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RESEARCH ARTICLE

Assessment of Flue Cured Virginia tobacco genotype performance in the light soil tobacco growing regions based on GGE biplot and AMMI analysis

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Abstract

Flue Cured Virginia (FCV) tobacco is a major commercial crop grown as a rainfed crop in Andhra Pradesh and Karnataka states during *rabi* and *kharif* seasons, respectively. The tobacco leaf yield is often affected by aberrant weather conditions leading to significant reduction in leaf yield and quality. To identify yielding and stable FCV genotypes, two groups of genotypes were evaluated, consisting of 9 light-cast and 7 medium-cast FCV tobacco types, along with three checks, over a period of three years in the *rabi* season. The stability analysis was carried out using AMMI and GGE analysis and identified three breeding lines (KB 32, KB 50, and KB 67) in respect of stable leaf yield and quality grade index. These lines were selected on the bases of yield stability indices and genotype stability index for cured leaf yield. They were further evaluated at four locations during 2021 representing the southern light soils, northern light soils and Karnataka light soils regions of FCV tobacco growing regions of the country. The superior performance of genotype KB 67 for high leaf yield was advanced for further large-scale testing in SLS areas of Andhra Pradesh.

Keywords: FCV tobacco, AMMI, GGE biplot, cured leaf yield, grade index.

Introduction

FCV (Flue cured Virginia) tobacco (*Nicotiana tabaccum* L.) is most important commercial crop grown in Karnataka and Andhra Pradesh states. India is the second largest producer of tobacco (769 Mkg) covering an area of 0.42 Mha (FAOSTAT 2023). Tobacco crop provides employment opportunity to 45.7 million peoples and about Rs 12,005 crores of foreign exchange to national exchequer (Tobacco Board Report 2023). Tobacco production is affected by drought and water logging conditions, resulting in significant reduction of leaf yield and quality particularly in Southern light soils (SLS) and Southern black soils (SBS) areas of Andhra Pradesh.

Tobacco is grown diverse agro-climatic regions with varied temperature, rainfall pattern, sunshine hours, which all together significant influences the leaf yield and quality (Tang et al. 2020). The G×E interaction effects and stability analysis of different tobacco types for leaf yield and quality index have been reported earlier by many researchers (Bonilla 1987; Sun et al. 2013; Kurt 2020; Kurt et al. 2 021). Among the various stability parameters applied in different crops, the additive main effect and multiplicative interaction (AMMI) model and the genotype-genotype-environment (GGE) biplot procedures have been used earlier for processing multi-environmental data to determine the genotype-environment interaction (GEI) effects and to identify relationship between mean performance and stability under various environmental variables such as soil types or seasonal variations/climatic conditions (Gauch 1988; Yan 2001; Gauch et al. 2008; Heidari et al. 2016; Reddy

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et al. 2022; Patel et al. 2023; Khandelwal et al. 2024). The stability analysis for leaf yield, as well as quality grades, with respect to their effects of genotype (G), environment (E), and GE interaction is essential for identifying high-yielding and stable genotypes. The present study aimed to identify stable genotypes for leaf yield through a multi-environment evaluation of breeding lines and the selection of ideal genotypes, as assessed by GGE biplot and AMMI stability methods.

Materials and methods

Experimental details

Two experiments were conducted at the National Institute for Research on Commercial Agriculture, Research Station, Kandukur, during the rabi seasons of 2017, 2018, and 2019, following the RCBD design. The material for experiment one consists of nine light-cast FCV tobacco breeding lines, KB-45, KB-51, KB-67, KB-78, KB-86, KB-88, KB-89, KB-90 and KB-92 and experiment two consisted of seven medium-cast FCV tobacco breeding lines, namely, KB-32, KB-46, KB-50, KB-52, KB-60, KB-63 and KB-70 along with three check varieties viz., Siri, VT-1158 and N-98. Each genotype was planted in four rows in a plot size of 2.6 × 5.85 m with a spacing of 0.65 × 0.65 m. Standard agricultural practices and plant protection measures were adopted for SLS tobacco to ensure healthy crop production.

Based on the initial three-year evaluation of breeding lines at the CTRI Research Station, Kandukur, three selected breeding lines (KB 32, KB 50 and KB 67) from two sets of experiments (Exp. I and Exp. II)) were further evaluated along with respective checks at four locations, namely, Kandukur (Sothern Light Soils), Jeelugumilli (Northern Light Soils), Hunsur and Shivamogga (Karnataka Light Soils) during rainy and post rainy seasons in 2021, respectively. The meteorological data for all four locations are presented in Fig. 1. Among all the locations, Kandukur had the highest temperature, followed by Jeelugumilli, Hunsur, and Shivamogga in 2021. The prevalence of maximum sunshine hours was seen in Kandukur and the minimum was in Jeelugumilli. During *kharif*, the Shivamogga location received higher rainfall, whereas in rabi, Kandukur received high rainfall. A total of rainy days recorded at all the locations are given in Fig 1. The trials were laid out according to plot size, spacing, and the recommended package of practices followed by the respective centers. The observations on green leaf yield and cured leaf yield were recorded on nine random plants from each accession. Estimates of bright leaf and grade index were calculated as per Gopalachari (1984). Chlorides (Hanumantha Rao et al.1981), sugars and nicotine (Harvey et al.1969) were estimated from the cured leaf samples.

Statistical analysis

The phenotypic data of leaf yield per plot were collected from the three years and subjected to a combined ANOVA using a mixed linear model in R software version 4.2.1 (2020). The location-wise stable genotypes identified by AMMI analysis were assessed for significance using the Gollob (Gollob, 1968) and F-test approaches (Vargas and Crossa, 2000). Genotypes were ranked based on the AMMI stability values (Purchase et al. 2000), yield stability index (YSI) and genotype selection index (GSI) (Kang 1991,1993). Furthermore, the singular value decomposition (SVD) of the first two principal components was used to fit the model, and GGE biplots were plotted to analyze yield stability and G×E interaction effects (Yan, 2002).

Results and discussion

Preliminary yield evaluation in Kandukur in Exp. I (Southern light soils)

The combined analysis of variance (ANOVA) was carried out on the data recorded on evaluation of light-cast FCV breeding lines for leaf yield and its attributing traits from an experiment I, which showed that mean squares from genotype and environment obtained for green leaf yield, cured leaf yield, bright leaf and grade index were significant (Table 1). Significant differences were observed for genotype, environment, and G×E interaction effects for all traits studied. The green leaf yield ranged from 12.84 to 17.12 kg with an average of 15.67 kg; cured leaf yield ranged from 1.97 to 2.41 kg with an average of 2.24 kg; bright leaf

Table 1. Combined ANOVA of 12 light-cast FCV genotypes (Exp. I) across three years for leaf yield and its component traits

Effect	df	Green leaf yield	Cured leaf yield	Bright leaf	Grade index
Years	2	124.08	0.08	0.37	0.26
Replication (Years)	9	5.77	0.14	0.03	0.10
Genotype	11	24.29**	0.23**	0.11**	0.17**
Genotype x Years	22	10.05**	0.16**	0.09**	0.12**
Residual	99	1.62	0.04	0.02	0.03
Total	143	6.64	0.08	0.04	0.06
C.V. (%)		8.13	8.46	9.40	9.18
L.S.D. (0.05)		1.03	0.15	0.10	0.14

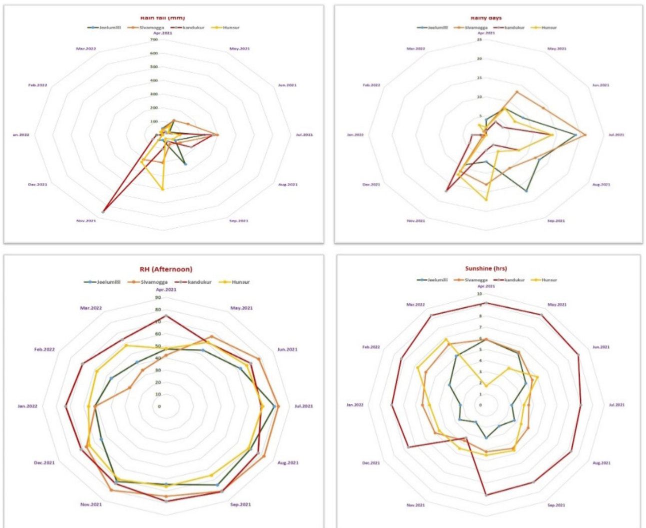

Fig. 1. Radar plot showing the weather situation at four locations of Andhra Pradesh and Karnataka

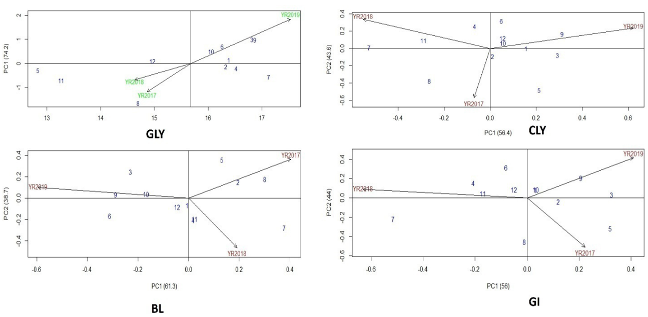

Fig. 2. AMMI Biplot analysis showing the environmental interaction of light cast FCV tobacco genotypes (Exp. I)

ranged from 1.21 to 1.54 kg and grade index ranged from 1.62 to 1.97 with an average of 1.83.

AMMI analysis

The result of ANOVA showed that the genotype, environment, and G×E interaction effects were significant for all the traits studied. The proportion of the total variance explained by genotype, environment, and G×E interaction for portion of total variance explained by genotype, environment, and G×E interaction for green leaf yield were 22.8, 21.1, and 18.81%, respectively. For cured leaf yield, genotype, G×E interaction and environmental effects explained 17.2, 23.9, and 1.13% of the total phenotypic variance, respectively. In the case of bright leaf, genotype, G×E interaction and environmental effects explained 15.8, 25.3, and 9.2% of the total variance in that order (Fig. 2). The findings obtained in the present study are consistent with the published reports of G×E interaction effects and AMMI analysis of Sadeghi et al. (2011) and Ahmed et al. (2019) for leaf yield.

The genotype's performance was assessed based on AMMI stability analysis using ASV and YSI parameters (Table 2). Genotypes G3, G6, G7, G9 and G4 had high green leaf yield and genotypes G6, G3, G4 and G9 had high mean cured leaf yield. Low G×E and stability per se should, however, not be the only criterion for selecting genotypes, because the stability of genotypes does not necessarily result in higher yield performance (Mohammadin et al., 2007a). Hence considering of rank of ASV as well as mean yield performance, a selection criterion was introduced by Kang (1991, 1993) called the genotype selection index (GSI). Two genotypesG3 and G6 (KB 67, KB 88) had high stability and higher mean leaf yield, bright leaf yield and grade index (Table 2).

Table 2. Mean performance of 12 light-cast FCV genotypes (Exp. I) across years for leaf yield based on AMMI stability value (ASV), and yield stability index (YSI)

Code No.	Genotype	Green leaf yield				Cured leaf yield				Bright leaf			
		Means	ASV	YSI	GSI	means	ASV	YSI	GSI	means	ASV	YSI	GSI
G1	KB-45	16.4	0.8	9	9	2.3	0.2	11	11	1.4	0.1	10	10
G2	KB-51	16.3	0.5	8	8	2.3	0.1	6	6	1.4	0.3	14	14
G3	KB-67	16.8	2.9	14	14	2.4	0.4	10	10	1.5	0.4	9	9
G4	KB-78	16.5	0.6	7	7	2.3	0.3	9	9	1.5	0.2	6	6
G5	KB-86	12.8	1.4	17	17	2.0	0.6	22	22	1.2	0.4	19	19
G6	KB-88	16.3	2.2	16	16	2.4	0.3	7	7	1.4	0.5	16	16
G7	KB-89	17.1	1.7	8	8	2.3	0.7	18	18	1.4	0.7	16	16
G8	KB-90	14.7	4.7	22	22	2.2	0.5	18	18	1.4	0.5	17	17
G9	KB-92	16.9	2.8	12	12	2.3	0.4	12	12	1.4	0.5	12	12
G10	SIRI (C)	16.1	1.5	14	14	2.2	0.1	11	11	1.4	0.3	11	11
G11	VT-1158 (C)	13.3	2.2	19	19	2.0	0.4	19	19	1.2	0.2	14	14
G12	N-98 (C)	15.0	0.4	10	10	2.2	0.1	13	13	1.3	0.1	12	12

GGE biplot graphical analysis

Fig. 3 showed the GGE biplot analysis of the 12 genotypes in three environments. Results of the GGE biplot showed that the first and second PCs accounted for 68.4 % and 20.9 % of the variation in green leaf yield, respectively. The GGE biplot method showed that the PC1 and PC2 could explain 53.2 and 31% of total GE variation in cured leaf yield. The biplot ranking of genotypes showed that G3, G6, G7, G9, and G4 had high green leaf yields, and G6, G3, G4, and G9 had high mean cured leaf yields. The line crossing the biplot's origin based on sections of singular values is the mean coordinate of the environment. The projection of the AEC abscissa, therefore, examines the stability of the genotypes. The genotype with the longest contract in any direction with AEC abscissa is the most stable. Two genotypes G3 (KB 67) and G6(KB 88) showed higher leaf yield performance and stability genotype among them.

Evaluation of medium cast FCV breeding lines for yield and its attributing traits (EX. II)

The combined analysis of variance over years resulted in significant differences for genotype, environment, and G×E interaction for leaf yield and quality traits (Table 3). The green leaf yield ranged from 13.11 to 16.8 kg with an average of 14.78; cured leaf yield ranged from 1.83 to 2.58 with an average of 2.17 kg and bright leaf ranged from 1.02 to 1.55 kg with an average of 1.23 kg.

AMMI analysis

The genotype, environment, and G×E interaction effects were significant for leaf yield traits studied. The proportion of the total variance explained by genotype, environment, and G×E interaction for green leaf yield were 40, 35 and 24%, respectively. For cured leaf yield, genotype, G×E interaction and environmental effects explained 47, 30 and 21% of the total phenotypic variance, respectively. In the case of bright leaf, genotype, G×E interaction and environmental effects explained 32, 43 and 24% of the total variance in that order (Fig. 4). Genotypic performance was assessed based on AMMI stability analysis using ASV, YSI and GSI parameters (Table 4). The lowest values of GSI indicate stability as well as higher yield performance. G1 (KB 32) and G3 (KB 50) showed high stability and higher mean leaf yield, bright leaf yield and grade index. Similar results of G×E interaction for leaf

Table 3. Combined ANOVA of 10 medium cast FCV genotypes (Exp. II) across three years for leaf yield and its component traits

Effect	df	Green leaf yield	Cured leaf yield	Bright leaf	Grade Index
Years	2	75.42	1.74	1.66	2.18
Replication (Years)	9	4.64	0.11	0.04	0.06
Genotype	9	19.04**	0.61**	0.27**	0.41**
Genotype x Years	18	5.67**	0.14**	0.10**	0.14**
Residual	81	1.30	0.04	0.01	0.02
Total	119	4.80	0.13	0.08	0.11
C.V. (%)		7.70	8.90	9.64	8.41
L.S.D. (0.05)		0.92	0.16	0.10	0.12

Table 4. Mean performance of 10 medium cast FCV genotypes (Exp. II) across years for leaf yield based on AMMI stability value (ASV), and yield stability index (YSI)

Code No.	Genotype	Green leaf yield				Cured leaf yield				Bright leaf			
		Means	ASV	YSI	GSI	means	ASV	YSI	GSI	means	ASV	YSI	GSI
G1	KB-32	16.8	0.3	2	2	2.5	0.3	4	4	1.4	0.4	5	5
G2	KB-46	13.5	0.4	11	11	2.1	0.3	9	9	1.1	0.3	9	9
G3	KB-50	16.7	0.7	8	8	2.6	0.3	2	2	1.6	0.7	5	5
G4	KB-52	15.3	0.5	7	7	2.3	0.5	9	9	1.2	0.8	12	12
G5	KB-60	14.9	0.9	11	11	2.2	0.7	12	12	1.2	1.3	14	14
G6	KB-63	14.8	1.4	14	14	2.1	0.9	15	15	1.1	0.9	18	18
G7	KB-70	13.1	1.5	20	20	1.8	0.7	19	19	1.0	0.4	12	12
G8	SIRI (C)	14.7	0.5	9	9	2.1	0.5	12	12	1.3	0.8	8	8
G9	VT-1158 (G)	13.6	1.2	16	16	2.0	0.7	16	16	1.2	0.9	15	15
G10	N-98 (C)	14.4	0.6	12	12	2.1	0.4	12	12	1.2	0.8	12	12

Table 5. Green leaf yield and cured leaf yield of 6 FCV tobacco genotypes at four locations of Andhra Pradesh and Karnataka

Genotype	Green leaf yield (kg/ha)				Cured leaf yield (kg/ha)			
	Jeelugumilli	Kandukur	Hunsur	Shivamogga	Jeelugumilli	Kandukur	Hunsur	Shivamogga
KB 32	11342	9252	6177	7730	1745	1439	1060	1070
KB 50	12131	10123	6652	7079	1866	1643	1147	999
KB 67	13275	11399	8558	10495	2042	1763	1605	1447
Check 1	10792	9119	8210	10199	1660	1405	1505	1429
Check 2	10983	9958	9048	10282	1690	1548	1274	1408
Check 3	11258	8730	9460	6888	1732	1528	1758	958
GM	11630	9763	8184	8779	1789	1554	1391	1218
S.EM	543	301	1250	722	83	68.5	74	103
CD (0.05)	1570	909	1250	2175	241	206	219	309
CV (%)	10.43	6.17	11.58	16	10.43	8.83	11.95	17

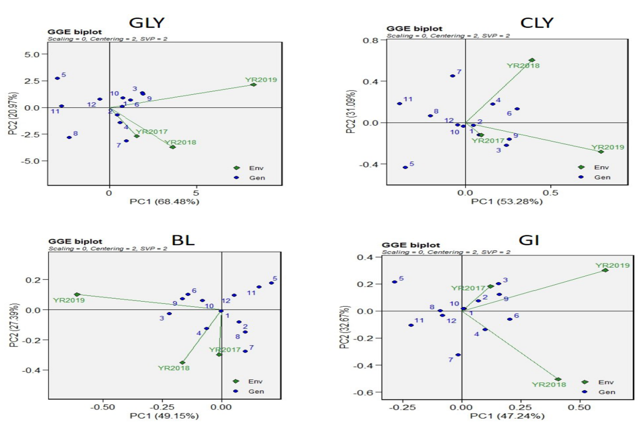

Fig. 3. GGE biplot for green leaf yield (GLY) and cured leaf yield (CLY) of light cast FCV breeding lines during 2017-19 (Exp. I)

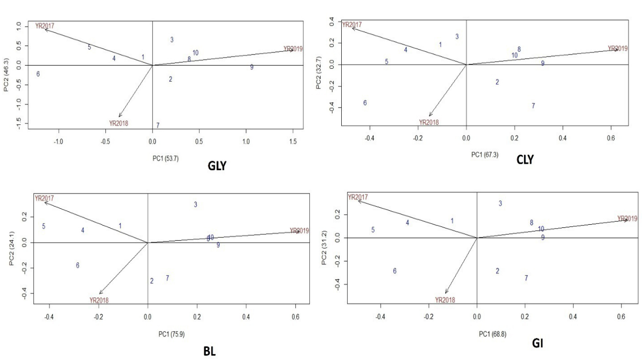

Fig. 4. AMMI Biplot analysis showing the environmental interaction of medium cast FCV tobacco genotypes (Exp.II)

yield were reported earlier (Sadeghi et al. 2011; Sun et al. 2013; Ahmed et al. 2019).

GGE biplot graphical analysis

The results of the GGE biplot for green leaf yield showed that the first and second principal components accounted for 73.4 and 19.7% of the variation, respectively. For cured

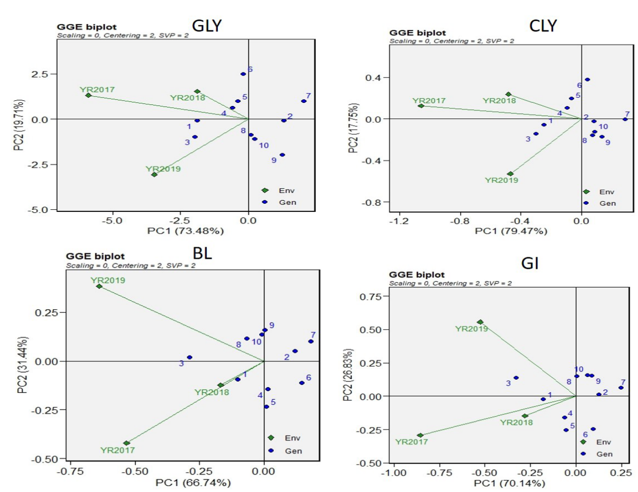

Fig. 5. GGE biplot for green leaf yield (GLY) and cured leaf yield (CLY) of medium cast FCV breeding lines during 2017-19

leaf yield, PC1 and PC2 could explain 79.4 and 17.7% of total GE variation, respectively. The biplot ranking of genotypes showed that genotypes G1, G2, G6, and G8 had high GLY, and genotypes G1, G3, G4, and G6 had high CLY. For bright leaf yield, genotypes G3, G1, G8 and G5 had high mean values (Fig. 5). The line crossing the biplot's origin based on sections of singular values is the mean coordinate of the environment. The projection of the AEC abscissa determines genotypic stability. The genotype with the longest contract in any direction with AEC abscissa is the most stable. Two genotypes, G1 (KB 32) and G3 (KB 50) showed higher leaf yield and stability among the genotypes studied.

Multi-location evaluation of three breeding lines at SLS, NLS and KLS areas of Andhra Pradesh and Karnataka

There are significant differences were observed among genotypes for green and cured leaf yield at all locations. The mean values of leaf yield characteristics at four locations are presented in Table 5. A high range of variability and mean values of green leaf yields were observed in SLS and NLS locations compared to KLS locations. The highest green leaf yield was observed inJeelugumilli (11630 kg/ha), and the lowest green leaf yield was realized in Shivamogga (6888 kg/ha). At Jeelugmilli and Kandukur, genotype KB 67 recorded significantly higher green leaf yield (13275 and 11399 kg/ha) than check (11258 and 9958 kg/ha), respectively. KB 67 recorded significantly higher cured leaf yield (2042 and 1763 kg/ha) than check (17322 and 1548 kg/ha) at Jeelugmilli and Kandukur, respectively. The superior performance with high leaf yield, KB 67 will be advanced for further large-scale testing in southern light soils (SLS) areas of Andhra Pradesh. Although KB 67 was found to have a significantly higher leaf yield in Jeelugmilli, it was not selected due to its light-cast nature, but it could serve as a potential donor for a breeding programme. The superior performance of genotype KB 67 for high leaf yield was advanced for further large-scale testing in southern light soils (SLS) areas of Andhra Pradesh.

The variability in growing environments and genetic composition have resulted in significant differences among the genotypes studied. The preferred leaf quality characteristics for each of the growing regions of FCV tobacco demand specific types, such as the cast (light, dark), curability, and flavor characteristics. Hence, region-specific breeding with more test locations within each growing region and targeting specific leaf quality requirements of different growing regions in India is suggested.

Authors' contribution

Conceptualization of research (PVVR, KG); Designing of the experiments (PVVR, KG); Contribution of experimental materials (PVVR, KG); Execution of field/lab experiments and data collection (PVVR, KG, MA, KS, CN, KS); Analysis of data and interpretation (PVVR, KG); Preparation of manuscript (PVVR, KG, MS).

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