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Exploring genetic diversity and trait associations with foliar blast disease among parental lines in pearl millet [*Pennisetum glaucum* (L.) R Br]

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ABSTRACT

Thirty-seven pearl millet genotypes were evaluated for morphometric traits and disease incidence and severity during *summer and kharif*, 2022. Pooled ANOVA revealed significant variation were present in all genotypes across different season. Association studies identified high positive correlations between grain yield and traits such as single earhead weight, single ear head threshed weight, and test weight, with direct and indirect effects on grain yield through key characters. E_2 (*kharif* – 2022) showed favourable conducive weather parameters for disease infection throughout the growing season compared to E_1 . The higher PCV relative to GCV for disease incidence underscores the environmental influence in disease resistance programs. Negative correlations between disease metrics and yield traits highlight blast disease's detrimental effect on grain yield. Disease severity indirectly suggests environmental factors may enhance its impact. Disease incidence exhibited a direct negative impact on yield, supported by negative genotypic correlations. The line, PT 6679, exhibit both high yield and highly resistant to blast. Restorer lines (PT 6029, PT 6067, PT 6300, PT 6707, PT 7068) and B lines (ICMB 01666, ICMB 02777) showed promising yield attributes with high to moderate disease resistant for future breeding programs. In D^2 analysis, five clusters revealed distinct genetic diversity with Clusters II and V indicating strong hybrid vigor, while Clusters IV (PT 6946, ICMB 06111) and V (ICMB 93111, ICMB 95444) excelled in disease resistance. Clusters I (PT 6029, PT 7068) and II (GMR 58) exhibited superior grain yield, particularly Cluster I, had potential restorer lines for future breeding. Clear differentiation between B and R lines underscored genetic distinctions in trait expression, validating the use of morphological data for assessing genetic diversity.

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INTRODUCTION

Pearl millet is a resilient and versatile cereal crop crucial for regions where major cereals struggle to grow. Recognized for its adaptability and drought tolerance, it is a lifeline in arid areas. India dominates global cultivation, contributing 43.3% of the cultivation area and 42% of the yield. The cultivation area spans around 7.55 million and 59,956 hectares in India and Tamil Nadu producing 9.22 and 1.46 lakh tones, respectively. Productivity stands at 1747 kg/ha in India and 2,437 kg/ha in Tamil Nadu (DES, 2021). While previously regarded as a minor, blast disease has now become a significant problem for pearl millet in India, as Thakur *et al.* (2009) reported. This shift in research focus from downy mildew to blast disease is attributed to the extensive spread of blast disease throughout the country (Sharma *et al.*, 2020). To address the challenge of biotic stress

and boost crop productivity, a viable approach is to engage in breeding programs aimed at enhancing disease resistance in pearl millet genotypes.

Pooled analysis assesses the average response of experimental materials under diverse environmental conditions, examining consistency in treatment-induced variations across years and locations. GCV and PCV quantify variation, providing insights into material variability. Heritability and genetic advance as a percentage of mean are crucial for successful crop improvement. Considering both primary and associated traits, along with validating the true influence of grain yield through path coefficient analysis, enhances breeding program effectiveness (Patil *et al.*, 2018). Mahalanobis's generalized distance calculated using D^2 statistics (Rao, 1952), is one of these methods, stands out as a valuable and distinctive approach

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for evaluating the diversity of traits, identifying clusters of genotypes that are genetically distant, and mainly focusing the significant traits that contribute to the overall variation within the germplasm can be established for identifying optimal parents in trait-specific line/hybrid development programs (Khandelwal *et al.*, 2023). Hence, this study aims to identify genotypes with high yield and blast disease resistance across seasons. Genetic parameters and association studies will guide breeding programs, providing insights into genetic diversity and aiding in the selection of blast-resistant parental lines for pearl millet hybrid development.

MATERIALS AND METHOD

A set of 37 parental lines of pearl millet which includes 19 restorer and 18 designated lines were raised by using Randomized Block Design in clay loam soil at Department of Millets, TNAU, Coimbatore (11° N latitude, 77° E longitude and 426.72 m MSL) during two different seasons (*summer* (E₁) and *khariif* (E₂), 2022) with average rainfall 680 mm. Season wise weather parameters details are given in Table 1. Each parental lines was raised in two rows for each replication with a plant spacing of 50 x 15 cm and a row length of 4 m. Systematic susceptibility assessments were conducted to elevate the disease pressure by planting ICMB 89111 and ICMB 95222 on every sixth row in an alternatively. At the hard dough stage (60 days after sowing), the level of disease severity was assessed using a progressive scale ranging from 1 to 9, which was devised by the International Rice Research Institute (IRRI) in the Philippines (2013). The prescribed agronomic procedures and practices were adhered to throughout period of experiment.

Data were recorded for 14 morphometric traits from five randomly selected plants in each experimental material as well as disease related traits *viz.*, disease incidence and disease severity were recorded. Morphometric traits include days to fifty flowering; leaf blade length (cm); leaf blade width (cm); leaf sheath length (cm); number of economic tillers/plants (no of productive tillers); number of nodes; spike length (cm); spike diameter (cm); plant height (cm); days to maturity; single earhead weight (g), single earhead threshed weight (g); test grain weight (g); Grain yield (g) was observed. Disease incidence and disease severity were calculated based on blast disease infection in experimental material. These traits were recorded as percentage data and it was transformed to arcsine values before proceeding to analysis. Categorization of genotypes for disease reaction was carried out based on disease severity score as reported by Thakur *et al.* (2009) [Score 1 - highly resistant, score 2-3 - Resistant, score 3-5 - Moderately resistant, score 5-7 - Susceptible and score 7-9 - Highly susceptible].

Disease incidence and disease severity will be calculated by using the formula

$$\text{Disease severity} = \frac{\text{Sum of individual disease rating}}{\text{total number of plants} \times \text{max grade}} \times 100$$

$$\text{Disease incidence} = \frac{\text{Number of infected plants}}{\text{total number of plants}} \times 100$$

Bartlett ‘t’ test was carried out for testing the homogeneity variance in the experimental material before proceeding the pooled analysis. The collected data was subjected to variability and association analysis using Windowstat ver 7.1 and D² by Tocher method.

RESULT AND DISCUSSION

Season-wise and combined ANOVA were conducted, with pooled analysis confirming homogeneity of variance through Bartlett’s test. Combined ANOVA revealed significant genotype differences across both seasons (E₁ & E₂), indicating substantial variation in the experimental material (Table 2). Genotype x season interaction analysis showed significant differences in most traits, highlighting the major contributions of genotypes and their interaction effects (Govindaraj *et al.*, 2020; Gangashetty *et al.*, 2021).

Relationship between Blast Disease Traits and Yield

Performance of parental lines of pearl millet for foliar blast disease across two different seasons were presented in Table 3. Out of 37 genotypes, six lines and one line showed highly resistant in E₁ and E₂ respectively. Six lines in E₁ and twelve lines in E₂ were grouped under resistant category, whereas 19 lines in E₁ and 18 lines in E₂ were registered moderately resistant. Four lines from E₂ environment were registered highly susceptible and none showed highly susceptible in E₁. The variations in blast infection levels from E₁ to E₂ can be attributed to the corresponding fluctuations in weather parameters documented throughout the growing season (Koutroubas *et al.*, 2009).

Comparing mean performance of resistant lines with susceptible check in E₁, among the R lines, PT 6679 (110 g) and PT 6029 (89 g) and among the B lines ICMB 01666 (82 g) and ICMB 02777 (83 g) showed high mean performance of grain yield as well as resistant to blast disease over susceptible check namely, ICMB 89111 (63 g) and ICMB 95222 (70 g) whereas in E₂, only PT 6679 were recorded resistant against blast disease with high grain yield. Considering both grain yield with disease - related traits, the lines namely, PT 6679 showed high yield with highly resistance to blast whereas PT 6029, PT 6707, PT 7068, ICMB 01666 and ICMB 05888 were recorded high yield with high to moderate resistance to blast. ICMB 02777 and ICMB 04111 were comes under moderate yield with highly

Table 1: Weather information of experimental season

S. No.	Season	Average Temp (°C)		Average Humidity (Morning) (%)	Average Humidity (Evening) (%)	Rainfall (mm)
		Min	Max			
1	Summer - 2022 (Feb - May)	33.6	22.9	82.6	45.6	69.4
2	Khariif - 2022 (Aug - Oct)	30.4	22.4	82.9	57.8	235

Table 2: Mean sum of squares of blast disease incidence, disease severity and yield related traits over two seasons (E₁ & E₂) in pearl millet

Source of variation	Treatment	Season	Treatment x Season	Pooled error
Df	36	1	36	72
DFF	54.09*	20.43*	2.06	2.51
LSL	8.08*	17.08*	0.15	0.65
LBL	346.58*	86.31*	3.24*	5.23
LBW	1.85*	0.39*	0.09*	0.06
NON	6.93*	2.07*	0.49*	0.24
SPL	150.49*	265.70*	5.56*	1.63
SPD	9.20*	7.05*	0.67*	0.06
NOET	1.80*	38.00*	0.34*	0.27
PLH	4638.72*	972.35*	64.53*	6.81
DTM	49.54*	636.82*	0.85*	2.65
SEWT	165.69*	436.73*	6.77*	6.73
SETWT	113.29*	293.80*	6.54*	5.31
TWT	11.24*	3.80*	0.03	0.10
DI% (T)	447.47*	3716.88*	33.87*	14.39
DS% (T)	203.73*	1985.61*	18.40*	4.69
GYP	597.34*	11199.30*	31.96*	12.02

DFF - days to fifty per cent flowering, LSL - leaf sheath length, LBL - leaf blade length, LSW - leaf blade width, NON - Number of nodes, SPL - spike length, SPD - spike diameter, NOET - number of economic tillers/plant, DTM - Days to maturity, PLH - plant height, SEWT - Single Earhead Weight, SETWT - Single Earhead Threshed Weight, TWT - 1000 grain weight, GYP - Grain yield/Plant, DI% (T) - Disease Intensity % transformed data, DS% (T) - Disease Severity % transformed data

Table 3: Performance of parental lines of pearl millet for foliar blast disease across two different seasons

Material	No of lines with blast disease severity										Total number of genotypes
	1		2-3		3-5		5-7		7-9		
	E1	E2	E1	E2	E1	E2	E1	E2	E1	E2	
B lines	4	-	3	4	8	9	3	1	-	3	18
R lines	2	1	4	8	12	9	1	1	-	1	19
Total	6	1	7	12	20	18	4	2	-	4	37

resistant. Kumar *et al.* (2013) reported that, resistant genotypes consistently exhibited higher level of phenols, orthodihydroxy phenols and chlorophyll compared to susceptible genotypes across all the time interval and growth stages of crop and this component has been recognized in disease resistance reaction (Walker, 1923).

When considering mean performance of susceptible to moderate susceptible category, the genotypes *viz.*, PT 6476 (81 g), PT 6676 (88 g), ICMB 94333 (74 g) were displayed moderate mean performance of grain yield over susceptible check ICMB 89111 (63 g) and ICMB 95222 (71 g) over both E₁ and E₂ seasons.

Pooled PCV and GCV estimates highlighted higher phenotypic variation than genotypic variation, indicating genotype-environment interactions (Ram *et al.*, 2014; Sharma *et al.*, 2018). The elevated PCV compared to GCV in disease incidence and severity percentages underscored the substantial influence of environmental factors during the trial, emphasizing the environment’s role in promoting disease

prevalence among diverse genotypes (Table 4). Heritability and GMA studies for disease severity and incidence revealed high heritability coupled with significant GMA. Namrata *et al.* (2019) reported specifically in relation to disease severity in rice blast disease.

Genotypic correlations between quantitative traits were examined across two seasons, summer (E₁) and kharif (E₂), as presented in Table 5. In E₁, disease incidence and severity exhibited a significant negative correlation with grain yield. A similar trend was observed in E₂, indicating a consistent negative correlation of grain yield with disease-related traits across both seasons. The findings emphasize a substantial reduction in grain yield when affected by blast disease, highlighting the significant contribution of leaf blast infection to yield decline. Namrata *et al.* (2019) had previously reported a similar result in rice blast disease. Unlike E₁, E₂ demonstrated a notably high and significant negative correlation with grain yield. The differences in blast infection between the two seasons are likely attributed to variations in weather parameters during the respective growing periods. Environmental conditions, particularly relative humidity, play a crucial role in blast disease development. Blast infection negatively impacts canopy photosynthesis by affecting the photosynthetic rate and inducing senescence, contributing to yield losses (Koutroubas *et al.*, 2009).

The specific values related to the path analysis can be found in Table 6. The disease-related traits, specifically disease incidence directly affected grain yield in Rice (Kumar *et al.*, 2022). Disease incidence showed negative indirect effect *via* all traits except disease severity. Disease severity exhibited positive indirect effect and indicate that, environmental factors conducive to disease spread increase, disease severity also rises, affecting the dependent variables.

Relationship between Grain Yield with other Yield Attributing Traits

Among the fourteen characters, the yield attributing characters like spike length, spike diameter, single earhead weight, single earhead threshed weight and test weight showed higher mean values and range in the restorer lines *viz.*, PT 6029, PT 6067, PT 6300, PT 6676, PT 6679 and PT 7068. Among the B lines, ICMB 93222, ICMB 01666 and ICMB 02777 showed high to moderate mean values for majority of yield contributing traits. The parental lines showed significant variation, which can be employed for future breeding programme, according to their *per se* performances.

The characters *viz.*, spike length, plant height, single earhead threshed weight, and grain yield exhibited high GCV and PCV (>22%), indicating substantial genetic variation for improvement (Table 4). Studies by Talawar *et al.* (2017) and Subbulakshmi *et al.* (2022) supported these findings for plant height, while Subbulakshmi *et al.* (2018) reported similar trends for single earhead threshed weight. Sharma *et al.* (2018) found comparable results for spike diameter and grain yield. Selection

Table 4: Genetic parameters for blast disease incidence, disease severity and different yield attributing traits over two seasons (E_1 & E_2) in Pearl millet

Traits	Mean	Range		ECV	GCV	PCV	h ²	GAM (5%)
		Minimum	Maximum					
DFF	46.78	36	53.3	3.29	7.69	8.36	84.60	14.56
LSL	12.39	9.3	15	5.63	11.13	12.47	79.60	20.46
LBL	57.71	41.7	74.6	3.28	16.15	16.48	96.00	32.60
LBW	4.06	2.6	6.1	6.76	16.43	17.76	85.50	31.29
NON	6.72	4.3	9	8.49	19.14	20.94	83.60	36.03
SPL	25.23	13.2	38	6.80	24.07	25.01	92.60	47.72
SPG	9.04	4.3	11.2	5.72	16.54	17.50	89.30	32.20
NOET	4.36	3	6.3	13.82	17.60	22.38	61.80	28.51
PHT	154.10	74.4	223.5	3.31	22.04	22.28	97.80	44.89
DTM	86.48	76	92.5	1.66	3.99	4.32	85.30	7.58
SEWT	32.95	21.8	44.9	8.09	19.62	21.22	85.50	37.35
SETWT	25.69	14.6	36.3	9.58	20.76	22.86	82.50	38.83
TWT	11.13	7.5	14.5	2.56	15.00	15.22	97.20	30.46
GYP	33.03	51.3	102.8	13.84	31.27	34.19	83.60	58.90
DI% (T)	26.61	13.9	57.7	20.51	46.97	51.25	84.00	88.67
DS% (T)	46.78	14.8	44.3	5.40	14.80	15.76	88.30	28.64

DFF - days to fifty per cent flowering, LSL - leaf sheath length, LBL - leaf blade length, LSW - leaf blade width, NON - Number of nodes, SPL - spike length, SPD - spike diameter, NOET - number of economic tillers/plant, DTM - Days to maturity, PLH - plant height, SEWT - Single Earhead Weight, SETWT - Single Earhead Threshed Weight, TWT - 1000 grain weight, GYP - Grain yield/Plant, DI% (T) - Disease Intensity % transformed data, DS% (T) - Disease Severity % transformed data

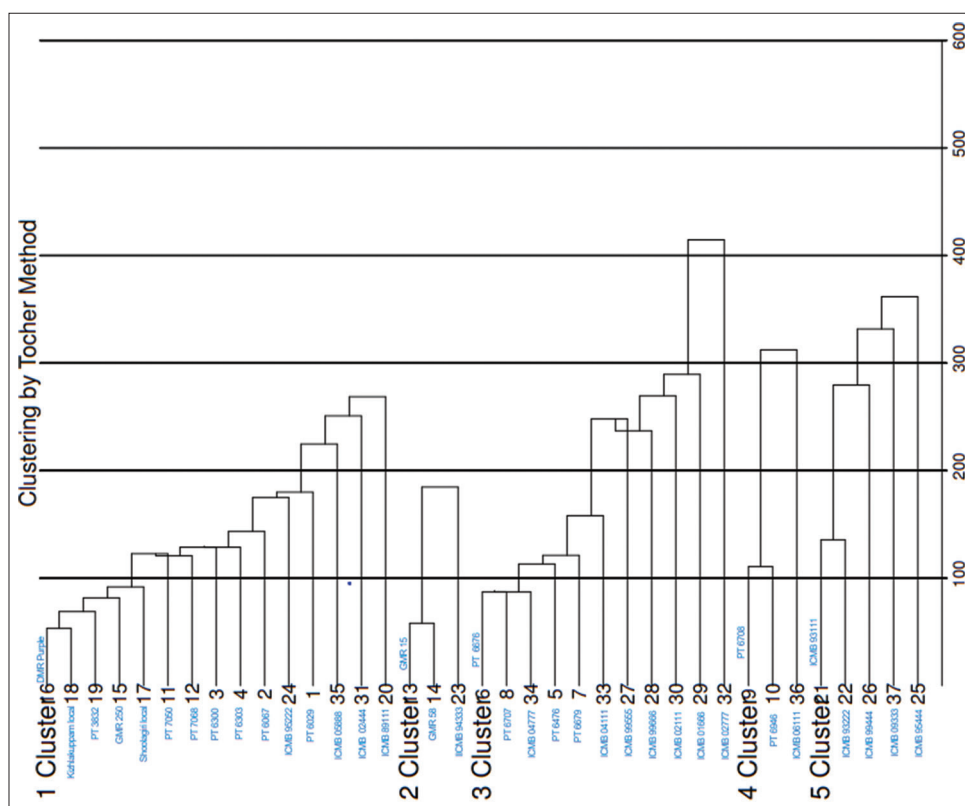


Figure 1: Dendrogram of parental genotypes of pearl millet by Tocher's method

strategies for these traits may be challenging, but reliable conclusions in plant breeding can be drawn from heritability estimates coupled with genetic advance. Generally high heritability, combined with varying values of GAM, suggests additive gene action influencing these traits, making direct selection procedures effective for improvement. Rajpoot *et al.* (2023) reported similar trends in variability studies in pearl

millet germplasm, aligning with observations of high heritability and genetic advance by Anuradha *et al.* (2018).

In both E_1 and E_2 , all morphometric traits, except the number of economic tillers per plant, showed a positive and significant genotypic correlation with grain yield (Table 5). Notably, leaf blade length, single earhead weight, single earhead threshed

Table 5: Correlation coefficient of blast disease incidence, disease severity and yield attributing traits for different traits in pearl millet

Traits	Season	DFD	LSL	LBL	LBW	NON	SPL	SPG	NOET	PLH	DTM	SEWT	SETWT	TWT	DI % (T)	DS% (T)	GPY
DFD	E1	0.22	0.33*	0.21	0.14	0.50*	0.16	-0.22	-0.28	1	0.18	0.18	0.12	-0.34*	-0.35*	0.17	
	E2	0.17	0.26	0.19	-0.02	0.51	0.02	-0.08	-0.29	0.91*	0.39*	0.39*	0.10	-0.37*	-0.37*	0.33*	
	Pooled	0.19	0.30*	0.21	0.06	0.49*	0.09	-0.17	-0.28	0.96*	0.28	0.27	0.11	-0.36*	-0.36*	0.25	
LSL	E1		0.42*	0.36*	0.35*	0.31*	0.73*	0.03	0.42*	0.22	0.40*	0.42*	0.31*	-0.37*	-0.38*	0.55*	
	E2		0.40*	0.40*	0.35*	0.38*	0.61*	0.05	0.44*	0.16	0.47*	0.43*	0.27	-0.35*	-0.39*	0.46*	
LBL	Pooled		0.41*	0.38*	0.33*	0.34*	0.66*	0.04	0.43*	0.20	0.44*	0.42*	0.28	-0.36*	-0.38*	0.51*	
	E1			0.53*	0.51*	0.61*	0.19	-0.19	0.44*	0.33*	0.57*	0.59*	0.27	-0.04	-0.2	0.63*	
	E2			0.63*	0.55*	0.58*	0.10	-0.17	0.53*	0.28	0.58*	0.47*	0.32*	-0.05	-0.18	0.60*	
LBW	Pooled			0.59*	0.51*	0.59*	0.14	-0.18	0.48*	0.31*	0.57*	0.53*	0.30*	-0.04	-0.19	0.61*	
	E1				0.36*	0.34*	0.32*	-0.04	0.41*	0.21	0.39*	0.31*	0.44*	-0.12	-0.10	0.59*	
	E2				0.59*	0.39*	0.29	-0.04	0.57*	0.18	0.42*	0.27	0.51*	-0.12	-0.12	0.57*	
NON	Pooled				0.48*	0.37*	0.29	-0.04	0.49*	0.20	0.40*	0.28	0.47*	-0.13	-0.11	0.57*	
	E1					0.24	0.30*	-0.14	0.61*	0.14	0.49*	0.43*	0.43*	-0.22	-0.18	0.49*	
	E2					0.25	0.36*	-0.15	0.69*	0.01	0.29	0.08	0.46*	-0.06	-0.07	0.36*	
SPL	Pooled					0.22	0.30*	-0.12	0.64*	0.08	0.38*	0.25	0.43*	-0.14	-0.11	0.40*	
	E1						0.19	-0.19	0.17	0.50*	0.42*	0.45*	-0.05	-0.25	-0.35*	0.38*	
	E2						0.10	-0.03	0.28	0.41*	0.56*	0.49*	0.09	-0.25	-0.41*	0.58*	
SPG	Pooled						0.15	-0.17	0.22	0.48*	0.48*	0.46*	0.01	-0.25	-0.37*	0.47*	
	E1							0.18	0.36*	0.16	0.28	0.24	0.29	-0.31*	-0.27	0.32*	
	E2							0.01	0.33*	0.02	0.25	0.23	0.44*	-0.31*	-0.36*	0.37*	
NOET	Pooled							0.13	0.34*	0.10	0.26	0.24	0.35*	-0.30*	-0.30*	0.34*	
	E1								0.01	-0.22	-0.04	0.04	-0.30*	-0.20	-0.13	-0.04	
	E2								0.01	-0.13	0.07	0.07	-0.24	-0.05	-0.03	-0.08	
PHT	Pooled								0.01	-0.19	0.00	0.04	-0.25	-0.11	-0.06	-0.06	
	E1									-0.28	0.36*	0.32*	0.14	0.03	0.06	0.38*	
	E2									-0.24	0.34*	0.19	0.21	0.03	-0.02	0.30*	
DM	Pooled									-0.27	0.36*	0.27	0.18	0.03	0.02	0.34*	
	E1										0.18	0.18	0.12	-0.34*	-0.35*	0.17	
	E2										0.34*	0.32*	0.11	-0.34*	-0.33*	0.27	
SEWT	Pooled										0.26	0.25	0.13	-0.34*	-0.33*	0.22	
	E1											0.89*	0.23	-0.17	-0.25	0.64*	
	E2											0.88*	0.30	-0.32*	-0.4	0.71*	
SETWT	Pooled											0.88*	0.26	-0.24	-0.33*	0.67*	
	E1												0.16	-0.18	-0.28	0.60*	
	E2												0.13	-0.2	-0.28	0.56*	
TWT	Pooled												0.14	-0.19	-0.27	0.58*	
	E1													-0.13	-0.08	0.57*	
	E2													-0.13	-0.15	0.54*	
DI	Pooled													-0.13	-0.12	0.55*	
	E1														0.88*	-0.27	
	E2														0.89*	-0.42*	
DS	Pooled														0.88*	-0.35*	
	E1															-0.28	
	E2															-0.52*	
	Pooled															-0.42*	

*Significant at 5%

weight, and test weight displayed high positive genotypic correlations with grain yield in E₁ and E₂, respectively. This positive association could arise from coupling linkage between genes or pleiotropic effects governing these traits in the same direction (Kyi et al., 2021). Positive and significant correlations between grain yield and contributing traits, as reported by Annamalai et al. (2023) and Narasimhulu et al. (2021) suggest that crucial yield-contributing traits consistently exhibit positive correlations in different seasons. Simultaneous selection of parental lines possessing these desirable traits is likely to yield favorable results.

In path analysis, the traits namely, leaf blade length, spike length, single earhead threshed weight and test weight showed positive direct effect on grain yield on both seasons (Table 6).

Path analysis confirmed the direct positive effects of leaf blade length, spike length, single earhead threshed weight, and test weight on grain yield in both seasons. Pooled analysis identified single earhead weight, single earhead threshed weight, and test weight as crucial traits for selecting high-yielding pearl millet genotypes, supported by significant positive genotypic correlations. According to Reddy et al. (2023), choosing parents based on these traits will be beneficial in achieving a higher grain yield.

Genotype Identification in Relation with Blast Disease and Yield Traits through Diversity Approach

Genetic diversity and identification of genotypes related to disease traits and yield were accomplished through D² analysis

Table 6: Path analysis of blast disease incidence, disease severity and different yield attributing traits over two seasons (E_1 & E_2) in Pearl millet

Traits	Season	DFF	LSL	LBL	LBW	NON	SL	SG	NOET	PLH	DTM	SEWT	SETWT	TWT	DI %	DS%	GYP
DFF	E1	-1.870	0.074	0.077	0.024	-0.002	0.083	-0.038	-0.036	0.015	1.719	0.057	-0.004	0.053	0.071	-0.054	0.17
	E2	0.044	-0.002	0.048	0.038	0.002	0.137	0.002	0.003	0.055	-0.246	0.144	-0.010	0.027	0.032	0.056	0.33
	Pooled	0.231	0.044	0.069	0.048	0.014	0.114	0.022	-0.040	-0.066	0.222	0.066	0.063	0.025	-0.083	-0.084	0.25
LSL	E1	-0.423	0.326	0.096	0.039	-0.004	0.054	-0.174	0.006	-0.022	0.389	0.129	-0.009	0.133	0.077	-0.057	0.55
	E2	0.007	-0.011	0.073	0.077	-0.024	0.101	0.048	-0.001	-0.086	-0.048	0.174	-0.011	0.076	0.030	0.059	0.46
	Pooled	0.021	0.107	0.044	0.041	0.036	0.036	0.071	0.005	0.047	0.022	0.047	0.045	0.031	-0.039	-0.041	0.51
LBL	E1	-0.628	0.137	0.228	0.059	-0.006	0.103	-0.044	-0.030	-0.023	0.577	0.181	-0.013	0.115	0.008	-0.031	0.63
	E2	0.012	-0.005	0.181	0.122	-0.038	0.160	0.007	0.004	-0.102	-0.075	0.214	-0.012	0.094	0.004	0.027	0.6
	Pooled	0.058	0.079	0.194	0.115	0.098	0.115	0.027	-0.034	0.093	0.059	0.111	0.103	0.058	-0.009	-0.036	0.61
LBW	E1	-0.413	0.116	0.123	0.109	-0.005	0.060	-0.075	-0.006	-0.021	0.380	0.125	-0.007	0.185	0.028	-0.016	0.59
	E2	0.009	-0.005	0.115	0.193	-0.041	0.110	0.022	0.001	-0.110	-0.049	0.156	-0.007	0.147	0.011	0.018	0.57
	Pooled	0.024	0.044	0.068	0.114	0.054	0.042	0.033	-0.005	0.056	0.023	0.046	0.032	0.054	-0.015	-0.013	0.57
NON	E1	-0.297	0.109	0.111	0.038	-0.013	0.037	-0.069	-0.017	-0.030	0.273	0.153	-0.009	0.178	0.051	-0.028	0.49
	E2	-0.001	-0.004	0.095	0.111	-0.072	0.063	0.026	0.003	-0.133	-0.003	0.103	-0.002	0.129	0.006	0.009	0.36
	Pooled	-0.004	-0.024	-0.036	-0.034	-0.071	-0.016	-0.022	0.009	-0.045	-0.006	-0.027	-0.018	-0.030	0.010	0.008	0.40
SL	E1	0.924	0.105	0.140	0.039	-0.003	0.168	-0.045	-0.037	-0.009	0.850	0.133	-0.010	-0.022	0.051	-0.055	0.38
	E2	0.022	-0.004	0.107	0.078	-0.017	0.272	0.008	0.002	-0.057	-0.122	0.210	-0.012	0.022	0.023	0.063	0.58
	Pooled	0.085	0.058	0.102	0.063	0.039	0.172	0.026	-0.029	0.038	0.082	0.083	0.079	0.001	-0.043	-0.064	0.47
SG	E1	-0.295	0.237	0.042	0.034	-0.004	0.031	-0.239	0.030	-0.019	0.272	0.087	-0.005	0.125	0.066	-0.042	0.32
	E2	0.001	-0.007	0.017	0.055	-0.024	0.027	0.079	-0.001	-0.063	-0.005	0.091	-0.006	0.126	0.026	0.054	0.37
	Pooled	-0.006	-0.040	-0.009	-0.018	-0.019	-0.009	-0.061	-0.008	-0.021	-0.006	-0.016	-0.015	-0.021	0.019	0.019	0.34
NOET	E1	0.429	0.012	-0.044	-0.004	0.001	-0.039	-0.045	0.159	0.000	-0.395	-0.014	-0.001	-0.117	0.041	-0.018	-0.04
	E2	-0.005	-0.001	-0.030	-0.008	0.010	-0.021	0.003	-0.024	-0.002	0.036	0.024	-0.001	-0.066	0.004	0.004	-0.08
	Pooled	-0.010	0.003	-0.011	-0.003	-0.007	-0.010	0.008	0.060	0.001	-0.011	0.000	0.003	-0.015	-0.007	-0.004	-0.06
PLH	E1	0.530	0.138	0.100	0.045	-0.008	0.028	-0.086	0.001	-0.052	-0.487	0.116	-0.007	0.061	-0.005	0.009	0.38
	E2	-0.013	-0.005	0.095	0.110	-0.049	0.080	0.026	0.000	-0.193	0.067	0.127	-0.005	0.063	-0.003	0.002	0.3
	Pooled	0.011	-0.016	-0.018	-0.018	-0.024	-0.008	-0.013	0.000	-0.037	0.010	-0.013	-0.010	-0.007	-0.001	-0.001	0.34
DTM	E1	-1.870	0.074	0.077	0.024	-0.002	0.083	-0.038	-0.036	0.015	1.719	0.057	-0.004	0.053	0.071	-0.054	0.17
	E2	0.041	-0.002	0.051	0.036	-0.001	0.125	0.001	0.003	0.049	-0.264	0.129	-0.008	0.037	0.029	0.050	0.27
	Pooled	-0.356	-0.074	-0.114	-0.074	-0.030	-0.176	-0.036	0.069	0.100	-0.370	-0.097	-0.091	-0.046	0.125	0.124	0.22
SEWT	E1	-0.338	0.133	0.131	0.043	-0.006	0.070	-0.066	-0.007	-0.019	0.311	0.316	-0.019	0.096	0.035	-0.040	0.64
	E2	0.017	-0.005	0.105	0.081	-0.020	0.154	0.019	-0.002	-0.066	-0.092	0.370	-0.022	0.085	0.027	0.060	0.71
	Pooled	0.077	0.119	0.156	0.110	0.103	0.131	0.072	0.001	0.097	0.071	0.272	0.239	0.070	-0.067	-0.090	0.67
SETWT	E1	-0.340	0.142	0.137	0.034	-0.005	0.074	-0.058	0.006	-0.017	0.312	0.279	-0.022	0.067	0.038	-0.045	0.6
	E2	0.017	-0.005	0.086	0.051	-0.005	0.133	0.018	-0.001	-0.038	-0.089	0.325	-0.025	0.035	0.018	0.042	0.56
	Pooled	0.013	0.020	0.026	0.014	0.012	0.022	0.012	0.002	0.013	0.012	0.042	0.048	0.007	-0.009	-0.013	0.58
TWT	E1	-0.231	0.101	0.061	0.047	-0.005	-0.009	-0.069	-0.043	-0.007	0.212	0.071	-0.003	0.430	0.028	-0.013	0.57
	E2	0.004	-0.003	0.059	0.098	-0.032	0.021	0.034	0.005	-0.042	-0.034	0.109	-0.003	0.289	0.012	0.023	0.54
	Pooled	0.044	0.115	0.121	0.192	0.175	0.002	0.142	-0.102	0.072	0.051	0.105	0.057	0.406	-0.055	-0.050	0.55
DI % (T)	E1	0.636	-0.121	-0.009	-0.015	0.003	-0.041	0.076	-0.031	-0.001	-0.585	-0.053	0.004	-0.057	-0.208	0.136	-0.27
	E2	-0.017	0.004	-0.009	-0.024	0.005	-0.073	-0.024	0.001	-0.007	0.090	-0.118	0.005	-0.040	-0.085	-0.132	-0.42
	Pooled	0.037	0.037	0.005	0.013	0.015	0.025	0.031	0.012	-0.003	0.035	0.025	0.019	0.014	-0.102	-0.090	-0.35
DS% (T)	E1	0.664	-0.121	-0.046	-0.011	0.002	-0.060	0.065	-0.019	-0.003	-0.610	-0.083	0.006	-0.035	-0.185	0.153	-0.28
	E2	-0.016	0.004	-0.032	-0.023	0.005	-0.115	-0.029	0.001	0.003	0.088	-0.148	0.007	-0.044	-0.075	-0.149	-0.52
	Pooled	0.032	0.034	0.016	0.010	0.009	0.033	0.027	0.006	-0.001	0.030	0.029	0.024	0.011	-0.078	-0.089	-0.42

Table 7: Average inter intra cluster distance of parental genotypes of Pearl millet

Clusters	I	II	III	IV	V
I	14.08	21.6	22.48	20.37	30.44
II		14.29	32.93	26.86	42.41
III			16.99	29.89	28.65
IV				18.69	32.76
V					20.27

using the toucher method. The materials were categorized into five main clusters, with Clusters I and III having the highest number of genotypes. Further division into subclusters occurred in Clusters I, II, III, and IV (Figure 1). The prevalence of genotypes within these clusters suggests a common ancestry and the potential for grouping in a single cluster. This finding

implies that parents within these clusters exhibit genetic diversity, making them suitable for hybridization programs to elicit a substantial heterotic response. The intercrossing of selected lines can establish foundational populations with desired traits, facilitating effective breeding strategies (Ramya et al., 2017; Rasitha et al., 2019; Rajpoot et al., 2023).

The average inter and intra cluster distances were displayed in Table 7. Cluster V displayed the highest intra-cluster distance (20.27), followed by Cluster IV (18.69), Cluster II (16.99), and Cluster I (14.29), while Cluster I had the lowest intra-cluster distance (14.08). This suggests that clusters with lower intra-cluster distances exhibit less genetic divergence, making selection within these clusters less effective for producing vigorous hybrids. The observed genetic diversity within clusters

may stem from factors like heterogeneity, pedigree variations, and overall combining ability. Similar findings were reported by Rajpoot *et al.* (2023). The highest inter-cluster distance was noted between Cluster II and Cluster V (42.41), followed by Cluster II and Cluster III (32.93). Parental lines within these clusters exhibit genetic diversity, potentially leading to a strong heterotic response in hybridization programs. Notably, high mean yield with resistant genotypes was found in these clusters, suggesting the suitability of designated lines for establishing foundational populations with desirable traits, as supported by studies by Ramya *et al.* (2017) and Shashibhushan *et al.* (2022).

Cluster mean values of parental genotypes of studied traits were presented in Table 8. For disease related traits, cluster IV (PT 6946, ICMB 06111) and V (ICMB 93111, ICMB 95444) had high mean values whereas grain yield and its contributing traits were recorded lower mean values. These clusters contain majority of B lines with intermediate performance in grain yield and disease related traits.

At the same time, cluster I (PT 6029, ICMB 02444) and II (GMR 58) were recorded low mean values of disease related traits and high mean values of grain yield and its attributing traits. It clearly explained that, cluster I comprised majorly restorer lines with better grain yield. Hence choosing parents from this group will act as potential breeding material to enhance the yield traits in a hybridization program (Rasitha *et al.*, 2020). Cluster III had both B lines (PT 6476, PT 6676) and R lines (ICMB 99666, ICMB 04777) with highest mean value for grain yield, and disease severity. The analysis indicated a clear separation between B and R lines, emphasizing the usefulness of morphological data in identifying promising parents for hybridization programs. This approach simplifies the preliminary evaluation of genetic materials and is universally

Table 8: Cluster mean values for different traits in Pearl millet genotypes

Traits	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V
DFF	46.63	49.67	47.73	50.67	40.4
LSL	13.28	14.48	13.35	11.77	11.76
LBL	64.24	64.35	67.03	60.23	51.99
LBW	4.45	5.42	4.4	5.2	3.78
NON	7.7	7.17	8.18	7.83	6.2
SPL	26.22	32.02	28.51	20.87	22.29
SPG	9.86	10.83	9.37	8.78	9.15
NOET	3.83	4.17	4.23	4.17	3.9
PHT	172.02	144.73	195.15	147.93	181.33
DTM	86.43	88.5	87.64	90.33	80
SEWT	35.13	38.91	38.41	30.5	24.35
SETWT	27.34	28.67	29.68	22.72	19.42
TWT	13.13	14.17	10.75	13.32	9.62
DI% (T)	30.35	18.92	33.55	41.37	41.54
DS% (T)	12.31	5.42	13.89	25.68	20.38
GYP	89.76	114.25	84.21	76.25	68.8

DFF - days to fifty per cent flowering, LSL - leaf sheath length, LBL - leaf blade length, LSW - leaf blade width, NON - Number of nodes, SPL - spike length, SPD - spike diameter, NOET - number of economic tillers/plant, DTM - Days to maturity, PLH - plant height, SEWT - Single Earhead Weight, SETWT - Single Earhead Threshed Weight, TWT - 1000 grain weight, GYP - Grain yield/Plant, DI% (T) - Disease Intensity % transformed data, DS% (T) - Disease Severity % transformed data

applicable for assessing genetic diversity among different genotypes (Shashibhushan *et al.*, 2022).

CONCLUSION

Pooled ANOVA revealed significant seasonal variation in studied materials (E_1 & E_2), highlighting resistance to blast disease and improved yield in lines like PT 6029, PT 6067, PT 6300, PT 6676, PT 6679, PT 7068, ICMB 02777, and ICMB 01666 compared to susceptible checks. Higher PCV and GCV in disease traits highlighted environmental factors, stressing the importance of genotype-environment interactions in disease resistance breeding. Negative correlations between disease and yield-contributing traits indicated blast disease's adverse impact on grain yield. Disease severity showed a positive indirect effect, suggesting an association between environmental factors conducive to disease spread and increased severity. Heritability and GMA suggested predominant additive gene action, making traits suitable for direct selection, except for economic tillers/plant and days to maturity. Association studies revealed genotypic correlations, highlighting positive correlation between quantitative traits and grain yield, especially single earhead weight and test weight. In D^2 analysis, five clusters revealed distinct genetic diversity with Clusters II and V indicating strong hybrid vigor, while Clusters IV (PT 6946, ICMB 06111) and V (ICMB 93111, ICMB 95444) excelled in disease resistance. Clusters I (PT 6029, PT 7068) and II (GMR 58) exhibited superior grain yield, particularly Cluster I, had potential restorer lines for future breeding. B and R lines validated the use of morphological data for assessing genetic diversity.

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