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Identification of potential pearl millet genotypes for early drought tolerance using PEG 6000

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Abstract
Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is an important staple millet for supplying food security in the world's dry regions. Despite its natural habitat for drought resistance, there is a considerable loss in plant stand because of water stress induced during the development period. In the present study, total of 40 genotypes have been studied for drought tolerance at seedling stages with varying levels of osmotic stress, *viz*., -3, -5, -7.5, and -10 bar using PEG 6000. Under increasing osmotic stress, a substantial decrease in germination%, shoot length, root length, and seedling vigour index was observed. The root shoot ratio had increased with the varying levels of treatments. Osmotic stress at -7.5 bar was highly relevant to evaluating drought tolerance in pearl millet, and genotypes Among the different genotypes screened for drought tolerance, the parental line such as PT5456, PT5721, PT5748, PT6303, PT7035, seed parent ICMB 99222, and landraces such as *Cumbu1, Cumbu2,* and *Nattu cumbu* had showed better performance with varying level of osmotic stress under laboratory conditions. These genotypes could be utilized for hybridization programme after assessing under field performance.

Keywords: Pearl millet, drought tolerance, PEG-6000, osmotic level, seedling vigour index

Introduction
Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is the 5th most important cereal crop in the world after rice, wheat, maize and sorghum and in India, it is the fourth most widely cultivated food crop after rice, wheat and maize. It is grown on more than 30 million acres globally, with Africa (>18 million ha) and Asia (>10 million ha) having the largest crop areas. It is used as a staple diet for humans as well as fodder and feed in the livestock industry. It grows quickly with minimal inputs, has a high photosynthetic efficiency, has established excellent, healthy, nutritious characteristics, and is tolerant to harsh climatic conditions and biotic stresses. It can withstand challenging environmental conditions such as low soil fertility, high soil pH, high soil Al^{3+} saturation, low soil moisture, high temperature, high soil salinity, and limited rainfall. In India, Pearl millet is cultivated in 7.55 Mha. area with a production of 9.74 million tonnes during 2022-23 with an average yield 1374 kg per (Source: Ministry of Agriculture & Farmers Welfare (MoA&FW)).

Drought studies in pearl millet have been carried out at each of the three major stages: vegetative, panicle growth, and grain filling and indicated that resistance mechanisms involving drought avoidance, tolerance, escape, and recovery (Shivhare *et al*., 2020) [38]. Drought stress is one of the major abiotic stresses that affect crop production worldwide, which is characterised by reduction of water content, decline in leaf water potential and turgor loss, stomatal closure, and decrease in cell enlargement and growth and other physiological processes like activation of reactive oxygen species and lipid peroxidation (Ghatak *et al*., 2016) [39]. Better germination percentage and better growth parameters at early seedling growth stages can be considered important traits for screening pearl millet genotypes for drought tolerance (Shivhare *et al*., 2019) [3]. Polyethylene glycol (PEG)-6000, an inert and non-ionic stress that can form almost impermeable chains to preserve uniform water potential, is a commonly used stress for this purpose (Sinhababu and Kar 2003; Basu *et al*., 2010) [16, 17]. Though the moisture stress induced by PEG-6000 is more rapid in triggering an osmotic shock, the physiological and biochemical reactions of plants to such treatments would indicate the comparative ability of different genotypes to survive drought stress at the seedling stage (Shivhare *et al*., 2019) [3]. The present research aimed to determine the influence of polyethylene glycol on root and shoot traits in seedling of pearl millet genotypes, and identification of potential genotypes for drought tolerance.

Materials and Methods

The experimental material consists of 40 genotypes including restorer lines, maintainer lines, landraces, and released hybrids and varieties were subjected to drought tolerance trials using PEG 6000 in the Department of Millets, Tamil Nadu Agricultural University (TNAU), Coimbatore. The experiment was conducted by enforcing different levels of osmotic stress using different concentrations of PEG 6000 with two replications under completely randomized design.

Mode of action of Polyethylene Glycol (PEG)

Polyethylene glycol (PEG) is a water-soluble, non-ionic natural polymer with higher molecular weight of 6000. PEG 6000 has been proven to imitate drought stress, resulting in a decrease in plant water potential due to osmotic stress under *in-vitro* conditions.

Table 1: PEG-6000 osmotic solution concentrations.

Water Potential	PEG 6000 Concentration
Control	distilled water
-3 bar	11.50%
-5 bar	19.60%
-7.5 bar	23.50%
-10 bar	33%

The seeds were treated with five different treatments for examination of pearl millet genotypes at early seedling growth stages, namely control with double distilled water (T1) and -3 bar (T2), -5 bar (T3), -7.5 bar (T4), and -10 bar (T5) PEG-6000 solution at 25 ±2 °C with two replications in factorial experiment including genotype. The seeds were placed in 90 mm petri dishes with two layers of germination paper. 10 ml treatment solution or distilled water was put over the germination paper, and then solution was poured over it as needed. When the radicle and plumule of a seed grew for at least 2 mm, it was considered as germinated. The physiological parameters were examined 10 days after incubation. After 10 days of germination, all physiological parameters were measured including germination %, shoot length (cm), root length (cm), root/shoot ratio, and seed vigour index (SVI). SVI was estimated by (Baloch *et al*., 2012) [18] method. Two way analysis of variance (ANOVA) was performed to determine significant differences and then the average values were compared using GRAPES software.

SVI= (Root length+Shoot length)×Germination Percentage

Result and Discussion

The analysis of variance revealed a significant difference among each seedling characteristics associated with pearl millet under different osmotic stress levels in the current study. According to Manga (1998) [19], Patil (2010) [25] and Surendhar *et al*., (2020) [2], osmotic stress at -7.5 bar was highly relevant to evaluating drought tolerance in pearl millet, and genotypes that performed better at this stress level were chosen for further field screening. At -10 bar, there was less seedling growth so observation could not taken. PEG 6000 showed that the effect of genotype, treatment and the interaction between genotype and treatment. All treatments were significant for germination percentage, root length, shoot length and seedling vigour index except root shoot ratio.

Impact of PEG on germination percentage

The lack of water in the soil has a detrimental impact on seed germination and seedling development. PEG-induced water stress at various levels had significant impacts on seed germination. Among the 40 genotypes evaluated at various concentrations, there was a reduction in germination percentage with an increase in osmotic potential observed. It is probably due to the low hydraulic conductivity of the environment, where PEG 6000 makes water unavailable to seeds, affecting the imbibitions process of the seed, which is essential for germination (Lobato *et al.*, 2008) [20]. Significant difference were observed among the treatments. In controlled condition, the range of germination % was from 60 to 100%. Germination % decreases from 100% to 20% in different conditions. At -3 bar osmotic potential, germination % varies from 60% to 100% with mean value 81%. 17 genotypes had more germination percentage than mean value. Among them, 14 genotypes had more than 90% germination and 20 genotypes had more than 70% germination and 6 genotypes had more than 60% germination. Among the genotypes, ICMB99222 (100%) had recorded highest germination percentage and PT6693 (60%) had lowest germination%. At -5 bar osmotic potential, germination % varies from 50% to 90% with mean of 73%.18 genotypes had more germination percentage than mean value. Among them,13 genotypes had more than 80% germination and 15 genotypes had more than 60% germination and 12 genotypes had more than 50% germination. Among the genotypes, PT5748 (90%) had observed highest germination percentage in contrary PT6674, PT6679, ICMB1508 (50%) had lowest germination %. At -7.5 bar osmotic potential, germination % varies from 20% to 75% with mean of 53%. 25 genotypes have more germination percentage than mean value. Among them,20 genotypes had more than 60% germination and 15 genotypes had more than 40% germination and 5 genotypes had more than 15% germination. Among the genotypes, PT5748 (75%) had observed highest germination percentage and PT6679, ICMB99111 (20%) had lowest germination%. Among the genotypes evaluated in different concentrations, the genotypes *viz.*, *Nattu cumbu*, PT5748, *Cumbu1*, have high germination percentage and indicated drought-tolerance under different stress condition. In contrary, the genotypes *viz.*, PT6679, and ICMB1508 show lower germination percentages, which are considered as susceptible. The variation in germination percentage has also been documented in maize (Parmar and More, 1966; Farsiani and Ghobadi, 2009) [21, 22], sorghum (Saint-Clair, 1976) [23], rice (Goswami and Baruah, 1994), and pearl millet (Govindaraj *et al.*, 2010; Sani and Boureima, 2014) [1, 13].

Impact of PEG on root length (cm)

Root is the principal component which is immediately exposed to osmotic stress. The important characteristic of drought resistance is early and fast root elongation (Kulkarne and Deshpande, 2007) [24]. In the current study, increasing external water potential induced a substantial decrease in root length, and all treatments caused a decrease in root elongation in all genotypes compared to their controls. Significant difference were observed among the treatments. Root length under controlled condition was varied from 6.8 to 14.3 cm. Under different condition, there was a reduction in root length from 14.3 cm to 2.1 cm At -3 bar osmotic potential, root length varies from 2.4 to 14 cm with mean value 8.016 cm. 22 genotypes had more root length than mean value. Among them, 21 genotypes had more than 8 cm root length and 12 genotypes had more than 6 cm root length and 7 genotypes had more than 2 cm root length. Among the genotypes, PT5748 (14 cm) had recorded highest root length and ICMB1508 (2.4 cm) had lowest root length. At -5 bar osmotic potential, root length varies from 2.7 to 10.1 cm with mean value 6.94 cm. 21 genotypes had more root length than mean value. Among them, 17 genotypes had more than 7 cm root length and 15 genotypes had more than 5 cm root length and 8 genotypes had more than 2 cm root length. Among the genotypes, PT5456 (10.1 cm) had recorded highest root length and ICMB1508 (2.7 cm) had lowest root length. At -7.5 bar osmotic potential, root length varies from 2.2 to 5.9 cm with mean value 4.09 cm. 21 genotypes had more root length than mean value. Among them, 11 genotypes had more than 5 cm root length and 19 genotypes had more than 3 cm root length and 10 genotypes had more than 2 cm root length. Among the genotypes, PT5721 (5.9 cm) had recorded highest root length and PT6067 (2.1 cm) had lowest root length. Among the genotypes evaluated in different concentrations, the genotypes *viz*., PT5748, PT5721, PT5456, PT7035, *Cumbu 1* have high root length and indicated drought-tolerance under different stress condition. In contrary, the genotypes *viz.*, PT6067 and ICMB1508 show lower root length, which are considered as susceptible. Jajarmi *et al.*, (2009), Whalley *et al*., (1998) [12], and Govindaraj *et al*., (2010) [1] all showed a decrease in root length at high concentration. Walter (1963) [29], Radhouane (2007) [5], and Kulkarni and Desphpande (2007) [24], have reported similar outcomes. According to Saxena and Toole (2002), the genetic material that performs better under stress is drought resistant in nature, Hence PT5721, PT5748, PT5456, PT7035, *Cumbu 1* could be considered for further breeding programmes. Seedlings with longer roots may be able to perceive and cope with water-deficiency conditions than shorter roots seedling (Khodarahmpour 2011) [40].

Impact of PEG on shoot length(cm)

Shoot length was shown to decrease as the osmotic stress condition increased by (Patil, 2010) [25]. Due to decreased cell division and elongation, shoot length decreased (Kramer, 1983) [26]. The reduction in shoot and root length caused by due to cell division and elongation, resulting in tuberization. This tuberization and lignification of the root system allows the plant to enter a dormant state while waiting for conditions to improve (Fraser *et al.*, 1990) [27]. Drought affects shoot length more than root length; however, the symptoms and effects of drought are more visible on the shoot and aerial sections of the plant, which bear the majority of the economic components of field crops in field circumstances. When the external osmotic potential was increased, the shoot length decreased. Significant difference were observed among the treatments. Among 40 genotypes, the shoot length differed from 5 to 11.7 cm with grand mean value of 8.014 in controlled condition. At -3 bar osmotic potential, shoot length varies from 2.6 to 8.9 cm with mean value 5.887cm. 21 genotypes had more shoot length than mean value. Among them, 20 genotypes had more than 6 cm shoot length and 11 genotypes had more than 5 cm shoot length and 9 genotypes had more than 2 cm shoot length. Among the genotypes, *Nattu cumbu1* (8.9 cm) had recorded highest shoot length and ICMB1508 (2.6 cm) had lowest shoot length. At -5 bar osmotic potential, shoot length varies from 2.7 to 6.3 cm with mean value 4.554 cm. 22 genotypes had more shoot length than mean value. Among them, 16 genotypes had more than 5 cm shoot length and 12 genotypes had more than 4 cm shoot length and 12 genotypes had more than 2 cm shoot length. Among the genotypes, PT6303 (6.3 cm) had recorded highest shoot length and *Shoolagiri local* (2.7 cm) had lowest shoot length. At -7.5 bar osmotic potential, shoot length varies from 1.1 to 4.9 cm with mean value 3.12 cm. 20 genotypes had more shoot length than mean value. Among them, 13 genotypes had more than 4 cm shoot length and 13 genotypes had more than 2.5 cm shoot length and 14 genotypes had more than 1 cm shoot length. Among the genotypes, *Cumbu 1*, COH10 (4.9 cm) had recorded highest shoot length and ICMB99555 (1.1 cm) had lowest shoot length. Among the genotypes evaluated in different concentrations, the genotypes *viz*., PT5456, PT6303, *Cumbu 1*, COH10 have high shoot length and indicated drought-tolerance under different stress condition. In contrary, the genotypes *viz.*, ICMB99555 and ICMB1508 show lower shoot length, which are considered as susceptible. Lawlor (1969) [28] also observed the slowdown of shoot and root length growth in response to increased osmotic stress in both the field and the laboratory. Above results also correlated with previous research (Walter 1963; Parmer and Moore 1966; Radhouane, 2007; and Kulkarni and Desphpande, 2007) [29, 5, 30, 24]. It is widely acknowledged that when exposed to external osmotic potential, the roots suffer first, followed by their attributed plant parts (Misra and Dwivedi, 2004). Shoot length was reduced in all genotypes at early seedling growth stages, with more severe impacts which is in accordance to those reported in sorghum (Bibi *et al*., 2010) [31] and pea (Okçu *et al*., 2005) [32].

Impact of PEG on seedling vigour index (SVI)

Early seedling vigour can improve agricultural water usage efficiency (Siddique *et al.*, 1990) [33]. Several studies indicated that greater growth under stress conditions could be used to enhance drought-resistant genotypes (Abdel-Raheem *et al*., 2007). As a result, selecting drought-tolerant genotypes based on a high seedling vigour index under increased osmotic stress would be beneficial (Surendar *et al.*, 2019) [3]. A decrease in seedling vigour index might be related to a decrease in seed germination and seedling growth. Significant difference were observed among the treatments. Under controlled condition, the range differed from 2415 to 984.5. At -3 bar osmotic potential, SVI varies from 395.5 to 1896.3 with mean value 1138.51. Among them, 20 genotypes had more than 1200 SVI and 9 genotypes had more than 900 SVI and 11 genotypes had more than 300 SVI. Among the genotypes, *Nattu cumbu1* (1896.3) had recorded highest SVI and ICMB1508 (395.5) had lowest SVI. 21 genotypes had more SVI than mean value. At -5 bar osmotic potential, SVI varies from 286.6 to 1291.1 with mean value 815.65. Among them, 16 genotypes had more than 900 SVI and 14 genotypes had more than 600 SVI and 10 genotypes had more than 200 SVI. Among the genotypes, PT5748 (1291.1) had recorded highest SVI and PT6679 (286.6) had lowest SVI. 21 genotypes had more SVI than mean value. At -7.5 bar osmotic potential, SVI varies from 111.7 to 719.5 with mean value 418.96. 19 genotypes had more SVI than mean value. Among them, 15 genotypes had more than 500 SVI and 15 genotypes had more than 250 SVI and 10 genotypes had more than 100 SVI. Among the genotypes, PT5748 (719.5) had recorded highest SVI and PT6679 (111.7) had lowest SVI. Among the genotypes evaluated in different concentrations, the genotypes *viz*., PT5748, *Nattu cumbu*, *Cumbu 1* and PT6303 have high SVI and indicated drought-tolerance under different stress condition. In contrary, the genotypes *viz.*, PT6679 and ICMB1508 show lower SVI, which are considered as susceptible. The higher SVI was observed in different genotypes by combination of high germination, root length, and shoot length. Das and Kalika (2010) [41] reported a decrease in seedling vigour index of rice cultivars when PEG concentration increased and decrease in seedling vigour index could be related to a decrease in seed germination and seedling growth. Seed vigour index (SVI) and activities of the various characters under drought stress were confirmed by various workers (Jana, 1975; Acevedo, 1991) [34, 35].

Upon considering all these parameters, parental lines such as PT5456, PT5721, PT5748, PT6303, PT7035, seed parent ICMB 99222, and landraces such as *Cumbu1*, *Cumbu2*, *Nattu cumbu*, and Checks COH10 are highly drought tolerant among the 50 genotypes. PEG-induced osmotic stress had a significant impact on seed germination percentage as well as many early seedling parameters *viz*., root length, shoot length and SVI. The results supported previous findings in lettuce (Nasri *et al*., 2011) [42], wheat (Baloch *et al*., 2012) [18], and sorghum (Bibi *et al*., 2010) [31]. Seed germination and seed vigour index have been reported to be positively correlated, indicating a relationship between these characters (Gupta *et al*., 2014) [36]. SVI is positively correlated with all seedling parameters, so drought tolerant genotype was selected based on SVI.

Screening of stress-tolerant types of agricultural crops is required to create greater stress tolerance through molecular breeding and transgenic techniques. Plant breeders might use and exploit the differential response of genotypes to PEG-induced osmotic stress at the seedling stage to find drought tolerant cultivars before conducting extensive field experiments. These findings are consistent with previous research, which found that *in-vitro* screening with PEG is one of the reliable ways for selecting appropriate genotypes to explore in depth on water shortage on plant germination indices (Kocheva *et al*., 2003) [37].

Table 2: Mean performance of pearl millet genotypes on seedling traits under PEG induces osmotic stress conditions.

Genotypes	Germination (%)				Root Length (cm)				Shoot Length (cm)				Seedling Vigour Index			
	0	-3	-5	-7.5	0	-3	-5	-7.5	0	-3	-5	-7.5	0	-3	-5	-7.5
PT5456	100*	85	75	65	13.7	13.5	10.1*	5.1	8.6	5.8	5.0	4.4	2232.4	1635.0	1121.5	619.8
PT5721	95	95	80	70	12.1	11.1	8.7	5.9*	8.4	7.5	6.2	4.0	1950.3	1769.3	1187.2	696.9
PT5748	90	85	90*	75*	12.4	14.0*	8.4	5.8	7.3	7.1	5.9	3.8	1784.2	1791.0	1291.1*	719.5*
PT6029	100*	90	85	60	9.5	8.1	6.0	4.6	7.9	5.7	4.6	3.7	1737.5	1243.8	900.2	495.0
PT6067	100*	95	85	40	7.6	5.7	5.0	2.2	9.7	5.4	4.0	2.3	1723.3	1052.4	773.9	180.7
PT6303	95	95	85	65	13.1	9.3	7.8	5.6	9.3	8.0	6.3*	4.5	2126.9	1639.9	1196.6	657.8
PT6317	80	65	60	50	7.8	3.5	4.1	3.4	7.3	3.5	3.6	3.0	1210.5	446.0	461.5	320.0
PT6475	90	80	65	45	11.0	4.7	4.9	4.1	9.0	5.2	4.7	2.7	1797.8	794.3	621.6	293.3
PT6476	85	65	65	35	9.2	7.0	5.3	3.0	5.7	4.1	3.2	1.9	1264.5	724.0	551.1	169.5
PT6674	75	65	50	45	10.4	8.2	5.0	3.0	6.2	5.1	3.9	2.0	1251.2	865.5	467.8	221.4
PT6679	85	65	50	20	9.2	5.6	2.8	2.6	11.7*	5.3	3.0	2.7	1779.5	706.7	286.6	111.7
PT6680	95	85	75	65	10.4	8.8	8.4	5.8	7.2	6.8	5.6	3.8	1664.5	1325.9	1052.4	628.4
PT6686	95	80	75	65	12.1	10.9	9.3	5.8	9.1	6.5	5.1	4.3	2023.1	1390.0	1085.9	647.7
PT6687	85	65	65	35	10.7	6.3	5.1	3.3	9.2	6.0	4.2	2.6	1699.8	800.9	595.0	204.5
PT6693	90	60	85	55	12.9	6.3	5.9	3.1	10.0	6.0	4.5	2.2	2054.5	735.0	887.5	295.0
PPT6708	95	75	70	35	9.5	7.4	6.4	2.7	11.4	6.6	4.2	1.8	1984.3	1056.0	739.3	159.3
PT6752	80	70	55	60	12.0	12.3	6.6	5.1	9.1	6.8	4.5	4.1	1683.3	1341.0	595.9	552.9
PT7035	100*	80	80	70	11.8	8.6	8.0	5.8	8.9	6.5	5.9	4.4	2060.0	1208.8	1110.4	718.6
PT7037	65	70	60	30	9.4	6.6	6.3	3.0	5.8	4.6	3.2	1.9	984.5	788.6	568.5	145.5
PT7043	95	80	55	50	10.7	10.2	7.1	3.1	6.4	5.7	4.0	2.7	1627.4	1268.8	630.9	287.5
ICMB93111	95	85	80	60	11.1	8.4	7.5	4.0	9.7	6.0	5.3	3.6	1984.2	1226.4	1024.8	458.7
ICMB98222	90	90	70	45	8.3	6.7	5.9	3.0	5.0	4.3	3.6	2.4	1198.8	987.8	664.3	241.3
ICMB99222	95	100*	75	65	9.7	5.8	3.7	4.6	6.1	4.1	2.9	3.1	1501.0	988.5	494.0	499.0
ICMB99555	60	65	75	40	13.3	9.0	8.1	2.3	7.2	5.7	3.4	1.1	1249.5	966.5	850.5	134.5
ICMB00555	100*	80	65	50	7.9	7.7	5.7	3.2	7.9	7.4	5.6	2.5	1580.0	1204.7	741.0	286.3
ICMB02777	95	90	80	65	11.3	8.8	7.7	4.0	6.8	6.2	5.5	3.4	1717.0	1356.2	1059.6	479.0
ICMB04111	85	75	65	60	8.2	6.5	5.7	3.6	6.7	5.3	4.2	3.0	1264.3	882.3	647.6	398.1
ICMB06111	100*	80	75	60	11.0	8.3	7.1	3.6	8.6	7.3	5.3	3.1	1962.0	1243.2	935.8	404.4
ICMB10444	85	75	70	40	7.2	4.3	7.2	2.8	5.6	4.3	5.5	2.4	1088.0	644.7	889.5	208.0
ICMB1508	70	80	50	50	6.8	2.4	2.7	2.8	7.1	2.6	2.9	2.4	991.0	395.5	292.7	258.1
Cumbu 1	100*	85	75	70	14.3*	8.7	8.3	5.6	8.7	7.0	6.3	4.9*	2303.5	1337.5	1095.7	676.0
Cumbu 2	100*	90	70	65	11.8	8.8	6.8	4.9	9.2	6.9	5.7	4.4	2100.0	1411.2	871.0	598.9
Kizhikuppam Local	95	85	75	65	10.7	7.2	6.0	4.7	10.5	6.4	5.0	4.3	2003.1	1164.4	829.5	584.0
Nattu cumbu	100	95	85	70	13.9	11.1	8.7	5.4	10.3	8.9*	6.0	4.1	2415.0*	1896.3*	1256.9	666.8
Shoolagiri Local	85	90	75	50	10.7	6.6	4.9	3.2	7.0	4.2	2.7	2.0	1485.9	987.7	578.8	258.0
Uthangarai Local	70	90	85	65	10.9	8.3	6.5	4.6	8.0	7.7	4.8	4.1	1372.0	1435.7	965.9	612.4
86M38	95	80	70	40	9.3	7.0	5.6	4.1	5.8	4.5	3.2	2.5	1431.5	925.2	616.0	263.4
CO10	90	90	85	65	12.6	10.7	7.0	4.9	8.1	6.6	4.4	4.5	1859.1	1566.5	958.5	611.0
COH10	95	90	85	65	13.2	9.1	8.1	5.7	7.9	5.2	4.5	4.9	2007.3	1289.3	1081.8	686.0
Dhanasakthi	95	85	70	45	9.5	7.4	5.6	3.6	6.4	5.0	3.7	2.5	1510.5	1048.8	647.5	275.8

S.Ed (G)	3.428	0.467	0.32	70.849
S.Ed (T)	1.212	0.148	0.101	22.404
S.Ed (G*T)	7.665	1.045	0.716	158.422
CD 5% (G)	6.759	0.92	0.631	139.536
CD 5% (T)	2.39	0.291	0.2	44.125
CD 5% (G*T)	15.114	2.058	1.411	312.012

Table 3: Two factor Analysis of variance for seedling traits of pearl millet under different PEG 6000 treatments.

Source	df	Mean Square			
		Germination (%)	Root Length	Shoot Length	Seedling Vigour Index
Genotype	39	843.788**	22.219**	8.148**	488930.66**
Treatment	4	68989.750**	963.699**	575.924**	32005666.94**
G*T	156	123.212**	2.500**	1.526**	67920.71**
Error	200	58.750	1.092	0.513	26962.89

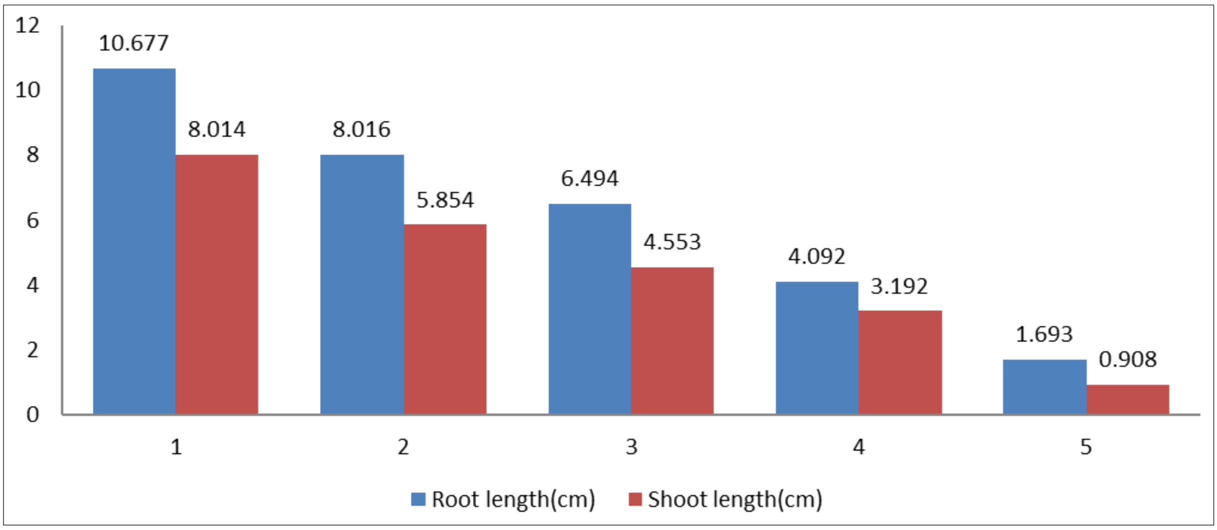

Fig 1: Effect of PEG on germination percentage among pearl millet genotypes

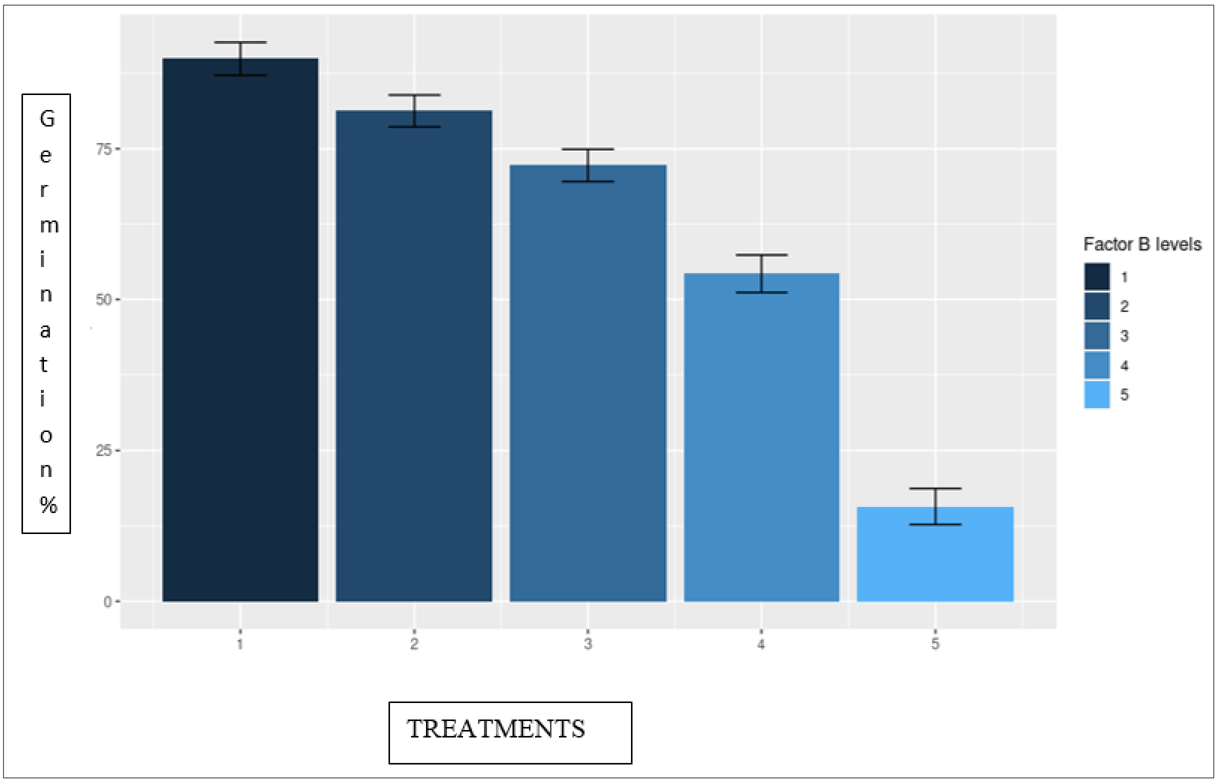

Fig 2: Performance of pearl millet genotypes and growth attributes under varying level of osmotic pressure.

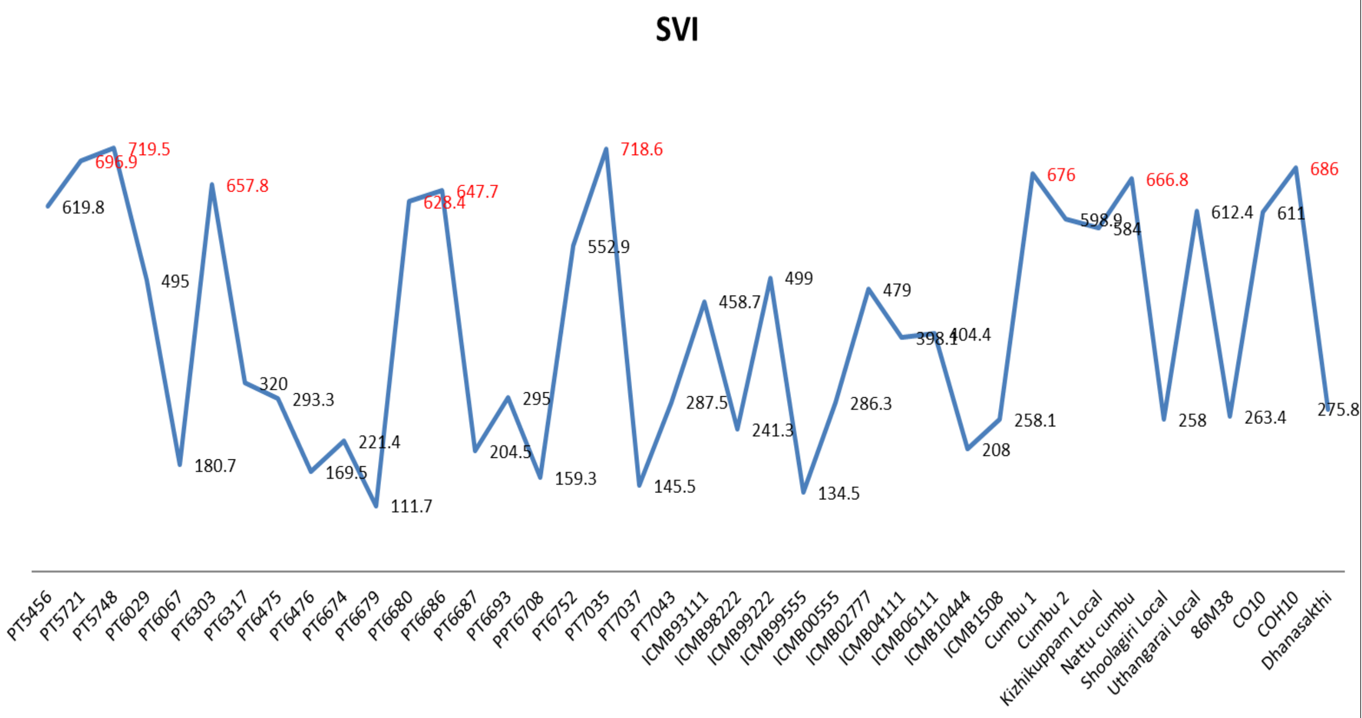

Fig 3: Effect of PEG on seedling vigor index of different genotypes under -7.5 bar osmotic stress

Control -3 bar -5bar -7.5bar -10bar

Fig 4: PEG screening under laboratory condition with different concentration

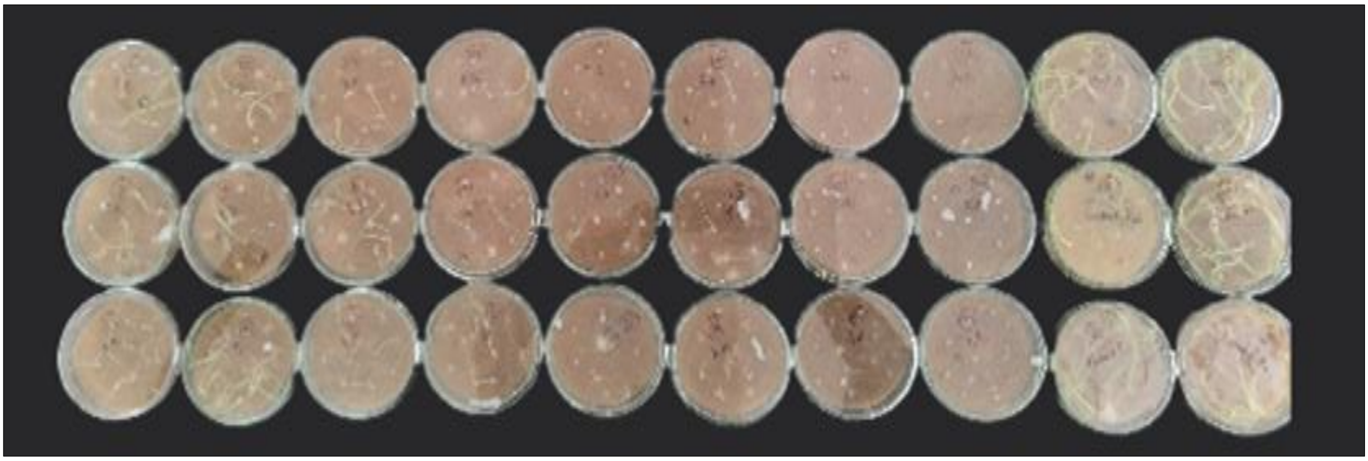

Fig 5: PEG screening

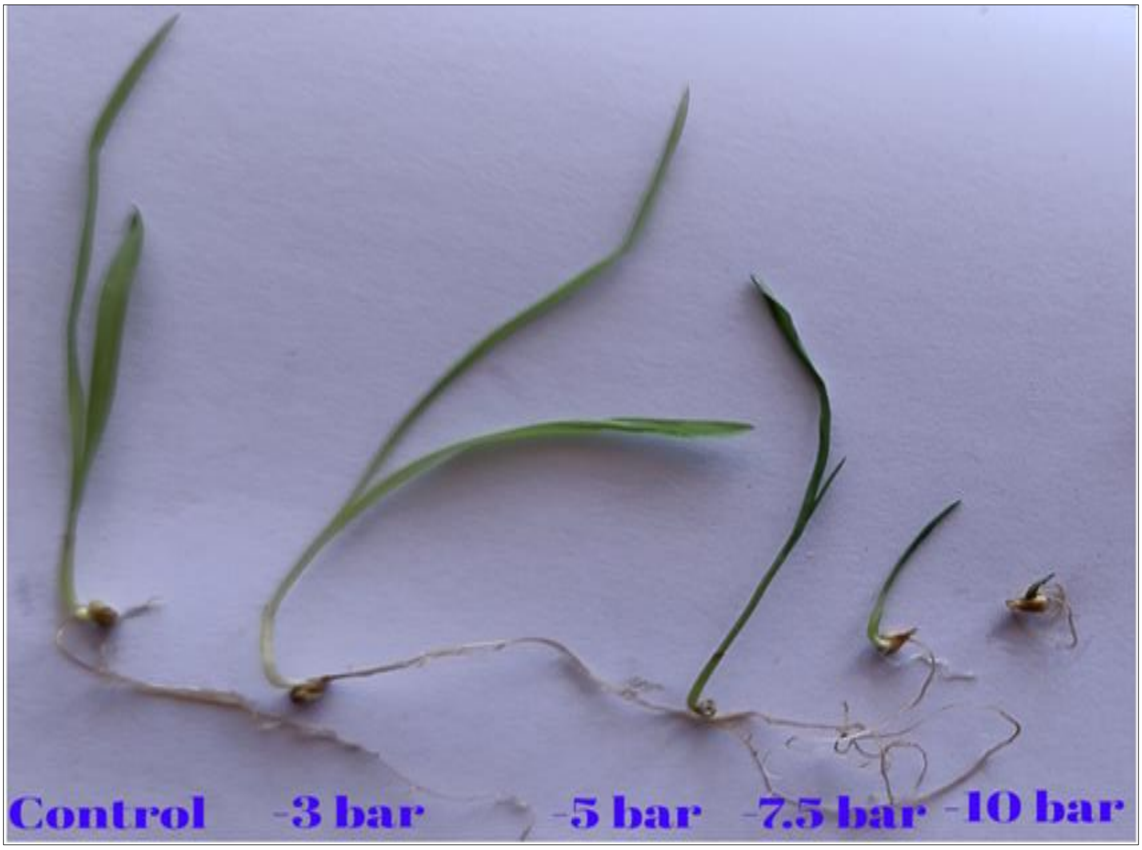

Fig 6: Effect of osmotic pressure/drought on seedling growth of pearl millet genotypes

Conclusion

The best and most cost-effective approach for selecting drought-tolerant genotypes from a wide range of collections is polyethylene glycol (PEG 6000)-mediated screening. Better germination percentage and early seedling development characteristics might be considered key attributes when screening pearl millet genotypes for drought resistance. Plant breeders can use the findings of this study to choose cultivars that are most suited for generating drought-resistant cultivars and for future research in this sector. Selection based on the seedling vigour index will be productive under stress conditions since it includes an overall assessment of germination percentage, shoot length, and root length. At controlled condition and -3 bar osmotic potential, there was less growth reduction. Osmotic stress at -7.5 bar was highly relevant to screening drought tolerance in pearl millet, and genotypes that performed better at this stress level were chosen for further field screening. In this experiment, parental line *viz.*, PT5456, PT5721, PT5748, PT6303, PT7035, seed parent ICMB 99222, and landraces *viz.*, *Cumbu1*, *Cumbu2* and *Nattu cumbu* performed well and more drought tolerance than CO10 and CO10. As a result, these genotypes might be further tested under *in-vivo* to identify their drought tolerance capacity. Plant development in arid and semi-arid terrain is dependent on the plant's tolerance to drought stress, as well as the capacity of seeds to germinate optimally under these adverse conditions. As a result, it is critical to identify drought-tolerant hybrids during the main growth stage.

Conflict of interest: none

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