage decrease in seedling length and biomass in chilli genotypes**

Genotype		Decrease (%)					
		Seedling length			Seedling biomass		
		PEG concentration			PEG concentration		
		5%	10%	15%	5%	10%	15%
G <sub>1</sub>	ACB 1	22.71	41.47	68.96	24.27	39.76	69.61
G <sub>2</sub>	BVC 42	22.03	47.83	59.43	20.40	40.00	67.13
G <sub>3</sub>	DKC 8	28.39	43.02	74.21	25.77	46.73	65.82
G <sub>4</sub>	JNA 1	32.45	49.11	82.82	35.68	48.44	79.99
G <sub>5</sub>	JNB 1	24.00	37.38	68.77	20.76	29.43	59.10
G <sub>6</sub>	PBC 80	31.29	58.30	76.24	30.83	51.85	70.02
G <sub>7</sub>	RAWE UP	33.89	48.20	80.84	20.28	47.80	79.86
G <sub>8</sub>	UARChH 42	20.26	44.05	59.17	16.55	43.27	63.43
G <sub>9</sub>	UARChH 43	21.04	43.64	58.92	17.76	43.65	63.08
G <sub>10</sub>	Arka Kyati	23.93	45.63	69.26	20.61	48.56	75.90
G <sub>11</sub>	Arka Haritha	32.77	51.03	63.61	43.38	58.70	76.30
G <sub>12</sub>	Arka Meghana	24.43	38.54	66.52	24.68	47.13	69.64
G <sub>13</sub>	Arka Swetha	20.39	43.34	58.83	17.02	46.57	63.51
G <sub>14</sub>	Byadgi Dabbi	30.85	43.67	52.61	24.30	47.79	66.10
G <sub>15</sub>	Raichur Local	51.87	61.08	76.80	56.29	62.31	78.17
G <sub>16</sub>	Raichur Bullet	31.29	52.75	74.36	26.67	51.63	76.89
Mean		28.22	46.81	68.08	26.67	47.11	70.28
		S.E.m ±		CD (0.01)	S.E.m ±		CD (0.01)
PEG concentration (A)		0.36		1.34	0.18		0.71
Genotype (G)		0.72		2.68	0.37		1.41
A x G		1.44		5.36	0.75		2.80

minimal reduction (43.27%, 43.65%, and 43.80%, respectively), contrasting with Raichur Local recorded maximum reduction (62.31%). At 15% PEG, UARChH 43, UARChH 42, and Arka Swetha had the least reduction (63.08%, 63.43%, and 63.51%, respectively), whereas JNA 1 showed the highest (79.99%). Significant variations were evident in per cent decrease in seedling biomass across different osmotic stress levels and genotypes. Stress-induced reduction in seedling dry weight across genotypes is supported by studies (Wani et al., 2010; Tyagi, 2010). Low water uptake and restricted metabolic activities under stress contribute to decreased seedling biomass.

### CONCLUSION

The study revealed that drought stress adversely affects chilli germination and seedling growth, likely due to stomatal closure, hindering carbon dioxide intake and photosynthesis. This stress triggers reactive oxygen species accumulation and alters hormone levels, particularly abscisic acid, prioritizing water conservation. Germination and growth traits declined with increasing drought severity. Among the 16 genotypes tested, UARChH 42, UARChH 43 and Arka Swetha were found as drought-tolerant. Utilizing drought-tolerant genotypes in breeding programs can lead to superior hybrids and enhance cultivation in drought-prone regions, offering promising solutions for agricultural resilience.

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