Genetic variability of early flowering and yield contributing traits in Barnyard Millet [*Echinochloa frumentacea* (Roxb). Link] grown at different altitudes

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Barnyard millet is an important up-surging minor millet in Asia countries for food and nutritional security. Being a minor crop, the genetics and inheritance of the traits need to understand, for effective utilization in the crop improvement programmes. Here, we investigated the genetic background and inheritance of different traits in barnyard millet genotypes. Forty germplasm accessions were evaluated in different ecological environments of southern India for 17 quantitative traits. Genetic variability analysis using GENSTAT and the pooled REML mean data results revealed that Phenotypic Co-efficient of Variation (PCV) was greater than the Genotypic Co-efficient of Variation (GCV) over the diverged ecological locations indicating the existence of environmental influence on all the biometric traits studied. In the Western Ghats region (E1), high heritability coupled with high genetic advance was exhibited for traits like days to flowering, plant height, days to maturity, number of nodes, length of nodes, number of basal tillers, stem diameter, flag leaf length, flag leaf width, inflorescence length, inflorescence width, length of peduncle, number of racemes, single ear head weight and grain yield per plant. Traits like thousand grain weight expressed moderate genetic advance with high heritability while, length of lower racemes expressed moderate heritability with high genetic advance. Similarly, high heritability coupled with high genetic advance was recorded for all the characters in valley region (E2) except thousand grain weight that showed high heritability but moderate genetic advance. In contrast, in the area of plains (E3), all the traits exhibited high heritability coupled with high genetic advance. Since, majority of the traits have shown high heritability with high genetic advance, selection might be effective in these traits irrespective of the environment.

Keywords: Genetic advance, Genetic environments, Genotypic\Phenotypic co-efficient of variation, Heritability, Western Ghats

Barnyard millet is an underexploited minor millet crop which has the cultivation history of 3000-5000 years and a major food crop in past[1]. At present, barnyard millet is grown in India, China, Japan and Korea, for human consumption as well as fodder[2,3]. India is the largest producer of barnyard millet both in terms of area (0.146 m ha) and production of (0.151 m t)[4]. This crop can be grown at the range of 2000 m height during summer season[5]. This nutrient-rich grain[6] is making a quick come back in the Indian agrarian landscape after decades of institutional neglect. This crop can grow in poor soil conditions with less water, fertilizer and pesticides and can withstand high temperatures, making it the perfect choice as climate-smart cereal[7]. The necessity of choosing early maturing crops becomes crucial

among the farmers due to climate resilience in the last couple of decades[8,9]. Further, the failure of monsoon results in wilting of crops planted at the beginning of the rainy season. Moreover, the weather extremes such as drought, heat waves and prolonged storm often cause severe damages in agro-ecosystems. Due to climate change during the 21st century, countries like Israel are expected to lead with 3-5°C rise in temperature by 2100[10]. Therefore, we are in need of new germplasm accessions with traits like early maturity and improved productivity under drought constraint to cope with the changing climatic conditions.

Genetic diversity has been eroded in many crops due to genetic bottlenecks associated with domestication and crop breeding[11]. Gene pools are an excellent reservoir of allelic diversity and source of genetic variability for crop improvement program. This is because over evolutionary time source of

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germplasm like wild species and wild relatives have gained adaptation to a wide range of environmental challenges[12]. Hence, the identification of useful genotypes with desirable introgressive traits can enhance suitability of selection criteria which can be handful for successful varietal improvement program.

For a fruitful breeding program, the analysis of variability among the traits of a crop is being essential with novel strategies. Improving the underutilized crop, gaining knowledge of variability traits could be a key to the success. Comparative characters variability is evaluated by estimating the GCV and PCV. Heritable genetic effects and non-heritable environmental influence contribute to variability presented in germplasm. The GCV flows the heritable portion, while the PCV is an expression of both genetic as well as the environmental effects on the trait. In barnyard millet, few studies on genetic variation have been documented. Higher PCV than the GCV indicates the meagre influence of environment on the expression of traits[13]. Other studies report that the information on genetic variation parameters like GCV, PCV, heritability and genetic advance is being absolute to begin an efficient breeding program[14]. In the present investigation, we analyzed the genotypic and phenotypic coefficient of variation, heritability and genetic advance for early flowering and yield attributing characters in barnyard millet over different environments.

Materials and Methods
Experimental germplasm, sites and Designs

Forty germplasm accessions including two local checks (MDU 1 and CO (Kv) 2) were used for the experiment (Suppl. Table S1. *All supplementary data are available only online along with the respective paper at NOPR repository at http://nopr.res.in*). The genotypes were obtained from the Gene Bank, International Crops Research Institute for Semi-Arid Tropics (ICRISAT), Hyderabad, India; Department of Plant Breeding and Genetics, Agricultural College and Research Institute, Tamil Nadu Agricultural University (TNAU), Madurai; Department of Millets, TNAU, Coimbatore, India and All India Coordinated Small Millets Improvement Project, Bangalore, India. The existing varieties are late maturing (95-110 days), and are susceptible to drought. The experiment was conducted in three ecologically diverged locations during summer season 2019 at Idukki, Kerala (Hilly region - E1), Theni, Tamil Nadu (Valley region - E2)

and Madurai, Tamil Nadu (Plain region - E3). The meteorological data of the distinguished environments is furnished in Table 1. The experiments were conducted following randomized complete block design with two replications. Each accession was sown on a single row 3 m length with plant-to-plant spacing of 15 cm and row-to-row spacing of 30 cm. Thinning and gap filling was carried out at 30th day after sowing to maintain optimum population. Adequate irrigation and recommended fertilizer application were provided on time with proper pest management as per the package of practices recommended by TNAU, Coimbatore (https://agritech.tnau.ac.in/agriculture/millets_kudirai vali.html).

Data collection and Analysis

The data was recorded on the seventeen biometrical traits namely days to flowering, days to maturity, plant height, number of nodes, length of internodes, number of basal tillers, stem diameter, flag leaf length, flag leaf width, length of inflorescence, inflorescence width, length of lower racemes, length of peduncle, number of racemes, single ear head weight, thousand grain weight and grain yield per plant. The morphological observations have taken based on the descriptors of IPGRI (1983). The data on quantitative traits were analysed for each environment separately and pooled the three environments using Residual Maximum Likelihood (REML)[15] in GenStat 17th edition (http://www.genstat.co.uk), considering the genotypes as random and environment as fixed. Best Linear Unbiased Predictors (BLUPs)[16] were obtained for all quantitative traits for each accession for combined of three environments. Further, the genetic components such as PCV, GCV, heritability and genetic advance were calculated[17-19].

Results and Discussion
Variance component and mean performance

The analysis of REML explained that, genotypic variance component (σ^2_g) was sizeable for all the traits in all three environments, however, the pooled data indicating the existence of acceptable variability

Table 1 — Meteorological data of three environments

Location	Date of sowing	Lati-tude	Longi-tude	Average rainfall	Average temp.
Idukki (E1)	07.01.2019	10.01^{0}N	77.34^{0}E	1082 mm	21.9°C
Theni (E2)	09.01.2019	9.93^{0}N	77.47^{0}E	791 mm	27.2°C
Maduraiv(E3)	11.01.2019	9.95^{0}N	78.01^{0}E	857 mm	28.8°C

for all the traits except for the traits such as length of nodes, length of peduncle, number of basal tillers and thousand grain weight which exhibited non-significance. A significant variance component due to genotypic and environment interaction (σ^2_{ge}) was observed for all the traits under pooled analysis, indicating the differential response of accessions to different environments (Table 2). The mean performance of barnyard millet genotypes for the quantitative traits in different environments are illustrated in Table 3 and it indicated that, plant height momentously varied in all three environments (75.86 cm in E2 to 148.47 cm in E1), while days to maturity was almost similar in all three locations (96.3.4 days E3 to 99.65 days in E2) as well as in pooled of three environments (97.66 days). Similarly, the grain yield per plant was remarkably differed between E1 (40.29 g) and E2 (23.34 g), but E3 (35.99 g) and pooled of three environments (33.20 g) exhibited almost equal data on yields. Likewise, the characters such as flag leaf length, inflorescence length, node length, length of peduncle and single ear head weight were varied significantly from E1 to E2, which divulged that the soil types, agroclimate ranges and other meteorological parameters might be impacted for the variation.

There are 12 genotypes such as IEc 71, IEc 82, IEc 107, IEc 108, IEc 109, IEc 296, IEc 350, IEc 356, IEc 385, IEc 386, IEc 391 and IEc 396, which were matured early (64-84 days) and has short plant stature (54-116 cm), while comparing other genotypes. Hence, these genotypes could be useful in the breeding programmes for developing early maturing genotypes. As far as grain yield per plant is concerned, three accessions *viz.* M5P1 (52.6 g), M36P1 (52.93 g) and M37P1 (50.86 g) recorded high grain yield per plant than checks (MDU 1 - 48.07 g and CO (Kv) 2 - 49.90 g), however, they showed no significant difference.

Estimation of Co-efficient of Variation, Heritability and Genetic advance

Improvement in breeding for yield and yield contributing traits of any crop is environmentally influenced, phylogenetically controlled and determined by the magnitude and nature of their genetic variability[20]. For trait-based genetic selection in the crop improvement program, the assessment of genetic parameters is an essential constituent for the perspective of trait characterization. The pooled BLUPs of three locations were used for further analysis. The traits such as days to flowering, stem diameter, inflorescence width, length of peduncle, number of racemes, single ear head weight and grain yield per plant[21] expressed high PCV and GCV in all the regions, while, days to maturity alone recorded moderate PCV and GCV for all three environments. In location E1, high PCV and GCV was estimated for plant height, days to flowering, number of nodes,

Table 2 — Variance components estimation for individual and pooled of three environments using REML approach

Variables	E1	E2	E3	Pooled	
	σ^2g	σ^2g	σ^2g	σ^2g	σ^2ge
Days to flowering	213.69**	213.39**	193.40**	191.18**	15.65**
Days to maturity	281.90**	292.77**	205.99**	252.00**	8.21**
Length of flag leaf (cm)	28.92**	21.69**	23.57**	15.87**	8.87**
Length of lower racemes	0.24**	0.40**	0.62**	0.16**	0.26**
Length of inflorescence	16.05**	13.65**	13.20**	10.60**	3.66**
Length of nodes	6.61**	4.90**	3.91**	1.20	3.92**
Length of peduncle	18.71**	2.66**	4.66**	1.09	7.59**
No. of tillers	15.48**	7.08**	4.48**	1.42	7.60**
No. of nodes	2.07**	2.01**	1.23**	0.84**	0.93**
No. of racemes	145.89**	143.37**	143.75**	130.31**	13.98**
Plant height	1198.72**	211.16**	827.25**	450.23**	294.88**
Stem diameter	1.42**	0.56**	1.07**	0.84**	0.18**
Single ear head weight	19.64**	4.02**	8.87**	7.89**	2.96**
Thousand grain weight	0.09**	0.11**	0.40**	0.01	0.19**
Width of flag leaf	0.29**	0.31**	0.50**	0.26**	0.11**
Width of inflorescence	0.65**	0.52**	0.57**	0.41**	0.17**
Grain yield per plant (g)	340.31**	70.44**	245.49**	172.26**	46.41**

[σ^2g, genotypic variance component; σ^2ge Genotype × environment variance component]

Table 3 — Mean performance of barnyard millet accessions at varied environments, and pooled of three environments

Trait	E1	E2	E3	Pooled
Days to flowering	63.74[ab]	65.58[a]	60.42[b]	63.24[ab]
Days to maturity	97.00[a]	99.65[a]	96.34[a]	97.66[a]
Length of flag leaf (cm)	29.03[ab]	17.68[b]	29.04[ab]	25.25[a]
Length of lower racemes	3.73[a]	2.15[b]	3.03[ab]	2.97[ab]
Length of inflorescence	20.26[a]	15.27[b]	17.28[ab]	17.60[ab]
Length of nodes	16.86[a]	8.71[b]	11.82[ab]	12.46[ab]
Length of peduncle	15.17[a]	6.33[b]	10.77[ab]	10.76[ab]
Number of tillers	16.44[a]	13.16[ab]	12.55[b]	14.05[ab]
Number of nodes	6.53[ab]	5.91[b]	7.51[a]	6.65[ab]
Number of racemes	43.17[ab]	37.43[b]	48.90[a]	43.17[ab]
Plant height	148.47[a]	75.86[b]	135.86[ab]	120.07[ab]
Stem diameter	2.77[ab]	1.95[b]	3.12[a]	2.61[ab]
Single ear head weight	9.83[a]	4.06[b]	9.68[a]	7.86[c]
Thousand grain weight	3.39[b]	3.44[b]	4.33[a]	3.72[b]
Width of flag leaf	2.76[a]	2.07[a]	2.77[a]	2.54[a]
Width of inflorescence	3.66[a]	2.54[b]	3.07[a]	3.09[a]
Grain yield per plant (g)	40.29[a]	23.34[b]	35.99[ab]	33.20[ab]

number of basal tillers, stem diameter, inflorescence width, length of peduncle, number of racemes, single ear head weight and grain yield per plant. Moderate PCV and GCV also recorded in this region for the traits like days to maturity, length of nodes, length of flag leaf and length of lower racemes. The only one trait 'thousand grain weight' recorded low PCV and GCV (Table 4). In contrast, high PCV and GCV was reported by some workers for this trait[22].

Similarly, in the location E2, the traits like days to flowering[23], number of nodes, length of nodes, number of basal tillers[24], stem diameter flag leaf length, flag leaf width, inflorescence length, inflorescence width, length of lower racemes, length of peduncle[25], number of racemes, single ear head weight, and grain yield per plant came out with the result of high PCV and GCV. Moderate PCV and GCV were also expressed only for the traits plant height and days to maturity. As observed in the E1 region, thousand grains weight similarly resulted low in PCV and GCV. Notably, no traits expressed high PCV and moderate GCV (Table 4). Considering location E3, eleven characters exhibited high PCV and GCV which cover plant height[26], days to flowering, stem diameter, flag leaf width, inflorescence length, inflorescence width, length of lower raceme, length of peduncle, number of racemes, single ear head weight and grain yield per plant[27]. Moderate PCV and GCV was expressed for the traits that includes days to maturity, number of nodes, length of nodes, number of basal tillers, flag leaf length and thousand grains weight. None of the characters was slapped into the category of low PCV and GCV. Overall, a minute difference was observed between phenotypic co-efficient of variation and genotypic co-efficient of variation which clearly

indicates the low contribution of environmental interaction in the individual locations[24]. However, comparing the different locations, the environment played a major role on genotypes and thus results significant genotype × environment interactions. The coefficient of variances is allowed only to the extent of total variability present for a character and does not divide the variability into heritable and non-heritable portions[28].

Mostly, high heritability coupled with high genetic advance was noted for majority of the traits in all three locations with slight changes in E1 as well as in the E2 regions, where, the trait thousand grain weights expressed high heritability connected with moderate genetic advance, while, length of lower racemes showed moderate heritability with high genetic advance, respectively. By contrast, higher heritability coupled with high genetic advance was noticed for all the traits studied in the E3 (Table 4). The slight changes of high and moderate heritability as well as genetic advance might be due to the factor of low temperature on filled grains/panicle at grain filling stage[29,30] and the influence of high temperature combined with difference in soil types while comparing other environments[27]. The better coupling of heritability with genetic advance must be required for gaining under selection of genotypes[31]. The traits categorized under high heritability along with high genetic advance alerts that the high heritability is most likely due to the control of additive gene effects[32]. Hence, the selection through these traits would be more effective. Interestingly, the trait days to flowering extra-ordinarily expressed its genetic variability and heritability in the diverged ecological locations due to its additive gene effect as it expressed high level of PCV, GCV, heritability and genetic

Table 4 — Estimation of genetic variability and heritability for 17 quantitative traits across environments

Traits		DF	DM	GY_P	LFL	LLR	LOI	LON	LOP	NBT	NN	NOR	PH	SD	SEW	TGW	WFL	WOI
	E 1	22.96	17.38	46.40	19.72	17.49	20.35	16.73	29.21	26.42	22.78	28.41	23.91	44.84	45.25	9.46	21.42	25.68
PCV	E 2	22.30	17.19	36.80	26.95	30.54	24.50	26.03	26.51	21.13	24.16	32.18	19.93	39.03	49.89	9.95	27.50	29.29
	E 3	23.08	14.93	43.78	17.05	26.44	21.22	17.64	20.59	17.71	15.32	24.61	21.37	33.51	31.18	15.49	25.87	24.87
	E 1	22.94	17.31	45.81	18.52	13.09	19.77	15.25	28.52	23.94	22.04	27.98	23.32	42.96	45.08	8.94	19.65	22.13
GCV	E 2	22.28	17.17	35.96	26.35	29.54	24.20	25.42	25.73	20.21	24.02	31.99	19.15	38.47	49.45	9.63	27.05	28.46
	E 3	23.02	14.90	43.54	16.72	26.00	21.03	16.74	20.05	16.87	14.80	24.52	21.17	33.20	30.77	14.55	25.52	24.61
	E 1	99.75	99.14	97.47	88.21	55.98	94.35	83.07	95.35	82.07	93.66	97.00	95.15	91.78	99.24	89.45	84.20	74.30
Herita-	E 2	99.81	99.83	95.51	95.57	93.52	97.60	95.37	94.21	91.54	98.79	98.87	92.33	97.16	98.24	93.63	96.74	94.40
bility	E 3	99.49	99.51	98.91	96.11	96.71	98.20	90.03	94.83	90.77	93.26	99.24	98.12	98.17	97.41	88.16	97.34	97.96
	E 1	47.19	35.50	93.18	35.84	20.17	39.56	28.63	57.38	44.67	43.95	56.76	46.86	84.78	92.51	17.43	37.15	39.31
Genetic	E 2	45.85	35.34	72.40	53.06	58.84	49.25	51.14	51.45	39.84	49.18	65.53	37.91	78.12	100.96	19.19	54.81	56.97
Adv.	E 3	47.30	30.61	89.20	33.77	52.68	42.92	32.72	40.23	33.11	29.44	50.32	43.20	67.77	62.56	28.14	51.87	50.18

[DF, Days to Flowering; DM, Days to Maturity; GY_P, Grain Yield per Plant; LFL, Length of Flag Leaf; LLR - Length of Inflorescence; LON, Length of Node; LOP, Length of Peduncle; NBT, Number of Basal Tillers; NN, Number of Nodes; NOR, Number of Racemes; PH, Plant Height; SD, Stem Diameter; SEW, Single Ear head Weight; TGW, Thousand Grain Weight; WFL, Width of Flag Leaf; and WOI, Width of Inflorescence. E 1, IDUKKI; E 2, THENI; and E,3 MADURAI]

advance in all three locations. Hence, the inheritance pattern for flowering time is stable over environments; this could be fruitful to select extra early flowering genotypes for introgressing early flowering genes into elite genotypes for developing extra early maturity cultivars.

Conclusion

In this study, we have used REML-MTME mixed model analysis for estimating the genetic parameters and prediction of genotype values in which genotype × environment interaction effects showed the best fit for all the traits studied. Most of the traits including early maturity have shown high heritability with high genetic advance irrespective of three environments, indicates the traits are governed by additive gene action. The identified early maturing genotypes could pave a way for developing barnyard millet with extra early maturing genotypes which would substitute cropping patterns and helps to escape from drought and also these genotypes can be utilized for hybridization programme for producing early maturity cultivars with better yield.

Conflict of interest

Authors declare no competing interests

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