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Root Response to Drought Stress: Insights from Peg-Induced Patterns in Rice Germplasms

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ABSTRACT

Drought stress threatens rice productivity, especially in South Asia, where climate variability exacerbates water scarcity. This study investigates root growth responses in 29 rice germplasms (7 varieties, 22 landraces) from the drought-prone Red-Lateritic Zone of West Bengal, India. Using polyethylene glycol at four different concentrations (i.e. 0%, 5%, 10%, and 20%), stress was induced and root growth of each germplasm was monitored over 10 days. Four distinct root growth patterns emerged: Pattern 1 showed the highest average root length (ARL) under control, with reductions of 33.68%, 57.72%, and 97.79% with increasing stress. Pattern 2 exhibited a sharp initial decline (~50%) that stabilized across higher stress levels. Pattern 3 displayed a decrease in mild stress, followed by a gradual increase in ARL under moderate and severe stress. Pattern 4, unique to the landrace Kalpana, showed an ARL increase under mild stress surpassing control, but ARL dropped to zero under severe conditions. These patterns highlight diverse root adaptations to drought. This study offers insights into breeding strategies to enhance drought tolerance through targeted root traits in rice.

Keywords: *Rice landrace, Poly ethylene glycol, Drought stress, Rooting pattern*

Introduction

Rice is the grain of life and a staple food for much of the world, particularly in South and Southeast Asia, where its cultivation is under growing pressure from surging populations and industrialization [1,2]. With shrinking arable land and shifting climate patterns, the impact of drought has become a critical threat to agriculture, leading to significant crop losses worldwide [3, 4, 5]. Drought affects over a third of the world's cultivated land, making it a key factor in the decline of global rice productivity [6].

Roots, as essential organs for water and nutrient absorption, are pivotal in determining a plant's response to drought [3, 7]. In rice, drought often triggers enhanced root growth while inhibiting shoot growth, thereby increasing the root-shoot ratio[8, 9, 10]. Ferreira et al., (2015) have also observed higher root-shoot ratio, higher root biomass accumulation, etc. in other plants including trees. Post-drought rewatering has also been shown to stimulate root growth, a vital process for crop recovery. Given the fluctuating moisture levels in drought-prone environments, evaluating root extension under different moisture conditions is crucial for understanding drought resistance [11, 12, 13].Polyethylene glycol (PEG) is frequently used in studies to simulate drought conditions and assess root growth. Research has demonstrated the utility of various PEG concentrations to induce drought responses, with concentrations ranging from 18.1% [14] to 25% [15] being proposed for evaluating rice cultivars. However, determining the optimal PEG concentration for drought resistance evaluation remains a challenge.Quantifying root growth under these stress conditions, particularly through mathematical modeling, provides valuable insights. Such models by Susilawati et al. (2022),and De et al., (2024b) allow for precise comparisons of root responses across different rice genotypes and environmental conditions, offering a powerful tool for simulating and predicting drought resilience.Therefore, the pattern of root growth under drought stress plays a pivotal role in determining a plant's drought tolerance and its ability to recover in terms of yield after facing such harsh abiotic conditions.

Materials and Methods

In our study, we examined 29 rice germplasms (Table 1), sourced from key institutions in drought-prone areas: the Zonal Drought Resistance Paddy Research Station in Hathwara, Purulia; Krishi Vigyan Kendra in Jahajpur, Purulia; and the Amarkanan Rural Welfare Society in Bankura, West Bengal for their drought resistance.Among these, 7 germplasms are established varieties, while 22 are traditional landraces, cultivated primarily in the Red Lateritic Zone (Zone IV) of Purulia and Bankura district, located in the Chhotanagpur Plateau region. These landraces, adapted to the region's challenging environmental conditions, offer unique insights into natural drought resilience and could hold the key to future breeding programs aimed at improving drought tolerance in rice.

Table 1: List of germplasms

Sl no.	Germplasm	Parentage	Specification
1	Sahabhagi Dhan	IR 55419-4*2/WAY RAREM [16]	DT
2	DRR44	IR 71700-247-1-1-2/IR 03 L120 [16]	DT
3	DRR42	Aday Sel/*3 IR 64 [16]	DT
4	Vandana	C22/Kalakeri[17]	DT
5	Nagina 22	Selection from landrace Rajbhog[18]	DT
6	Swarna	Vasista/Mahsuri [19]	DS, HYV
7	Lalat	Obs 677/IR2071//Bikram/W 1263 [20]	HYV
8	Kerala Sundari	-	L
9	Bhuri	-	L
10	Maniksal	-	L
11	Chhotodidi	-	L
12	Lohasal	-	L
13	Aswinsal	-	L
14	Morogjhota	-	L
15	Sonagori	-	L
16	Tulsikamal	-	L
17	Bhundi	-	L
18	Bhadoi	-	L
19	Kelesh	-	L
20	Vasamanik-I	-	L
21	Velchi-I	-	L
22	Langalmathi	-	L
23	Bhramarmali	-	L
24	Chandrakanti	-	L
25	Kalpana	-	L
26	Neta	-	L
27	Lakkansal	-	L
28	Vutmuri	-	L
29	Kashiphool	-	L

**DT: Drought Tolerant, DS: Drought Susceptible, HYV: High Yielding Variety, L: Landrace*

All 29 germplasms were subjected to varying levels of drought stress using polyethylene glycol (PEG) solutions, starting from 0% PEG concentration (no stress/control) to 5%, 10%, and 20% PEG, representing mild, moderate, and severe drought conditions respectively. The osmotic potential of each treatment was calculated based on the Michel and Kaufmann (1973) equation (Table 2). The equation is mentioned below, where the osmotic potential is represented by ψ.

$\psi = -(1.18 \times 10\text{-}2)\, C - (1.18 \times 10\text{-}4)\, C2 + (2.67 \times 10\text{-}4)\, CT + (8.39 \times 10\text{-}7)\, C2T$………Eq. (1)

Where C is the concentration of PEG 6000 in g/kg of H_2O So, for 5% PEG, C is 50; 10% PEG, C is 100; 20% PEG, C is 2000 and T= Temperature i.e. 25^{o}C

Each germplasm underwent treatment with all four PEG concentrations.

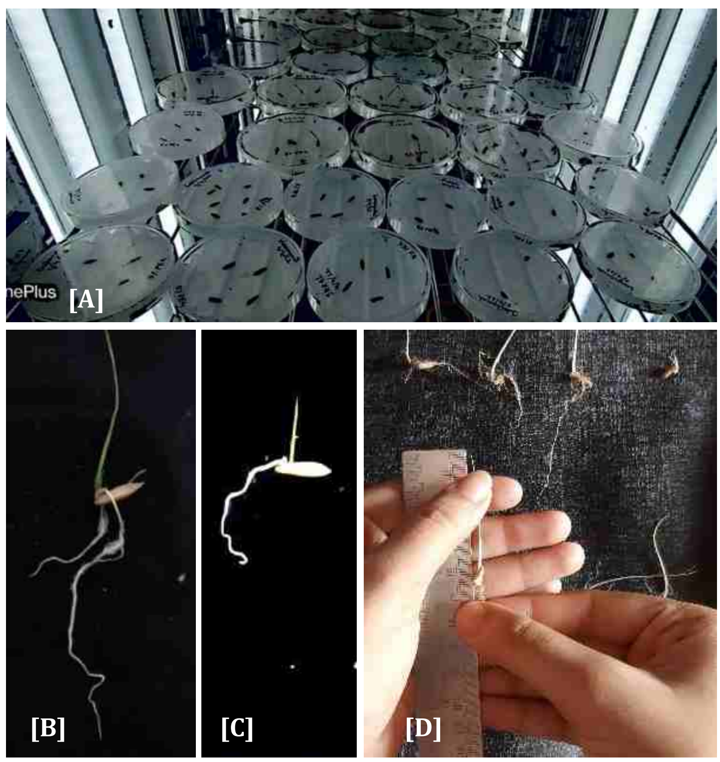
Table 2: Osmotic potential of different PEG solutions

Table 2: Osmotic potential of different PEG solutions	
PEG Concentration	**Osmotic potential (bar)**
0%	0
5%	-0.49
10%	-1.48
20%	-4.91

To begin, seeds were surface sterilized and exposed to various levels of drought stress for 10 days in petri plates and test tubes, with five replications per treatment. Root length was measured daily: in petri plates using a ruler, and in test tubes using non-contact image analysis via ImageJ software (Fig. 1). By the 10th day, total root lengths were recorded for each germplasm.

Results and Discussion

The average root length (ARL) on the 10th day for each treatment was then plotted to reveal distinct root growth patterns. Interestingly, four unique growth patterns emerged across the different stress levels, offering valuable insights into how each germplasm responds to varying drought conditions.



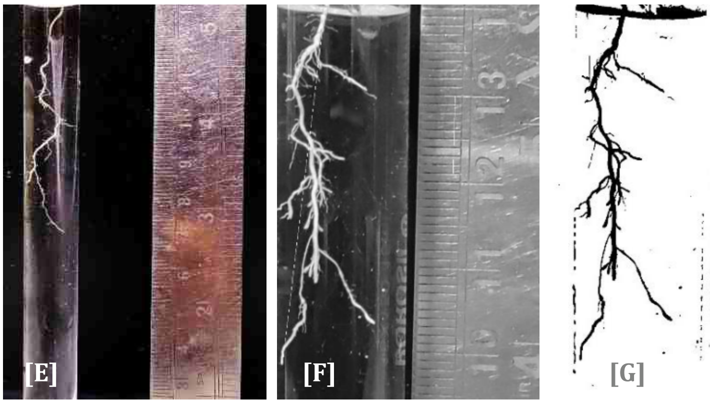
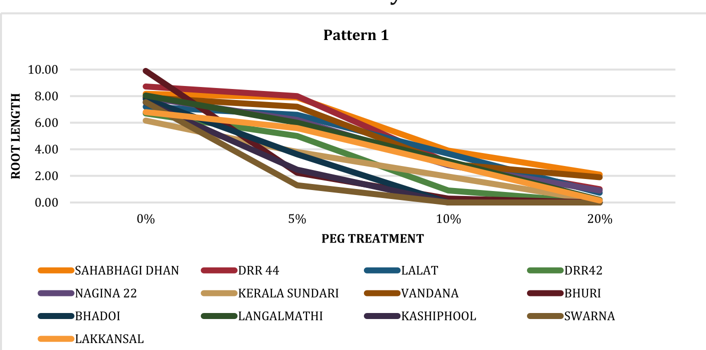


Fig 1: [A]- Treatment in Petri plates, [B]- Root growth under mild stress, [c]- Reduced root growth under severe stress, [D]- Measurement of root length in contact method using a scale, [E]- Treatment in test tubes, [F]- Original RGB image and [G]- its 8-bit image in Image J software [non-contact method].

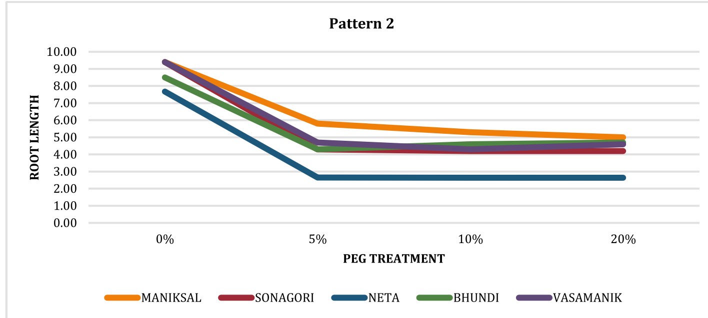
Pattern 1- Thirteen germplasms, including all seven established varieties, exhibited their highest average root length (ARL) under control conditions. However, as drought stress intensified, root growth declined significantly, with the ARL reaching its lowest under severe stress conditions. Specifically, the ARL dropped by an average of 33.68% under mild stress, 57.72% under moderate stress, and an alarming 97.79% under severe PEG stress.

Certain germplasms like Bhadoi, Kashiphool, and Swarna showed no measurable ARL under 10% and 20% PEG stress, while DRR42, and Bhuri had no root growth at severe stress conditions. Table 3 presents the detailed ARL and corresponding percentage reductions across different stress levels. These findings are in line with earlier studies [22, 23], which also demonstrated a sharp decrease in root length as drought stress increases. Similarly, Patmi and Pitoyo (2020) reported that drought not only reduces root length but also diminishes the area of root aerenchyma in rice.



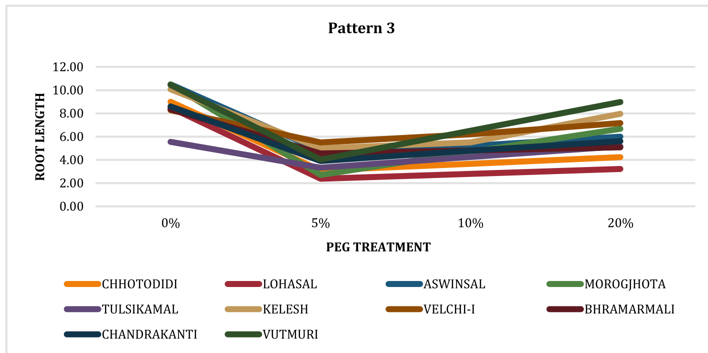
Pattern 2-In this pattern, the highest ARL was observed under control conditions, followed by a sharp decline in mild stress. Interestingly, beyond this initial drop, the ARL remained relatively stable under both moderate and severe stress conditions. The reduction in ARL was consistent, with values of 51.49%, 52.92%, and 52.70% for mild, moderate, and severe stress, respectively, indicating a steady 50% decrease in root growth across all stress levels.

Landraces such as Maniksal, Sonagori, Neta, Bhundi, and Vasamanik exhibited this distinct root elongation pattern, as detailed in Table 3. These results highlight a unique drought tolerance mechanism where root growth is sharply curtailed early on but then stabilizes under increasing drought intensity.



Pattern 3-This category encompassed the largest number of landraces, where ten germplasms showed their highest ARL under control conditions, followed by a sharp decrease at mild stress. Interestingly, as the stress level intensified, their ARL gradually increased, reversing the initial decline. The reduction in ARL was 55.18% under mild stress, 45.05% under moderate stress, and 32.40% under severe stress, indicating a notable recovery in root growth as drought conditions worsened.

The most pronounced increase in ARL under 20% PEG stress was observed in landraces such as Vutmuri, Kelesh, and Velchi-I. These findings align with previous studies on rice [7, 25, 26], which reported enhanced root growth under stress during key stages—seedling, vegetative, and reproductive—likely as an adaptive response to maximize water and nutrient uptake under drought conditions.



Pattern 4- Lastly, a unique root growth pattern was observed in the landrace Kalpana. Remarkably, its root length increased under mild stress, even surpassing the control condition by 10%. However, as drought stress intensified, root growth sharply declined, reaching zero under 20% PEG stress. This unusual growth response could be attributed to specific genetic or physiological factors unique to Kalpana.

A study by De et al., 2024(b) further explored this distinctive pattern where a logistic equation was applied to simulate its root growth and predict drought resistance. This mathematical modeling offers valuable insights into how such a rare growth response might contribute to drought tolerance in extreme conditions.eversing the initial decline. The reduction in ARL was 55.18% under mild stress, 45.05% under moderate stress, and 32.40% under severe stress, indicating a notable recovery in root growth as drought conditions worsened.

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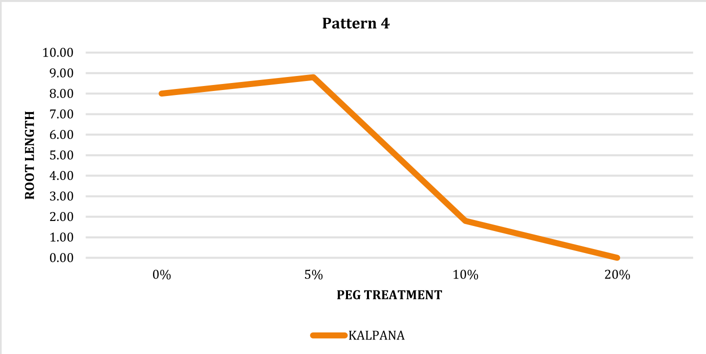

Conclusion

This study reveals distinct root growth patterns among 29 rice germplasms under varying levels of drought stress. The diversity in root responses, ranging from gradual reductions to increases in root length under stress, highlights the complex mechanisms of drought tolerance. Out of the twenty-two landraces, those following Pattern 2 (i.e. Maniksal, Vasamanik) and 3 (Vutmuri, Kelesh) where the root length either remained constant or increased under stress respectively, suggest potential genetic traits for enhancing resilience in extreme conditions of drought. Notably, the unique behavior of the landrace Kalpana leaves room for further studies regarding its genetic traits under stress. These findings are of paramount importance offering valuable insights for future breeding programs aimed at improving drought tolerance in rice.

Statements and Declarations

Funding and competing interests: No funding was received for conducting this study. The authors have no competing interests to declare that are relevant to the content of this article.

Data availability statement: All data generated or analyzedduring this study are included in this published article.

Author contribution

Conceptualization: Anirneeta De, Avishek Dey; Methodology: Anirneeta De, Avishek Dey; Formal analysis and investigation: Anirneeta De, Avishek Dey; Writing – original draft: Anirneeta De, Sailesh Chattopadhyay; Writing – review & editing: Anirneeta De, Avishek Dey, Sailesh Chattopadhyay, Subrata Raha, Dipak Kumar Kar; Supervision: Avishek Dey, Dipak Kumar Kar

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Table 3: Root length and its reduction percentage under stress for 29 germplasms

Variety name	0% PEG	5% PEG	10% PEG	20% PEG	Red. at 5% PEG	Red. at 5% PEG	Red. at 5% PEG
Sahabhagi Dhan	8.18	7.90	3.90	2.10	3.36	52.29	74.31
DRR 44	8.73	8.00	2.99	1.00	8.31	65.75	88.54
Lalat	7.20	6.60	3.67	0.73	8.33	49.07	89.81
DRR 42	6.70	5.00	0.90	0.00	25.37	86.57	100.00
Nagina 22	7.90	6.20	2.80	0.90	21.52	64.56	88.61
Kerala Sundari	6.16	3.80	1.95	0.10	38.31	68.34	98.38
Vandana	7.90	7.20	2.90	1.90	8.86	63.29	75.95
Bhuri	9.90	2.23	0.30	0.00	77.44	96.97	100.00
Bhadoi	7.78	3.63	0.00	0.00	53.38	100.00	100.00
Langalmathi	8.05	6.00	3.10	0.20	25.47	61.49	97.52
Kashiphool	7.53	2.48	0.00	0.00	67.11	100.00	100.00
Swarna	7.55	1.30	0.00	0.00	82.78	100.00	100.00
Lakkansal	6.80	5.60	2.88	0.15	17.65	57.72	97.79
PATTERN 1	AVERAGE				33.68	74.31	93.15
Maniksal	9.40	5.80	5.30	5.00	38.30	43.62	46.81
Sonagori	9.40	4.30	4.20	4.20	54.26	55.32	55.32
Neta	7.68	2.65	2.65	2.64	65.47	65.54	65.60
Bhundi	8.50	4.30	4.60	4.70	49.41	45.88	44.71
Vasamanik	9.40	4.70	4.30	4.60	50.00	54.26	51.06
PATTERN 2	AVERAGE				51.49	52.92	52.70
Chhotodidi	9.00	3.10	3.66	4.23	65.56	59.31	53.06
Lohasal	8.50	2.38	2.80	3.22	72.06	67.09	62.12
Aswinsal	10.50	4.80	5.20	6.00	54.29	50.48	42.86
Morogjhota	10.50	2.70	4.69	6.68	74.29	55.33	36.38
Tulsikamal	5.55	3.33	4.24	5.16	40.09	23.56	7.03
Kelesh	10.08	5.00	5.50	7.96	50.37	45.41	20.99
Velchi-I	8.28	5.50	6.20	7.16	33.53	25.08	13.47
Bhramarmali	8.35	4.55	4.82	5.08	45.51	42.34	39.16
Chandrakanti	8.60	3.90	4.80	5.60	54.65	44.19	34.88
Vutmuri	10.45	4.03	6.50	8.98	61.48	37.78	14.07
PATTERN 3	AVERAGE				55.18	45.05	32.40
Kalpana	8.00	8.80	1.80	0.00	-10.00	77.50	100.00

**Red. at 5% PEG, Red. at 5% PEG, and Red. at 5% PEG indicates the reduction percentage of RL under 5%, 10%, and 20% stress from control condition respectively. Only Kalpana had a negative reduction i.e. an increment in RL at 5% PEG stress.*

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