

## Multi Season Evaluation of Foxtail Millet (*Setaria italica* (L.) P. Beauv.) Genotypes for Forage Yield Stability at the Nagaland Ecosystem

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### ABSTRACT

Foxtail millet is a significant crop for the ethnic tribes of Nagaland, playing a major role in their traditions. In Nagaland, most farmers cultivate foxtail millet for forage and grain purposes to maintain sustainable agriculture. Developing hybrids in foxtail millet is challenging due to the small flower morphology and the self-pollinating nature of the crop. Therefore, identifying stable forage elite variants from pure lines is a priority in foxtail millet breeding. Genotypic stability analysis conducted in this region will help to identify stable performance genotypes, which can be incorporated into crop improvement programs for developing elite lines. The current study was conducted from July 2022 to May 2023, with four different sowing dates considered as four environments. Two

environments were maintained under rainfed conditions, and the other two were irrigated. Analysis of variance showed a significant ( $p < 0.05$ ) difference among genotypes and genotype-environment interactions, while replications showed non-significant differences. Three genotypes, namely G1, exhibited constant mean performance in fodder yield across the four environments, followed by G18 and G23. AMMI biplot 1 revealed that genotypes G1, G25 and G18 exhibited stable performance among 30 foxtail millet genotypes and these results were confirmed by AMMI stability values for G18 and G25, which exhibited the lowest AMMI stability values.

**Keywords** AMMI, ASV, Foxtail millet, Forage yield, GEI.

### INTRODUCTION

Foxtail millet (*Setaria italica* (L.) P. Beauv.) is one of the minor millet crops in the Poaceae family and originated from the Yellow River in China (Ataei and Shiri 2020). Foxtail millet secure the third rank among the global millet production after sorghum and pearl millet. Projected global foxtail millet production is around 6 million tons in 2023, while India contributes half of the global production. In India, foxtail millet cultivation is around 0.8 lakh hectares with 0.6 lakh metric tons of production (Hariprasanna 2023).

Foxtail millet is a significant forage crop in the Nagaland region due to its adaptability and tolerance to drought conditions. It is used for both food and

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forage purposes. In Nagaland, most farmers cultivate foxtail millet for forage to maintain sustainable agriculture. Sustainable agriculture relies on crops that can consistently produce under varying conditions, minimizing the risk of crop failure. By identifying stable genotypes through stability analysis, farmers can cultivate varieties that ensure consistent forage production. This is essential for livestock farming and maintaining soil health.

The analysis of grain yield and adaptability of crop varieties is an important aspect for identifying suitable crop varieties for general and specific cultivation in different agricultural zones (Omran *et al.* 2022). The stability performance of genotypes across different agricultural zones has been assessed through the application of various statistical tools, such as univariate and multivariate methods (Enyew *et al.* 2021). The Additive Main Effects and Multiplicative Interaction (AMMI) is a multivariate method. The first part of AMMI, the additive part, uses analysis of variance, while the second part, the multiplicative part, employs principal component analysis to study Genotype by Environment Interaction (GEI) (Kebede *et al.* 2023). The AMMI stability value (ASV) is the interface principal component (IPCA1 and IPCA2) scores of the AMMI model (Pramanik *et al.* 2024).

The study of forage stability in foxtail millet in Nagaland is crucial due to its unique agro-climatic conditions, which differ from other regions. Evaluating foxtail millet genotypes in this area allows for the identification of varieties that are well-adapted to local environmental challenges, ensuring a stable forage yield. Therefore, the current experiment was conducted on 30 foxtail millet accessions to assess the stable forage performance of genotypes in diverse environmental conditions in the Nagaland foothills region.

## MATERIALS AND METHODS

### Experiment location

The experiment was carried out from July 2022 to May 2023, incorporating four different sowing dates, which are presented in Table 1. Each sowing date aimed to establish varied environmental conditions

during crop growth stages. Two environments were maintained under rainfed conditions, while the other two were under irrigated conditions with a seven-day interval. The experiment took place at the Research Farm of the Department of Genetics and Plant Breeding, School of Agricultural Sciences, Nagaland University, located in Medziphema, India.

### Soil sampling and analysis

In all four situations, the top 15 cm of soil were randomly selected from the field. The university lab analyzed this composite sample. The materials were dried in the shade and pulverized with a glass mortar and pestle to guarantee nutrient distribution, homogeneity, and plot representation. After sifting, the sample was tested for chemical characteristics and particle size distribution. These tests measured sand, clay, silt, pH, organic carbon (OC), nitrogen (N), potassium (K), and phosphorus. Complete soil analysis is presented in Table 2.

### Plant materials and experimental design

A total of 30 foxtail millet genotypes, including one check variety (Surya Nandi), were collected from the Indian Institute of Millets Research (IIMR), Hyderabad. These accessions were used in the current experiment to identify stable variants for forage yield performance. The list of accessions is provided in Table 3. The experiment was conducted using a Randomized Complete Block Design (RCBD) with three replications across all four environments. Each replication consisted of 30 plots (1 m × 1 m), with a 10 cm spacing between plots. Plants within rows were spaced 10 cm apart, while rows were spaced 22.5 cm apart. Recommended agricultural practices were followed throughout the experiment.

### Data collection

Dry fodder yield per plant (g) data were recorded from ten randomly selected plants in each block of every genotype in three replications.

### Statistical analysis

The data analysis was carried out using the R Studio

**Table 1.** Environment description of the experimental site. Env=Environment.

	Sowing date	Season	Latitude	Longitude	Altitude	Temperature (°C)		Humidity (%)			Year
						Min	Max	Min	Max	Rain-fall (mm)	
Env-1	01-07-2022	<i>Kharif</i>	25°45' 15.95" N	93°51' 44.71 E	310 MSL	22.30	31.66	69.64	91.75	51.92	2022
Env-2	26-07-2022	<i>Kharif</i> (Late)	25°45' 15.95" N	93°51' 44.71 E	311 MSL	22.84	32.09	69.99	92.10	55.19	2022
Env-3	01-01-2023	Summer	25°45' 15.95" N	93°51' 44.71 E	312 MSL	17.40	29.11	61.84	94.48	15.58	2023
Env-4	26-01-2023	Summer (Late)	25°45' 15.95" N	93°51' 44.71 E	313 MSL	15.97	28.28	60.11	95.29	8.46	2023

**Table 2.** Characterization of soil properties of the experimental region.

Determination	Field-1	Field-2	Field-3	Field-4
Physical analysis	Value			
Sand (%)	42.8	43.4	42.9	45.1
Silt (%)	24.9	26.7	35.1	34.5
Clay (%)	32.2	29.8	21.9	14.2
Textural classes (USDA)	Clay loam	Clay loam	Loam	Sandy loam
Chemical analysis	Value			
pH	4.68	5.49	6.48	5.74
Organic matter (%)	0.89	0.98	0.94	1.03
Available nitrogen (kg ha <sup>-1</sup> )	193.56	197.94	195.75	207.20
Available phosphorus (kg ha <sup>-1</sup> )	17.08	17.56	16.05	16.85
Available potassium (kg ha <sup>-1</sup> )	124.54	128.36	121.87	120.89

**Table 3.** List of selected genotypes based on the mean yield.

ACC No.	IC No.	Source	Code
ELS 20	IC 0621991	Andhra Pradesh	G1
FOX 4438	IC 0077702	West Bengal	G2
FOX 4394	IC0610541	Andhra Pradesh	G3
FOX 4339	IC 0597715	Andhra Pradesh	G4
ERP 82	IC 0622113	Tamil Nadu	G5
FOX 4384	IC 0610531	Andhra Pradesh	G6
FOX 4396	IC 0610543	Andhra Pradesh	G7
FOX 4403	IC 0610550	Andhra Pradesh	G8
FOX 4428	IC 0850064	Unknown	G9
ESD 79	IC 0618660	Maharashtra	G10
FOX 4336	IC 0597710	Andhra Pradesh	G11
FOX 4386	IC 0610533	Andhra Pradesh	G12
ERP 26	IC0622071	Tamil Nadu	G13

**Table 3.** Continued.

ACC No.	IC No.	Source	Code
ESD 3	IC 0618597	Maharashtra	G14
ELS 40	IC 0622003	Andhra Pradesh	G15
ERP 90	IC 0622117	Tamil Nadu	G16
FOX 4478	IC 0078006	Uttar Pradesh	G17
FOX 4489	IC 0078200	Tamil Nadu	G18
FOX 4392	IC 0610539	Andhra Pradesh	G19
FOX 4390	IC 0610537	Andhra Pradesh	G20
FOX 4330	IC 0596783	Arunachal Pradesh	G21
ESD 75	IC 0618657	Maharashtra	G22
ESD 46	IC 0618634	Maharashtra	G23
ERP 57	IC 0622094	Tamil Nadu	G24
FOX 4341	IC 0597722	Andhra Pradesh	G25
FOX 4440	IC 0077761	Gujarat	G26
FOX 4420	IC 0613573	Andhra Pradesh	G27
ELS 36	IC 0621999	Andhra Pradesh	G28
ELS 34	IC 0621998	Andhra Pradesh	G29
Surya Nandi	Check	Andhra Pradesh	G30

environment and R version 4.1.2. Stability analysis for different models with varied parameters was conducted using the “metan” package within the R Studio environment (Olivoto and Lúcio 2020).

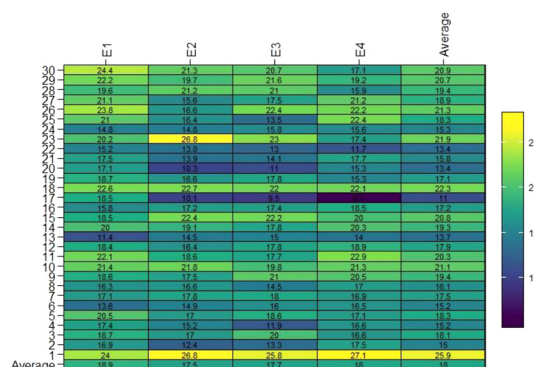
## RESULTS AND DISCUSSION

### Analysis of variance

The analysis of variance was used to assess the variation among 30 accessions across four environments for fodder yield. The results were presented in Table 4 and Fig. 1. A significant difference ( $p < 0.05$ ) was observed among the genotypes, seasons, and the interaction between year and season, indicating a sufficient amount of variation among the genotypes

**Table 4.** Analysis of variance for forage data from 30 foxtail millet genotypes grown in four diverse environments. Significant at 5%.

Source of variation	DF	Sum of squares	Mean squares	F-calculated	Significance
Seasons	3	1,274.63	424.88	474.01	0
Rep within season	8	7.17	0.90		
Genotype	29	1,044.13	36.01	36.01	0
Year×Season	87	390.61	4.49	4.49	0
Pooled error	232	231.99	1.00		
Total	359	2,948.55			



**Fig. 2.** Environmental wise treatment means.

under study. Similar findings were reported by Ataei and Shiri (2020) and Ataei *et al.* (2020).

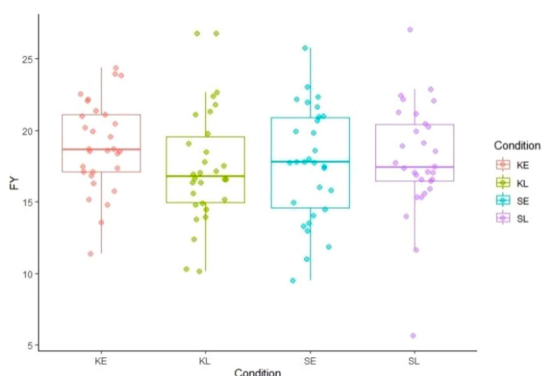
**Mean performance**

The average performance of 30 foxtail millet genotypes across four environmental conditions is detailed in Table 5 and Fig. 2. Genotype G1 consistently demonstrated high fodder yield across all environments, leading in Environment 1 (23.97 g<sup>-1</sup>), Environment 2 (26.83 g<sup>-1</sup>), Environment 3 (25.77 g<sup>-1</sup>), and Environment 4 (27.07 g<sup>-1</sup>). In Environment 1, G30 (24.36 g<sup>-1</sup>) and G26 (23.83 g<sup>-1</sup>) also showed strong performance. G23 (26.83 g) and G18 (22.70 g) excelled in Environment 2, while G23 (23.03 g<sup>-1</sup>) and G26 (22.37 g<sup>-1</sup>) performed well in Environment 3. In Environment 4, G11 (22.90 g<sup>-1</sup>) and G25 (22.43 g<sup>-1</sup>) followed G1. Overall, G1 was the top performer

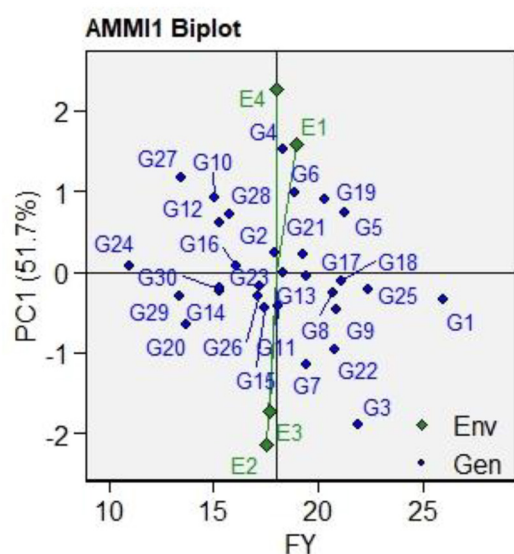
in fodder yield across all conditions, with G18 and G23 also showing notable consistency.

**Table 5.** Environmental wise treatment means of fodder yield per plant (g<sup>-1</sup>).

Sl. No.	Genotype	E1	E2	E3	E4	Mean
1	G1	23.97	26.83	25.77	27.07	25.91
2	G2	16.87	12.40	13.33	17.50	15.03
3	G3	18.73	16.97	19.97	16.57	18.06
4	G4	17.37	15.17	11.87	16.60	15.25
5	G5	20.47	17.00	18.63	17.13	18.31
6	G6	13.57	14.93	16.03	16.47	15.25
7	G7	17.10	17.80	18.03	16.90	17.46
8	G8	16.30	16.57	14.47	17.03	16.09
9	G9	18.60	17.53	20.97	20.47	19.39
10	G10	21.40	21.83	19.83	21.30	21.09
11	G11	22.10	18.57	17.73	22.90	20.33
12	G12	18.40	16.37	17.80	18.93	17.88
13	G13	11.40	14.50	14.97	14.00	13.72
14	G14	19.97	19.10	17.80	20.27	19.28
15	G15	18.53	22.43	22.17	19.97	20.78
16	G16	15.77	17.17	17.40	18.53	17.22
17	G17	18.53	10.13	9.53	5.67	10.97
18	G18	22.57	22.70	21.97	22.10	22.33
19	G19	18.73	16.60	17.77	15.33	17.11
20	G20	17.10	10.33	11.00	15.33	13.44
21	G21	17.47	13.93	14.07	17.73	15.80
22	G22	15.17	13.80	12.97	11.67	13.40
23	G23	20.20	26.83	23.03	17.37	21.86
24	G24	14.80	14.80	15.80	15.63	15.26
25	G25	21.00	16.40	13.50	22.43	18.33
26	G26	23.83	16.63	22.37	22.20	21.26
27	G27	21.13	15.63	17.47	21.17	18.85
28	G28	19.57	21.17	21.00	15.93	19.42
29	G29	22.17	19.73	21.63	19.17	20.68
30	G30	24.37	21.33	20.67	17.07	20.86
Mean		18.91	17.51	17.65	18.01	



**Fig. 1.** Box plot representation variance of thirty foxtail millet genotypes performance for fodder yield across the four environments.



**Fig. 3.** AMMI 1 biplot analysis of thirty foxtail millet genotypes over four environments.

### Additive main effects and multiplicative interaction (AMMI)

#### Analysis of variance for the additive model

The AMMI analysis of variance for fodder yield among 30 foxtail millet genotypes across four environments is detailed in Table 6. Statistically significant effects ( $p < 0.05$ ) were observed for genotypes, environments, and their interactions, while replicates had no significant impact. The genotype component accounted for the highest variance at 50.16%, followed by the genotype  $\times$  environment interaction at 18.36%, the residual component at 11.35%, en-

vironments at 1.47%, and replication at 0.31%. The genotype  $\times$  environment interaction was further analyzed and partitioned into three principal components, all significant ( $p < 0.05$ ) for fodder yield across the environments. The first principal component (PC1) alone explained 51.7% of the total variance, with PC1 and PC2 together explaining 86.9%, and PC1 to PC3 accounting for 100%. The significant effect of genotype  $\times$  environment interaction for forage yield indicates that different foxtail millet genotypes responded differently to the environments. Hence, there is scope to select the genotypes suitable for specific environments. The significant effect of the environment indicates the need to study the multi-location trial data, which can provide the opportunity to determine the stable variants suited for wide adoption. Similar results were reported by Madhavilatha *et al.* (2022) and Enyew *et al.* (2021).

#### AMMI stability biplot 1

Stability analysis of 30 foxtail millet genotypes was performed using AMMI biplot 1, presented in Fig. 3. The biplot reveals that the first principal component (PC1) accounts for 51.7% of the total variation. Figure 3, displays IPCA1 scores for both genotypes and environments, plotted against the fodder yield per plant. Numerical markers in blue denote genotypes, while green lines indicate environments. These environment lines connect to their average trait values. The biplot features a central vertical line representing the grand mean and a solid horizontal line at the IPCA1 score of 0. The x-axis shows the main effects (means) while the y-axis displays interaction effects (IPCA1). Genotypes and environments to the right

**Table 6.** AMMI analysis for fodder yield per plant ( $g^{-1}$ ) of thirty foxtail millet genotypes evaluated in four environments.

Source	Df	Sum sq	Mean sq	F value	Pr (>F)	Significant levels	Proportion %	Accumulated %
ENV	3	106.54	35.51	12.83	0	Significant	1.47	
REP(ENV)	8	22.14	2.77	0.78	0.62	Non-significant	0.31	
GEN	29	3633.24	125.28	35.35	0	Significant	50.16	
GEN: ENV	87	1329.8	15.29	4.31	0	Significant	18.36	
PC1	31	687.95	22.19	6.26	0		51.7	51.7
PC2	29	468	16.14	4.55	0		35.2	86.9
PC3	27	173.85	6.44	1.82	0.01		13.1	100
Residuals	232	822.24	3.54				11.35	
Total	446	7243.77	16.24					



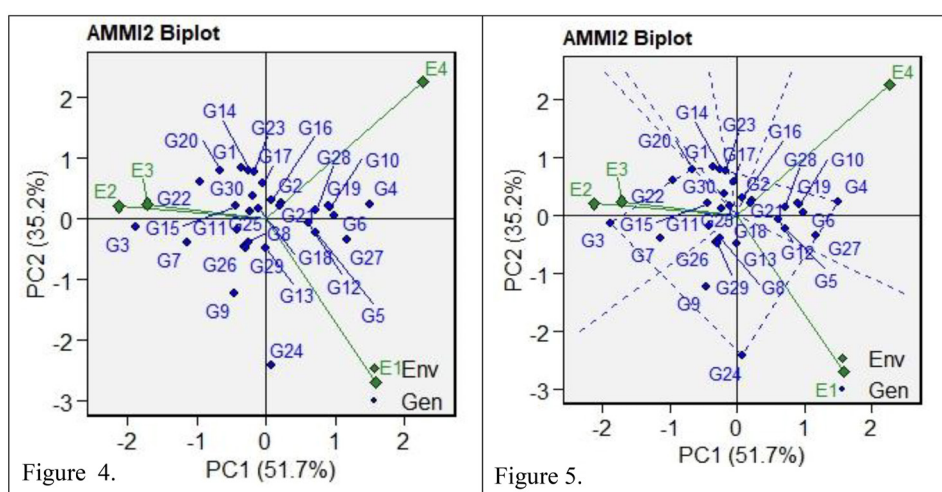


Fig. 4. AMMI 2 biplot analysis of thirty foxtail millet genotypes over four environments. Fig. 5. Polygon view of AMMI 2 biplot.

of the vertical line have higher fodder yields than the grand mean, whereas those to the left have lower yields. The intersection of the horizontal and vertical lines divides the biplot into four quadrants. Quadrants II and IV generally have more potential for higher yields compared to Quadrants I and III. Genotypes with lower IPCA1 values are considered more stable.

In the AMMI biplot, Quadrant I contain genotypes G16, G24, G2, G12, G28, G10, and G27. These genotypes have positive IPCA scores but yield below the average. Quadrant III includes genotypes G23, G30, G29, G14, G26, G15 and G20, which have negative IPCA scores and also yield below average. Genotypes G16, G24, G23, and G2 have IPCA values close to zero, indicating stability with minimal genotype  $\times$  environment interaction (GEI). However, these stable genotypes are non-adaptive and produce low yields, making them unsuitable for cultivation. Environments E2 and E3 are located in Quadrant III with negative IPCA1 values, suggesting they are less productive with below-average mean yields and thus not suitable for cultivation.

Quadrant II comprises genotypes G13, G21, G5, G19, G6 and G4, characterized by positive IPCA scores and above-average yields. Quadrant IV contains genotypes G17, G18, G8, G25, G11, G9, G7, G22, G3 and G1, which have negative IPCA

scores and above-average yields. G13, G17, G18, G1, G21 and G25 were IPCA values are closer to “Zero (0)”; hence, these genotypes are considered stable, high-yielding, adaptable, exhibit a minimum GEI interaction effect, and are recommended for general cultivation in the Nagaland region. Two environments (E1 and E4) were found in quadrant II, and these are considered productive environments due to above-average yields. Ideal genotypes are considered to have a high mean yield along with stable performance. For this effort, G1, G25 and G18 were ideal genotypes due to their high mean yield and low IPCA scores.

#### AMMI 2 stability biplot

AMMI 2 model was one the good model for analysis of stable performance of genotypes and identify the genetic variation present among the genotype and environment (Kebede *et al.* 2023). In the AMMI 2 biplot, the X-axis represents the first principal component (PC1), and the Y-axis represents the second principal component (PC2). These components capture the most significant patterns of variation in genotype  $\times$  environment interaction (GEI) (Fig. 4). This analysis helps breeders and researchers identify genotypes that perform consistently across environments and those specifically adapted to certain conditions. In the present study, AMMI biplot 2 reveals that PC1 contributes 51.7% and PC2 contributes 35.2% to the total varia-

**Table 7.** AMMI stability value (ASV) fodder yield in thirty foxtail millet genotypes over four environments.

GEN	ASV	ASV_R	GEN	ASV	ASV_R
G1	0.992	17	G16	0.328	2
G2	0.424	5	G17	0.578	9
G3	2.774	30	G18	0.225	1
G4	2.231	28	G19	1.35	21
G5	1.095	19	G20	1.253	20
G6	1.455	24	G21	0.368	4
G7	1.719	26	G22	1.523	25
G8	0.534	8	G23	0.804	14
G9	1.4	23	G24	2.43	29
G10	1.363	22	G25	0.351	3
G11	0.649	12	G26	0.643	10
G12	0.906	16	G27	1.759	27
G13	0.486	7	G28	1.063	18
G14	0.878	15	G29	0.647	11
G15	0.678	13	G30	0.474	6

tion (Fig. 4). Lines extending from the origin (0,0) to genotype or environment points show the direction and magnitude of their interactions. In this biplot, all environments (E1, E2, E3 and E4) connect to the origin. Environments E2 and E3, with shorter spokes, exhibit limited interaction strength, while E1 and E4, with longer arrows, display stronger interaction forces. Genotypes G3, G24, G27, G4, G1, G14, G20 and G22, positioned farthest from the origin, form a polygon (Fig. 5). The biplot divides into six sectors by rays extending from the origin. E2 and E3 fall into the same sector, with vertex genotypes G3, G22 and G20 indicating ideal performance in these environments. E4 falls into a sector with vertex genotypes G27 and G4, while E1 falls into another sector with vertex genotypes G24 and G9. Genotypes in sectors without associated environments are less favorable for cultivation under the specific environment conditions. Genotypes near the origin are more stable and show less interaction with the environment. According to Fig. 4, genotypes G2, G1, G25, G14, G15, G20, G23, G18 and G16, located near the center, exhibit high fodder yield stability.

#### AMMI stability value (ASV)

The AMMI Stability Value (ASV) measures the stability of genotypes for fodder yield per plant in this study. According to the ASV methodology, genotypes with the lowest ASV scores show high stability,

while those with higher scores indicate lower stability (Enyew *et al.* 2021). Table 7 presents the ASV values for 30 foxtail millet genotypes. In this study, genotype G18, with an ASV of 0.225, demonstrated the highest stability. It was followed by G16 (ASV = 0.328, rank = 2), G21 (ASV = 0.368, rank = 3), G25 (ASV = 0.351, rank = 4), and G2 (ASV = 0.424, rank = 5). These genotypes display significant stability in fodder yield per plant, indicating their consistent performance across different environmental conditions.

#### CONCLUSION

The current experiment analyzed forage data from 30 foxtail millet genotypes grown in four diverse environments in the foothills of Nagaland. Various stability analysis models were employed, and their results were compared. The findings identified Environment (E1), representing the timely *kharif* season, as the most suitable sowing environment for achieving optimal forage yield in foxtail millet. Genotypes G1 and G18 demonstrated stable forage yield performance across all four sowing environments, making them promising candidates for multi-environmental trials in this region for forage cultivation.

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