

Identifying Stable and High-Performing WA-CMS Rice Lines through Integrated Genotype $\times$ Environment Interaction (GEI) and Multi-Trait Stability Analysis

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Abstract

Identifying widely adoptable and stable cytoplasmic male sterile (CMS) lines with superior agronomic and floral traits under multi-location testing is essential for efficient hybrid rice seed production. The present study evaluated 12 rice CMS lines across four test locations during the *Kharif 2022* cropping season to assess genotype $\times$ environment interaction (GEI), stability, adaptability, and multi-trait performance. Eleven agronomic, floral, and yield-related traits were screened among CMS lines. Joint analysis of variance revealed highly significant ($P \leq 0.01$) genotype, environment and G $\times$ E effects for all traits except panicle length and number of panicles m^{-2} at the genotypic level. The AMMI analysis revealed that environmental effects explained the largest share of variation for most phenological and morphological traits (up to 70.33% for panicle number m^{-2}). In comparison, genotypic and G $\times$ E effects were relatively more significant for grain yield (33.81% and 30.85%, respectively). AMMI Stability Value (ASV) acknowledged APMS 17A as the most stable line for grain yield (ASV = 3.956). REML/BLUP-based variance components demonstrated that all evaluated traits exhibit moderate to high heritability except panicle length and number of panicles m^{-2} , and prediction accuracy (>0.70) for most traits, supporting the reliability of BLUP-based rankings. High Genotype $\times$ Environment Interaction Ratio (GEI_r^2) was observed for number of panicles m^{-2} , panicle length, natural outcrossing, number of filled grains, and grain yield, indicating that environmental fluctuations strongly influenced these traits. WAASB and WAASBY analyses acknowledged APMS 15A, APMS 16A, APMS 17A, and APMS 14A as broadly adapted and stable across test locations, and APMS 10A combined the highest mean performance with marked environment-specific sensitivity. BLUP identified APMS 10A as a promising line. Factor analysis extracted four factors that explained 83.46% of total phenotypic variance, forming the basis of the Multi-Trait Stability Index (MTSI); APMS 17A (MTSI = 1.481) and APMS 16A (MTSI = 1.57) emerged as the most stable lines across the multi-trait complex. These results demonstrate that integrating mixed-model and multi-trait stability analyses provides a more reliable basis than any single stability model for selecting broadly adapted, high-performing WA-CMS lines for hybrid rice breeding

Key words: ANOVA, AMMI, ASV, BLUP, WAASBY and MTSI

Introduction

Rice is the major staple food for most developing and developed nations. It provides nearly 21% of the per capita dietary energy and contributes approximately 15% of the total protein intake worldwide (Abo-yousef et al., 2026). In Asia, India is the second-largest rice-producing nation after China, supporting future food security (El-Nasharty et al., 2023). Hybrid rice technology is a key breeding strategy for breaking the existing yield plateau and offering a 15–20% yield advantage over conventional rice cultivars by effectively exploiting heterosis (Singh et al., 2022). The cytoplasmic genetic male sterility (CGMS) system is dominant for rice hybrid line development worldwide, including in India (LongKumar et al., 2020). Fertility restoration of the Wild Abortive (WA) CMS system derived from *Oryza rufipogon* is governed by fertility-restoring (*Rf*) genes, which are naturally accumulated in the rice gene pool (Pushpam et al., 2019). A total of 17 *Rf* genes are reported across various CMS systems in the rice gene pool. In the WA-CMS system, two genes were identified as highly potential restoration genes, i.e., *Rf3* located on Chr-1S and *Rf4* mapped on Chr-10L (Ragi et al., 2026). The efficiency of hybrid seed production largely depends on the performance of CMS lines. Therefore, identifying and evaluating superior WA-CMS lines is important for improving hybrid seed production efficiency and developing high-yielding rice hybrids (Unan, 2021).

Although male sterility in the WA-CMS system is maternally inherited through the mitochondrial genome, it remains mostly stable across different environments (Zhang et al., 2021). However, agronomic performance and floral characteristics of CMS lines are mainly governed by the nuclear genome (Toriyama et al., 2019). They are significantly influenced by genotype $\times$ environment ($G \times E$) interactions (Casco et al., 2021). Traits such as stigma exertion, panicle exertion, flowering synchronisation, plant height, natural outcrossing rate, and seed production efficiency are highly sensitive to environmental conditions and directly determine the success of hybrid seed production (El-Namaky and van Oort, 2017). Therefore, it is important to evaluate CMS lines in various agro-climatic environments to identify genotypes with stable expression of desirable floral traits, consistent natural outcrossing ability, and dependable seed production potential (Wang et al., 2012). Despite the widespread use of WA-CMS lines in hybrid rice breeding, most previous studies have focused mainly on fertility restoration, hybrid performance, or single-environment evaluation. There has been less focus on understanding how CMS lines perform across multiple environments (Ragi et al., 2026). The complex interaction between genotypes and environments often changes how floral and agronomic traits are expressed. This makes it difficult to distinguish genetically superior CMS lines from those exhibiting environment-specific responses (Anis et al., 2024).

In multi-environment trials (METs) data analysis, genotype and genotype-by-environment biplots (GGE) and the additive main effects and multiplicative interaction model (AMMI) are commonly applied to identify stable and broad-adapted genotypes across multiple locations (Sharifi et al., 2021). However, these methods provide valuable graphical interpretation of G×E interactions; they mainly use fixed-effect models, and both models fail to account for genotypic random effects, which may reduce the accuracy and consistency of genotype evaluation under heterogeneous environments (Karimizadeh et al., 2020). Additionally, traditional linear selection indices often face multicollinearity among related traits, which leads to biased estimates and reduced selection efficiency (Ashwini et al., 2021). Principal component analysis (PCA) is an effective exploratory tool for dimensionality reduction and visualization of trait relationships in multi-environment trials. However, these methods have limited ability to accurately rank genotypes based on trait weight (Taleghani et al., 2023). To overcome these limitations, linear mixed model (LMM) based stability indices, Weighted Average of Absolute Scores (WAAS) and Weighted Average of Absolute Scores from BLUP (WAASB), offer more reliable estimates of genotypic performance by considering genetic effects alongside G×E interactions to identify stable genotypes (Sharifi et al., 2020). The Weighted Average of Absolute Scores from BLUP for Yield (WAASBY) was developed to combine mean performance with stability. This helps in identifying superior genotypes across different environments (Olivoto et al., 2019). In addition, multi-trait selection methods like the Multi-Trait Stability Index (MTSI) consider several traits, interactions between genotypes and environments, and breeding goals (Singh et al., 2025). This approach allows for more reliable identification of stable and superior CMS lines for hybrid rice breeding. However, the use of integrated multi-environment and multi-trait analytical methods to systematically evaluate WA-CMS lines remains limited. Therefore, this study aimed to explore genotype × environment interactions, assess stability and adaptability using different stability indices, and identify superior WA-CMS lines through the Multi-Trait Stability Index (MTSI) for efficient hybrid seed production and hybrid rice breeding.

Materials and methods

Plant materials and Description of locations of study:

The current investigation was carried out across four environments under an irrigated rice system during the *Kharif* season of 2022 (**Table 1**). The test centres were ANGRAU-Regional Agricultural Research Stations at Maruteru (E1) and Nanydal (E2), and ANGRAU-Agricultural Research Stations at Utukur (E3) and Nellore (E4). A total of 12 CMS lines were used in this current investigation (**Table 2**); among them, 10 CMS lines were developed by ANGRAU-Regional Agricultural Research Station, Maruteru, using indigenous germplasm, and two lines were commercial checks. The experiment was conducted in a

randomized complete block design (RCBD) with three replications across all test environments. Each experimental plot consisted of alternating rows of maintainer (B) and CMS (A) lines in a B–A–B–A–B row arrangement, with a row length of 10 m, to facilitate natural cross-pollination. Every CMS line plot was isolated by polythene tarpaulin barriers to prevent pollen contamination from nearby plots. Same barriers materials were placed between replications to maintain genetic purity and stop any unintended pollen movement among treatments. Rice seedling nursery was raised in nursery plots were transplanted in the main field with a spacing of 20 cm between rows and 15 cm between plants. Recommended agronomic practices were carried out throughout the cropping system across all test locations.

Data recorded

Data were recorded for eleven agronomic and floral traits from ten randomly selected plants of each WA-CMS line in every replication. Days to 50% flowering (DFF, days) were recorded as the number of days from sowing in the seedling nursery stage to the stage when 50% of the plants in a plot had flowered. Days to maturity (DM, days) was counted as the number of days from sowing to physiological maturity, plot-wise. Plant height (PH, cm) was measured from the ground level to the tip of the panicle (excluding awns) at maturity, while panicle length (PL, cm) was measured from the base of the peduncle to the tip of the panicle. The number of panicles per square metre (NPS, m⁻²) was recorded by counting all panicles within a 1 m² quadrat. Test weight (TSW, g) was determined by weighing 1,000 well-filled grains. The number of filled grains per panicle (NFG) was recorded by counting the total number of well-filled grains from ten randomly selected panicles in each plot and expressed as the average number of filled grains per panicle. Grain yield (GY, kg ha⁻¹) was recorded from each experimental plot at harvest after adjusting the grain moisture content to the standard level and subsequently converted to a per-hectare basis. Agronomic traits were recorded following the Standard Evaluation System (SES) for Rice (IRRI, 2014). Panicle exertion (PE, %) was calculated as the proportion of spikelets exerted from the flag leaf out of the total spikelets in a given panicle (plant) at maturity. It was calculated by using this formula (IRRI, 2014)

$$PE \% = \frac{\text{No. of exerted spikelets}}{\text{Total spikelets}} \times 100$$

Stigma Exsertion % (SE) measured as ratio of spikelets with exerted stigma (one or both side) to the total no. of spikelets in each panicle (plant). It was recorded each day until the completion of spikelet flowering in a panicle. It was calculated by using this formula (IRRI, 2014)

$$SE \% = \frac{\text{No. of spikelets with exerted stigma}}{\text{Total no. of spikelets}} \times 100$$

Natural outcrossing % (NOC) refers to the extent of seed set in panicles under natural outcrossing. It was calculated by using this formula (IRRI, 2014)

$$\text{NOC \%} = \frac{\text{No. of filled spikelets}}{\text{Total no. of spikelets}} \times 100$$

Statistical analysis

Variance analysis

A joint analysis of variance (ANOVA) was conducted across environments using the `anova_joint` function of the `metan` package in R (v.4.2.3) (Olivoto and Lúcio, 2020). This analysis assessed the significance of genotype, environment, and the interaction between genotype and environment. The significance of random effects was evaluated using the likelihood ratio test (LRT) with a two-tailed chi-square test. We examined the homogeneity of error variances across environments with Bartlett's K-squared (Arsham and Lovric, 2011), which is implemented in the `GRAPES` software (Giugno et al., 2013). Differences among environments were evaluated by one-way ANOVA, followed by Tukey's honestly significant difference (HSD) test at $P \leq 0.05$, using the `metan` package in R (v.4.2.3).

AMMI model

The AMMI model (Gauch, 1988) analyses carried for stability assessment of agronomic and floral traits. It combines ANOVA and Principal Component Analysis (PCA). The GEI was assessed using the AMMI model with the first two principal components. In addition, the model analyses the main effects of CMS lines and environment without interaction for trait data; then IPCA was unified using standardised residuals that include experimental error and GEI effect.

The analytical AMMI model equation can be written as

$$Y_{ij} = \mu + g_i + e_j + \sum_n \lambda_n \gamma_{in} \delta_{ij} + \epsilon_{ij}$$

where Y_{ij} is the mean yield of i^{th} genotype in the j^{th} environment; μ is the overall mean; g_i and e_j represent the genotype and environment deviations from the grand mean, respectively λ_n stands for the eigenvalue of principal component (IPCA) axis n . γ_{in} and δ_{ij} are the genotype and environment principal components scores for axis n and ϵ represents the error term (Gauch, 1990).

Furthermore, AMMI's stability value (ASV) was calculated in order to rank genotypes in terms of stability using the formula suggested by (Purchase et al., 2020) as shown

Below:

$$ASV = \sqrt{\left[\frac{SS_{IPCA1}}{SS_{IPCA2}} (IPCA \text{ score } 1) \right]^2 + [IPCA \text{ score } 2]^2}$$

Where SS represents the sum of squares of first (IPCA1) and second (IPCA2) interaction principal component axes; and IPCA1 and IPCA2 are the genotypic scores obtained from the AMMI model. This analysis carried out by metan package in R (v.4.2.3) (Olivoto and Lúcio, 2020).

Estimates of REML/BLUP-based variance components

Multi-environment morphological data were analyzed using linear mixed models based on the best linear unbiased prediction and restricted maximum likelihood (BLUP/REML) approach. This method is commonly used to estimate variance components and select the best genotypes in multi-environment trials (Olivoto et al., 2019; Olivoto and Lúcio, 2020). In this study, genotype, genotype $\times$ environment interaction, and incomplete blocks nested within complete replicates were considered as random effects to improve the precision of variance estimation by utilizing inter-block information. Environments and complete replicates within environments were treated as fixed effects (Olivoto et al., 2019). Variance components were estimated using the standard linear mixed model described by Yang (2007). This analysis was carried out using the metan package in R (v.4.2.3) (Olivoto and Lúcio, 2020)

Weighted average of absolute scores (WAASB)

The Weighted Average of Absolute Scores from the Best Linear Unbiased Prediction (WAASB) index evaluated the stability of CMS lines across different environments. WAASB came from the singular value decomposition (SVD) of the BLUP-based genotype $\times$ environment interaction effects. This method combined mixed-model analysis with AMMI to measure stability (Olivoto et al., 2019). Genotypes with lower WAASB values were seen as more stable across environments, while higher values showed more sensitivity to environmental changes. The WAASB analysis used the waasb() function from the metan package in R (v.4.2.3) (Olivoto and Lúcio, 2020). WAASB index calculated by used this formula:

$$WAASB_i = \frac{\sum_{k=1}^p |IPCA_{ik} \times EP_k|}{\sum_{k=1}^p EP_k}$$

$$WAASBY_i = \frac{\left\{ W_Y \times \left[\left(\frac{GY_i}{GY_{max}} \right) \times 100 \right] \right\} + \left[W_S \times \left(100 - \frac{WAASB_i}{WAASB_{min}} \right) \right]}{W_Y + W_S}$$

Where, $WAASBi$ is the weighted average of absolute scores of the i th genotype and $IPCA_{ik}$ is the score of that genotype in the k th interaction principal component axis. EP_k is the proportion of genotype $\times$ environment interaction variance explained by the k th IPCA, and p is the number of IPCAs retained.

Multi-trait stability index

The Multi-trait Stability Index (MTSI) was used to identify CMS lines that have better average performance and stability across different environments based on multiple traits at the same time (Olivoto et al., 2019a). The index was computed using the factor scores obtained after analyzing the rescaled BLUP values for all evaluated traits. MTSI measures the Euclidean distance between each genotype and an ideotype, which represents the most desirable value for each trait. The MTSI was calculated as:

$$MTSI_i = \sqrt{\sum_{j=1}^f (F_{ij} - F_j)^2}$$

where $MTSI_i$ is the multi-trait stability index of the i th genotype, F_{ij} is the score of the i th genotype for the j th factor, F_j is the score of the ideotype for the j th factor, and F is the number of retained factors. MTSI values were considered closer to the ideotype and were identified as superior for overall performance and stability across environments. The MTSI analysis was performed using the `mtsi()` function of the `metan` package in R (v.4.2.3) (Olivoto et al., 2019a; Olivoto and Lúcio, 2020).

Results and discussion

Joint analysis of variance

The joint analysis of variance was estimated for 12 CMS lines across four test locations, as illustrated in **Table 3**. The analysis showed significant differences ($p \leq 0.01$) among CMS lines (G), GEI, and test locations (E) for all eleven evaluated traits except panicle length and number of panicles per square meter, which were not significant at the genotypic level. This significant outcome indicates that there is significant variation presented among 12 CMS lines across four test locations for all studied traits. The high magnitude of significant GEI indicates that the mean performance of CMS lines rankings changes due to variations in test locations. This means the agronomic performance of CMS lines differs from one test location to another, and this significant variance is adoptable for assessment of phenotypic stability and adaptability across environments. These results align with Ragi et al., (2026), who reported a

significant GEI effect among 240 rice restorer lines. Senguttuvel et al., (2021) reported a significant genotypic effect among 72 rice CMS lines across three temperature regimes.

Mean performance of CMS lines

Mean performance of 12 CMS lines for eleven agronomic and floral traits across four test locations is illustrated in **Table 4 and supplementary Table 1**. An elite CMS line is expected to exhibit superior performance for stable grain yield and floral traits across test environments. Among 12 CMS lines, phonological characters such as days to 50% flowering expression range from 76.77 days (IR 58025A in Utukuru) to 124.38 (APMS 14A in Maruteru) with mean 99.4 days, while day to maturity range from 102.61 (IR 58025A in Utukuru) to 151.79 (APMS 14A in Maruteru) with mean 129.85 days. Variations among phonological traits reflect different maturity patterns of CMS lines within the population, and they indicate that these lines are suitable for different agroecological regions across rice hybrid breeding regions. These results align with Unan (2021) and Rani et al., (2022), who reported diverse maturity duration genotypes within the population. Morphological traits exhibited more variation than phonological traits. Plant height ranged from 54.74 cm (IR 58025A in Nellore) to 119.87 cm (APMS 16A in Maruteru), with an average of 98.23 cm, suggesting a broad range of growth habit among CMS lines. The number of panicles per square meter ranged from 218.57 (APMS 11A in Maruteru) to 849.87 (PMS 3A in Nellore), with a mean of 459.94, indicating wide variation in plant density. Panicle length ranged from 17.74 cm (IR 58025A in Nellore) to 30.1cm (APMS 14A in Maruteru), with an average of 24.57 cm, suggesting a wide variation among CMS lines for panicle length. Number of filled grains per panicle ranged from 10.67 (APMS 11A in Nandyal) to 92.96 (APMS 10A in Utukuru), with a mean of 36.93. Similar results were reported by Chen et al., (2024) and John et al., (2025) in rice.

Panicle exertion (%) ranged from 74.83% (APMS 9A in Maruteru) to 100.00% (APMS 18A in Nandyal), with an average of 88.93%. Natural outcrossing (%) expressed range from 6.94% (APMS 11A in Maruteru) to 32.51% (APMS 10A in Nellore), with a mean of 18.26%, while stigma exertion (%) ranged from 76.51% (APMS 6A in Utukuru) to 99.58% (APMS 15A in Maruteru), with an average of 90.54%. These results suggest a wide range of floral trait variation within populations. However, yield-related traits also exhibited significant variation among CMS lines. Test weight ranged from 13.4g (APMS 19A in Nandyal) to 23.79g (APMS 10A in Utukuru), with a mean of 18.5 g, while grain yield per hectare expressed ranged from 0.20 t ha⁻¹ (APMS 11A in Utukuru) to 4.59 t ha⁻¹ (APMS 10A in Utukuru), with an average of 1.51 t ha⁻¹ (Fig. 1-2). These results align with Lee et al., (2023) and Nasution et al., (2025) in rice for yield traits. APMS 10A and APMS 15A are identified as candidate CMS lines for a high heterotic combination of grain yield.

AMMI analysis

The combined analysis of variance (AMMI ANOVA) showed that the effects of environment (E), genotype (G), and genotype $\times$ environment interaction ($G \times E$) were highly significant ($P < 0.001$) for all evaluated traits across four test locations (**Table 5**). This indicates substantial variation among environments, genetic diversity among CMS lines, and differential genotype responses across environments. These results support using the AMMI model to separate and understand the $G \times E$ interaction. The environmental effect accounted for the largest proportion of the total variation in most traits, demonstrating that the environment strongly influences trait expression. The contribution of the environment effect ranged from 3.93% for grain yield ($t\ ha^{-1}$) to 70.33% for the number of panicles m^{-2} . It also explained 62.36% of the variation in panicle length, 50.66% in days to 50% flowering, 44.61% in plant height, and 40.37% in days to maturity. In contrast, grain yield ($t\ ha^{-1}$) had a relatively small environmental contribution of 3.93%. This suggests that genotypic differences and $G \times E$ interactions were more important in determining yield performance. These results align with Sharifi et al., (2021) and Sharifi et al., (2026), who reported that the environment effect was larger than the genotype and GEI effects for phenological and plant traits in an elite rice population.

Genotypic variance ranged from 2.57% for panicle length to 35.54% for panicle exertion, indicating considerable genetic variability among the CMS lines. Floral traits and yield-related traits showed a higher contribution of genotypic effects than environmental effects, suggesting that genotypic factors, rather than the environment, primarily govern phenotypic variance in these traits. Relatively high genotypic contributions were observed for grain yield per hectare (33.81) and test weight (53.19), suggesting good prospects for genetic improvement through selection. The significant GEI effect was observed for all traits, suggesting that the relative performance of CMS lines varied across environments. The GEI ranged from 9.29 % (test weight) to 30.85% (grain yield per ha^{-1}). Except for floral and yield-related traits, the remaining phenological and plant traits exhibited a 10- 20% GEI effect. These findings indicate that genotype ranking was not consistent across environments and highlight the necessity of stability analysis for identifying widely adapted CMS lines. Similar results were reported by Sing et al., (2025) and Behera et al., (2023) in an elite rice population.

According to Gollob's F-test (1968), the first two principal components (PC1 and PC2) derived from the AMMI model were statistically significant for all traits. The phenological traits, days to 50% flowering, PC1 and PC2 explained 55.8% and 28.5% of the GEI variance, respectively. Together, they accounted for 84.3% of the interaction. Similarly, for days to maturity, PC1 contributed 66.0%, while PC2 explained 21.6%. Together, they captured 87.6% of the total GEI. Among the morphological traits, plant height,

PC1 and PC2 accounted for 66.1% and 30.6% of the GEI variance, respectively, explaining 96.7% of total variance. Number of panicles m^{-2} recorded 68.8% and 22.2% of the GEI variance in PC1 and PC2, respectively, while panicle length exhibited 64.4% and 32.2%, together explaining 96.6% of the total GEI. For the floral traits, Panicle exertion showed the highest contribution of PC1 (77.5%), followed by PC2 (22.5%), together explaining 100% of the GEI variance. Similarly, stigma exertion contributed 67.7% and 28.4% of the GEI variance in PC1 and PC, respectively, making up 96.1% of the interaction. For natural outcrossing, PC1 explained 57.0%, while IPCA2 accounted for 30.0%, together capturing 87.0% of the GEI. These results show that the first two interaction principal components effectively reflected the environmental sensitivity of the floral traits related to hybrid seed production. These results agree with Abd El-Aty et al., (2024) and Abdelrahman et al., (2020) for morphological and phonological traits in rice.

For the yield and yield-related traits, such as the number of filled grains per panicle, test weight, and grain yield (t ha^{-1}), the first two PCs explained most of the GEI variation. For the number of filled grains per panicle, PC1 and PC accounted for 57.6% and 27.0% of the interaction variance, respectively, together explaining 84.6% of the GEI. Similarly, test weight showed 73.2% and 22.4% of the interaction variance in PC1 and PC2, jointly accounting for 95.6% of the GEI. For grain yield (t ha^{-1}), PC1 accounted for 65.0%, and PC2 accounted for 35.0% of the interaction variance, indicating that the first two PCs captured most of the GEI. Thus, AMMI biplots based on the first two IPCAs were enough for understanding genotype stability and adaptability in yield-related traits across different environments. These findings are supported by Cheloe et al., (2020) and Chandramohan et al., (2023) in rice.

AMMI biplots

AMMI biplots partitioned the total variance into genotype, environment, and GEI for agronomic traits (Lakew et al., 2017). In the AMMI-1 biplot (**Fig. 3a**), the first principal component axis (PCA-I scores) is plotted on the Y-axis, while the main effects (mean performance of CMS lines and test locations) are drawn on the X-axis. The CMS lines located near the PCA-I = 0 line exhibit minimal GEI effect, suggesting broad adaptability. CMS lines located far from the zero line exhibit a high magnitude of the GEI effect, suggesting adoption to a specific environment (Islam et al., 2020). Among the evaluated CMS lines, APMS 14A, APMS 16A, IR 58025A, APMS 17A, APMS 10 and APMS 15A were positioned on the right side of the biplot and exhibited higher grain yields than the grand mean of grain yield (**Fig 3a**). Among these, APMS 17A (2.10 t ha^{-1}) and APMS 15A (2.20 t ha^{-1}) were located close to the PCA-I = 0 line, suggesting high stability. These CMS lines are also considered ideal candidate lines because they performed at high yield with stability compared with the best standard IR 58025A (1.56 t ha^{-1}). The

Among test locations, Nandyal exhibited greater discriminating ability, while Nellore was located closest to the origin, indicating comparatively lower interaction effects and greater environmental stability. The CMS line APMS 10A is a winning genotype for the Utukuru location (Fig. 3b), while APMS 18 CMS lines are winning genotypes for Nandyal, and APMS 11A is a specific adoption CMS line for Maruteru.

AMMI Stability Value (ASV)

The AMMI Stability Value (ASV) effectively differentiated the stability of CMS lines for all the traits evaluated (**Table 6**). Genotypes with lower ASV values showed greater stability across environments. In contrast, higher ASV values indicated greater sensitivity to environmental changes. For phonological traits, APMS 18A secured the first rank in ASV values, suggesting greater stability across test locations. Among morphological traits, APMS 18A exhibited stable performance for plant height, while APMS 15A expressed stable performance for number of panicles m^{-2} . For panicle length, APMS 10A recorded the lowest ASV values. Among floral traits, APMS 17A recorded stable genotypes for panicle exertion (%), while for natural outcrossing (%) APMS 14A exhibited the greatest stability. For stigma exertion (%), APMS 19A was the most stable CMS line. For number of filled grains per panicle, APMS 9A exhibited the greatest stability, whereas test weight APMS 10A recorded the lowest ASV values, indicating superior stability. For grain yield per hectare, APMS 17A (ASV = 3.956) was identified as the most stable genotype, followed by APMS 16A (5.572), APMS 19A (7.279), and APMS 15A (7.441). Conversely, APMS 10A (78.388) exhibited the highest ASV, followed by APMS 18A (52.609) and APMS 11A (27.253), indicating strong genotype $\times$ environment interaction and poor stability for grain yield. These results are supported by Habib et al., (2024) and Ahmed et al., (2024) in rice.

Estimates of variance components

The BLUP/REML-based variance components analysis of eleven agronomic and floral traits among 12 CMS lines across four test locations was presented in **Table 7 and Fig. 4**. The likelihood ratio test (LRT) against the chi-square distribution indicated highly significant ($P < 0.05$) of genotypic and genotype $\times$ environment ($G \times E$) interaction effects for all studied traits, except for the genotypic effect on the number of panicles m^{-2} (**supplementary Table 2**). These results confirm that there is usable genetic variation among the CMS lines and substantial differential responses across environments, justifying the use of the REML/BLUP mixed-model approach for accurate estimation of variance components and prediction of genotypic performance.

Genetic parameters provide the basis for effective selection in plant breeding (Paiva et al., 2026). Heritability (h^2) is a key attribute for quantifying the heritable phenotypic variance (Hossain et al., 2025).

Resende (2002) reported heritability classifications in annual species as low (<0.15), moderate ($0.15-0.50$), and high (>0.50). Pereira et al., (2013) classifies heritability as low (<0.15), moderate ($0.15-0.50$), and high (>0.50) in both annual and perennial species. High heritability means that trait expression is mainly controlled by genetic factors, with less influence from the environment. This improves the efficiency of selection based on predicted genotypic values rather than observed phenotypes (Ferreira et al., 2025). In the present investigation, heritability (h^2) ranged from 0.05% for panicle length to 0.77% for test weight. All evaluated traits exhibit moderate to high heritability except panicle length and number of panicles m^{-2} , which show low heritability. These results suggest that genetic factors predominantly control these traits and can be effectively improved through selection. Traits with low heritability reflect greater influence of environmental factors (Devi et al., 2026). The moderate heritability observed for grain yield indicates that, although environmental effects substantially influence trait expression, sufficient genetic variability exists to achieve effective genetic improvement through selection (Hasan et al., 2025). The low heritability found for some traits in this study shows that environmental factors played a bigger role in phenotypic variation than genetic effects (Santos et al., 2015). Nevertheless, deResende (2002) suggested that heritability estimates of approximately 20% are still adequate for achieving genetic gain in economically important quantitative traits through appropriate selection strategies. These results are consistently aligned with Devi et al., (2026) and Mukul et al., (2025) in rice for yield traits.

Grain yield ($t\ ha^{-1}$) showed the highest phenotypic variance, followed by the number of panicles m^{-2} and the number of filled grains. Yield-related traits therefore capture the greatest phenotypic variability, while panicle length shows the lowest variance among the traits evaluated. This substantial variability in yield and related traits offers significant potential for selecting superior genotypes. Similar results were reported by Hossain et al., (2025) in rice. In this study, the highest heritability of genotype means (h^2mg) was observed for most traits except the number of panicles m^{-2} and panicle length, with values ranging from 0.01 for panicle length to 0.942 for test weight. These high h^2mg values indicate reliable genotypic mean estimates in replicated multi-environment trials, improving the accuracy of genotype ranking and the efficiency of selection based on REML/BLUP-predicted genetic values. Alves et al., (2024) also found that high h^2mg supports reliable genotype prediction and selection in rice population.

The proportion of genotype $\times$ environment interaction variance (GEI_r^2) ranged from 0.170 for test weight to 0.876 for number of panicles m^{-2} . High GEI_r^2 values for number of panicles m^{-2} , panicle length, natural outcrossing, number of filled grains, and grain yield indicate that these traits were strongly influenced by environmental conditions and genotype $\times$ environment interaction. They need to be evaluated in different settings to assess their stability and adaptability properly. Olivoto et al., (2019) reported similar findings. They emphasized the value of WAASBY-based stability analysis for traits with strong $G \times E$ interaction.

Conversely, test weight, panicle exertion and stigma exertion were comparatively less affected by environmental variation. The prediction accuracy of BLUP estimates was high (>0.70) for all evaluated traits except number of panicles m^{-2} and panicle length. The highest accuracy was observed for test weight, followed by plant height and days to 50% flowering. Accuracy >0.70 is considered reliable for genotype selection. These high accuracy values indicate that the predicted genotypic values are reliable for genotype selection (Ferreira et al., 2025).

The genotypic correlation (r_{ge}) demonstrated the alignment between predicted and true genetic values, thereby reflecting the accuracy of genotype prediction (Vigneshwari et al., 2025). In the present study, high genotypic correlation (r_{ge}) was observed for number of filled grains, natural outcrossing, grain yield, and number of panicles m^{-2} , indicating high prediction accuracy and reliable ranking of the WA-CMS lines for these traits. In contrast, panicle exertion and stigma exertion exhibited relatively lower r_{ge} values, suggesting comparatively lower prediction accuracy, which may be attributed to greater residual variation and environmental influence. The genotypic coefficient of variation (CVg) was highest for grain yield, followed by number of filled grains and natural outcrossing, indicating substantial exploitable genetic variability for these traits. In contrast, stigma exertion, days to maturity, panicle length and panicle exertion exhibited relatively lower genetic variability.

The genotypic coefficient of variation (CVg) and the residual coefficient of variation (CVr) provide important information on how much genetic and residual variation contributes to trait expression. A high CVg along with acceptable CVr values suggests good prospects for genetic improvement through selection, as noted by Behera et al., (2024). The coefficient of relative variation (CVg/CVr) values greater than one considered favourable for effective selection (Naveen et al., 2025). In the present study, all traits, except panicle length, exhibited a CV ratio greater than one, indicating that genetic variation exceeded residual variation. The highest CV ratio was found for the number of filled grains (10.80), followed by natural outcrossing (7.10), grain yield (5.33), and test weight (3.57). This indicates a strong potential for genetic improvement through selection.

Among the evaluated traits, test weight and plant height demonstrated high heritability, strong genotypic variance, prediction accuracy, and a low magnitude of GEIr2, indicating strong genetic control. Traits like grain yield, number of filled grains, and natural outcrossing showed high genetic variability coupled with favourable CV ratios but were also associated with substantial genotype $\times$ environment interaction, highlighting the importance of stability analyses such as WAASB, WAASBY, and MTSI for identifying stable and high-performing WA-CMS lines across environments. However, the numbers of panicles m^{-2}

and panicle length were more strongly influenced by environmental effects and therefore require careful evaluation across multiple environments before selection.

WAASB biplot

In the circumstance of genotype $\times$ environment interaction (GEI) analysis, the WAASB (Weighted Average of Absolute Scores from BLUP) biplot provides a more robust informative framework compared to the conventional AMMI (Additive Main effects and Multiplicative Interaction) model, which largely interprets genotype $\times$ environment interaction ($G \times E$) using the first interaction principal component (IPCA1). At the same time, the WAASB approach integrates all significant interaction principal component axes from the BLUP-based $G \times E$ matrix (Naveen et al., 2025 and Sanku et al., 2026).

The WAASB biplot analysis of 12 CMS lines across eleven agronomic and floral traits at four test locations is shown in **Supplementary Fig. 1a–j**. Among yield traits, CMS lines APMS 15A, APMS 14A, APMS 16A and APMS 17A were located in Quadrant IV of the biplots for grain yield per hectare (**Fig 5**) and number of filled grains (**S. Fig 1g**), indicating high grain yield combined with low WAASB values and, consequently, superior stability across test locations. The WAASB biplot for number of panicles per square meter (**S. Fig 1d**) revealed that APMS 15A, APMS 11A, APMS 9A, APMS 19A, and APMS 18A were also located in Quadrant IV, demonstrating above-average panicle number with strong stability. For test weight (**S. Fig 1j**), APMS 10A, APMS 11A, APMS 9A, PMS 3A, and IR 58025A were identified as superior genotypes, as they exhibited higher test weight and lower WAASB values, and were situated in Quadrant IV. Across the four yield trait WAASB biplots, CMS lines APMS 15A, APMS 16A, APMS 17A, and APMS 14A consistently demonstrated high mean performance and low WAASB values for major yield-related traits, reflecting broad adaptation and high stability across test locations. In contrast, APMS 10A consistently showed superior mean performance but higher WAASB values, indicating specific adaptation and increased sensitivity to environmental variation.

The WAASB biplot for plant height (**S. Fig. 1c**) showed that CMS line APMS 11A was located in Quadrant III, which indicates below-average plant height but relatively stable performance. For panicle length (**S. Fig. 1e**), APMS 10A and APMS 18A exhibited longer panicles and greater stability across test locations. Among phenological traits (**S. Fig. 1a-b**), CMS lines APMS 18A, APMS 17A, APMS 16A, APMS 15A, and APMS 19A were located in Quadrant IV, suggesting later flowering with high stability. IR 58025A was positioned in Quadrant I, reflecting early maturity but poor stability across environments. For natural outcrossing (**S. Fig. 1h**), CMS lines APMS 14A, APMS 17A, IR 58025A, and APMS 15A were identified as the most outcrossed genotypes, as they were positioned in Quadrant IV, combining above-average natural outcrossing with high stability across test locations. Although APMS 10A recorded

the highest natural outcrossing percentage, it was located in Quadrant II, indicating high performance but greater genotype $\times$ environment interaction and lower stability. APMS 16A demonstrated good stability but only moderate outcrossing ability. For panicle exertion (S. Fig. 1f), CMS lines APMS 17A, APMS 10A, APMS 18A, and APMS 16A showed stable performance across test locations. The WAASB biplot for stigma exertion (S. Fig. 1i) identified APMS 15A, APMS 14A, and APMS 16A as desirable genotypes because they combined above-average stigma exertion with low WAASB values. Overall, APMS 15A, APMS 16A, APMS 17A, and APMS 14A consistently occupied Quadrant IV for most agronomic, floral, and yield-related traits, indicating broad adaptation and stable performance across test locations.

The WAASB biplots also partitioned the test locations according to their contribution to GEI. The Nellore exhibited the lowest WAASB values for grain yield, plant height, panicle exertion, and natural outcrossing, indicating a relatively stable testing environment. Nandyal was comparatively stable for days to 50% flowering, days to maturity, number of filled grains, and number of panicles m^{-2} , whereas Maruteru provided greater stability for stigma exertion. In contrast, Utukur consistently exhibited higher WAASB values for several traits, indicating greater environmental discrimination and stronger genotype $\times$ environment interaction. Likewise, Maruteru showed high WAASB values for plant height, panicle exertion, and stigma exertion, suggesting its usefulness in distinguishing genotype performance under contrasting environmental conditions.

Overall, the WAASB analysis demonstrated that CMS lines such as APMS 15A, APMS 16A, APMS 17A, and APMS 14A emerged as the most stable and widely adapted across test locations, with suitability for general adoption in hybrid rice breeding programmes. The Utukur and Maruteru served as highly discriminating environments for evaluating genotype $\times$ environment interaction. These findings confirm the usefulness of the WAASB approach for selecting stable parental lines with superior agronomic, floral, and yield performance across diverse environments. APMS 10A and APMS 18A exhibited superior performance for several traits but also showed high WAASB values, indicating specific adaptation and sensitivity to environmental fluctuations. These CMS lines may therefore be recommended for targeted environments where their yield potential can be fully exploited. Comparable findings were reported by Sharifi et al., (2020), Hossain et al., (2025) and Devi et al., (2026) in rice.

Best Linear Unbiased Prediction (BLUP)

Best Linear Unbiased Prediction (BLUP) biplots are widely used in plant breeding to evaluate genotype performance by simultaneously accounting for fixed and random effects in multi-environment trials. In the present study, the BLUP biplots (Supplementary Fig. 2a–j) effectively distinguished the CMS lines

based on their predicted genetic values for the evaluated agronomic, floral, and yield-related traits. CMS lines such as APMS 14A and APMS 11A exhibited high predicted mean values for phenological traits (S. Fig. 2a-b), while APMS 16A showed a high predicted mean value for plant height (S. Fig. 2c). Similarly, CMS lines of standers such as PMS 3A and IR58025A exhibited the best predicted mean values for panicle length (S. Fig. 2e) and number of panicles m^{-2} (S. Fig. 2d). Whereas, CMS line APMS 10A recorded the highest predicted genotypic values for grain yield (Fig.6), natural outcrossing (S. Fig. 2h) and test weight (S. Fig 2j). At the same time, APMS 15A showed higher predicted genotypic values for stigma extraction (S. Fig. 2i) and number of filled grains (S. Fig. 2g). APMS 18A recorded the best predicted genotypic value for panicle extraction (S. Fig. 2f). Overall, the BLUP analysis identified APMS 10A, APMS 15A, APMS 17A, APMS 14A, and APMS 16A as the most promising WA-CMS lines because they consistently exhibited above-average predicted genetic values for grain yield and several key agronomic and floral traits. Similar findings were reported by Wang et al., (2017), Huang et al., (2019) and Abdelkhalek et al., (2025) in rice elite populations.

WAASB × Yield heat map

The WAASBY heat map (Fig. 7a-b) illustrated changes in CMS line rankings across different combinations of yield performance and stability (WAASB) across test locations. The major advantage of the WAASB approach is its ability to categorize genotypes by integrating the contribution of all significant interaction principal component axes (IPCA), including those not represented in conventional AMMI1 and AMMI2 biplots (Olivoto et al., 2019). Figure 7b shows how assigning different weights to grain yield (GY) and stability (WAASB) affects the classification of WA-CMS lines using the WAASBY index. The weighting ratio ranged from 100:0 (stability only) to 0:100 (grain yield only). Each 5% increase in the weight for grain yield came with a 5% decrease in the weight for stability. This analysis allows for the evaluation of both productivity and stability at the same time. It also lets breeders adjust genotype rankings based on specific goals, helping to identify CMS lines that are best for either broad or targeted adaptation.

When the first and second IPCAs were deployed in WAASB assessment, APMS 17A, APMS 19A, and APMS 16A (blue colour bars in Fig. 7a) were demonstrated as top CMS lines across all WAASB × yield weighting ratios, demonstrating an excellent balance between grain yield and stability and confirming their broad adaptation across test locations. APMS 10 exhibited the poorest stability (yellow colour bar in Fig. 7a) across test environments. However, when more than three IPCAs were applied, APMS 17A, APMS 16A, and APMS 15A maintained consistently superior rankings regardless of the weighting ratio (100:0 to 0:100) at WAASB × Y bilpot (blue colour bars in Fig. 7b), indicating stable grain yield and low

genotype $\times$ environment interaction. However, the ranking of APMS 10A differs quite a bit from the weighting ratio. It ranked among the poorest genotypes when stability received greater emphasis (100:0 and 95:5), but progressively improved as the weight assigned to grain yield increased, ultimately becoming the highest-ranked genotype under the 0:100 ratio. This pattern indicates that APMS 10A possesses excellent yield potential but relatively poor stability across test locations. Similar studies reported by Adilakshmi et al., (2026) in sugarcane and Karimizadeh et al., (2026) in lentil.

Multi-trait Stability Index based Selection

The ranking of 12 CMS lines, based on their MTSI (Multi-trait Stability Index) scores, is presented in **Table 8**. Two CMS lines, namely APMS 17A (MTSI=1.481) and APMS 16A (MTSI=1.57), were identified as stable-expressed lines in terms of multi-trait combinations across four test locations.

Factor analysis

Factor analysis of 12 CMS lines investigated for eleven agronomic and floral traits across four test locations retained four factors, with eigenvalues exceeding 1 and cumulatively accounting for 83.46% of the total phenotypic variance (Table 9). This suggests that these four factors effectively capture a large portion of the variance in the dataset, and this magnitude of variance is meaningful for statistical robustness and biological interpretability (Rahman, 2025).

The factor loadings fell between -1 and $+1$. Both the size and direction of each loading showed how strong and what type of relationship existed between a trait and its factor (Habib et al., 2024). The communality (h^2) values ranged from 0.63 to 0.95. This suggests that the chosen factors explained a significant amount of the variation in the traits studied. Among these traits, days to 50% flowering, days to maturity, and number of filled grains had the highest communality values ($h^2 = 0.95$). This means the extracted factor structure well represented these traits.

On the other hand, test weight and stigma exertion had lower communality values (around 0.63). This indicates that more of their variation was unique to the traits and not fully captured by the retained factors. The extracted factor structure formed the basis for calculating the Multi-Trait Stability Index (MTSI) by grouping correlated traits into independent latent factors, thereby minimizing multicollinearity and redundancy while improving the efficiency of multi-trait genotype selection (Habib et al., 2024). FA1 correlated to phenological traits and plant height. FA2 was associated with yield-related traits (GY, NFG) and natural outcrossing (%). FA3 related to floral traits (PE, SE, PL). FA4 consisted of test weight and number of panicles m^{-2} . A similar pattern of results was reported by Hossain et al., (2025) and Sharifi et al., (2020) in rice genotypes.

Selection differentials and predicted genetic gains

The selection differentials (SD) and predicted selection gains (SG) for eleven agronomic and floral traits were estimated and presented in **Table 9** and **Fig. 8**. Selection differentials (SD) ranged from –8.64% for test weight to 17.08% for grain yield per hectare, with an average of 3.22%. Among the evaluated traits, the greatest expected selection differential (SD) was observed for grain yield (24.54%), followed by number of filled grains (22.76%), plant height (5.33%), and natural outcrossing (7.29%), indicating substantial improvement in these economically important traits after selection. Moderate gains were obtained for panicle exertion (1.92%), days to maturity (0.50%), and stigma exertion (0.75%), whereas panicle length showed a negligible selection differential (0.18%), indicating limited genetic improvement for this trait. Predicted selection gain (SG%) ranged from -9.18% for test weight to 24.54% for grain yield per hectare, with a mean of 4.85% and cumulative gain of 68.71% across all evaluated traits. The predicted selection gain (SG) followed a similar trend.

The MTSI analysis demonstrates that simultaneous selection of WA-CMS lines is expected to produce the greatest genetic improvement in grain yield, number of filled grains, natural outcrossing, and plant height, while maintaining favorable performance for phenological and floral traits. The negligible gain for panicle length suggests limited genetic variability and low selection efficiency for this trait. Overall, the MTSI approach brought together several related traits and found CMS lines with better productivity, floral features, and stability. This makes it a useful tool for choosing top parental lines for hybrid rice breeding. Compression among above stability models APMS 15A, APMS 16A, APMS 17A and APMS 14A (Table 10) were identified as promising CMS lines across four test locations.

Conclusion

The findings showed that genotype $\times$ environment interactions strongly influenced grain yield and various floral characteristics affecting seed set in WA-CMS lines, emphasising the need for multi-environment and model stability analyses before releasing the parental lines. Across AMMI/ASV, REML/BLUP, WAASB and WAASBY analyses, APMS 15A, APMS 16A, APMS 17A and APMS 14A repeatedly exhibited outstanding yields with low levels of environmental sensitivity. In contrast, APMS 10A represented the typical yield-stability trade-off, where good yield was accompanied by poor stability, making the line more appropriate for specific rather than broad deployment. The convergence of these single-trait stability measures with the multi-trait stability index, which identified APMS 17A and APMS 16A as the top-ranked lines, provides strong, cross-validated evidence that these two WA-CMS lines are the most promising candidates for broad-based hybrid rice breeding programmes. More broadly, the findings indicate that none of these stability models can function independently; fixed-effect models like

AMMI are ideal for visualisations, random-effect models like REML/BLUP and WAASB(Y) are better in terms of ranking accuracy, and MTSI is required whenever the breeding target involves improving yield and other agronomic characters simultaneously. It is suggested that this combined approach to stability analysis be employed in future assessment and development of WA-CMS lines within hybrid rice breeding programs.

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Author contributions statement

Conceptualization of the study, development of WA-CMS lines, designing and supervision of the research work, monitoring of all phases of the multi-environment trials, collection and analysis of data, interpretation of results, critical review and approval of the manuscript was done by BNVS RK. Development, improvement and maintenance of WA-CMS lines were done by PVS NR and YS. Coordination and conduct of the multi-environment trial were carried out by CSL and PLV respectively. Collection of experimental data, statistical analysis and interpretation of results, construction of figures and tables and drafting of the manuscript were done by DPR, KANT and CVKR. Contribution to the development, maintenance and improvement of WA-CMS lines and technical support during the study was provided by PVRR, MGR, N. NCMSWRI and TS.

Data availability statement: The datasets used and/or analyzed during the current study are available in this published article and supplementary material. Further information about the data underlying the results presented in this study is available from the corresponding author on request.

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Fig 1. Representative panicles of superior WA-CMS (A) lines and their corresponding maintainer (B) lines identified through multi-environment evaluation.

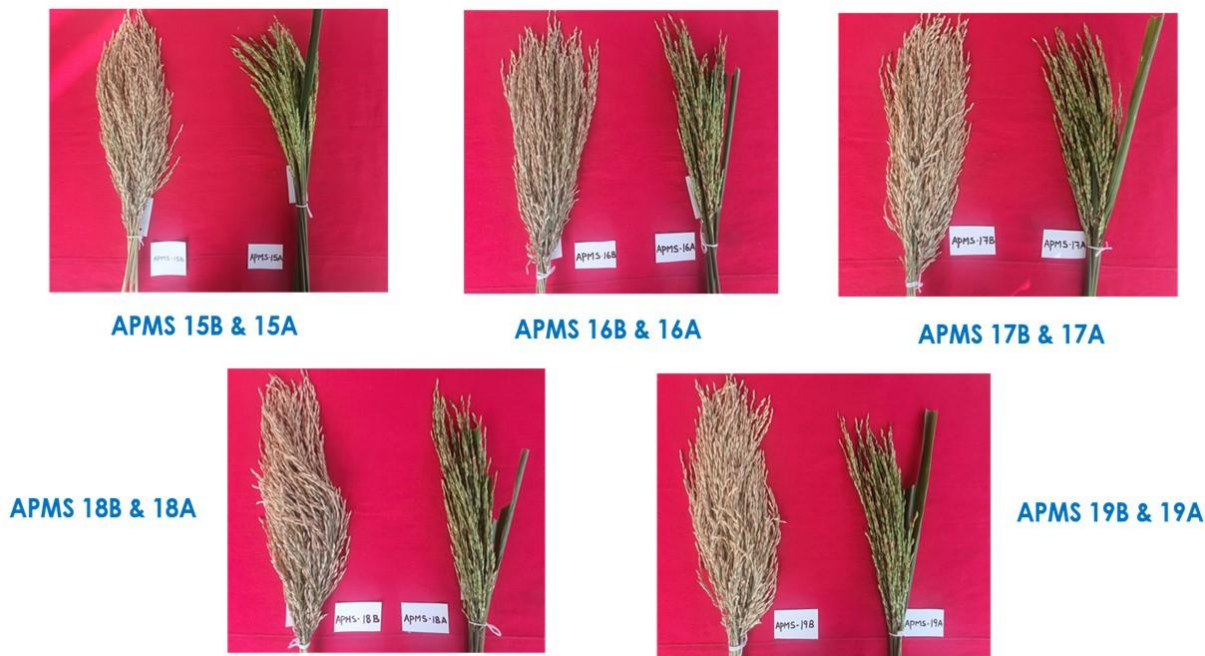


Fig 2. Grain yield mean performance of 12 CMS lines across four test locations

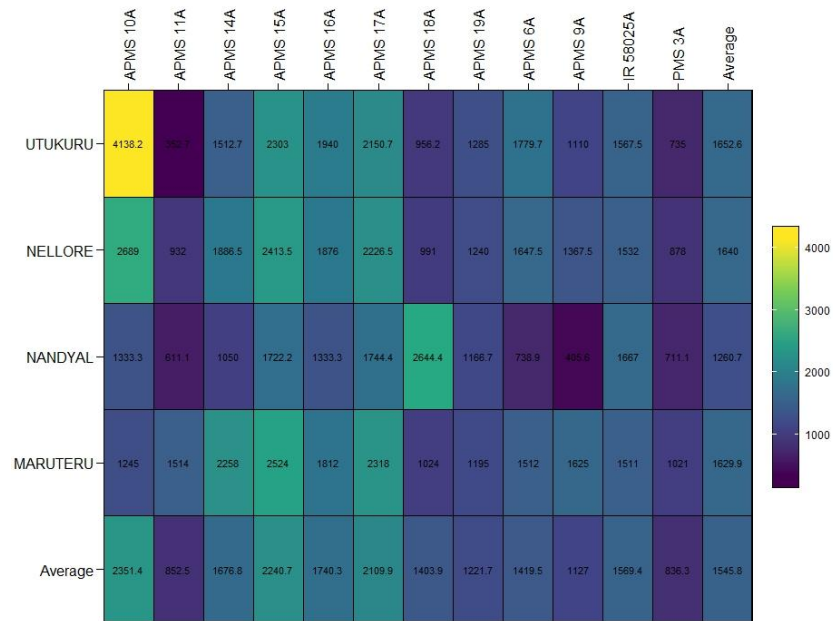


Fig 3a. AMMI1 biplots for grain yield per hectare among 12 CMS lines over four test locaions

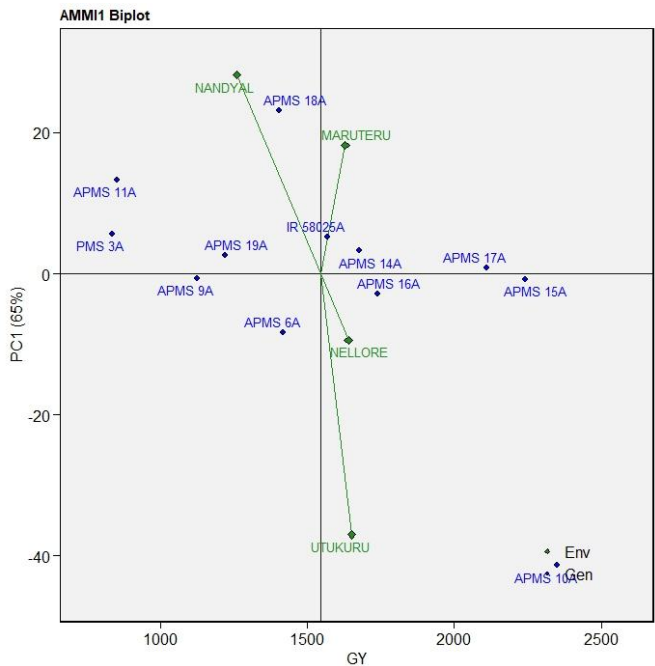


Fig 3b. AMMI2 biplots for grain yield per hectare among 12 CMS lines over four test locaions

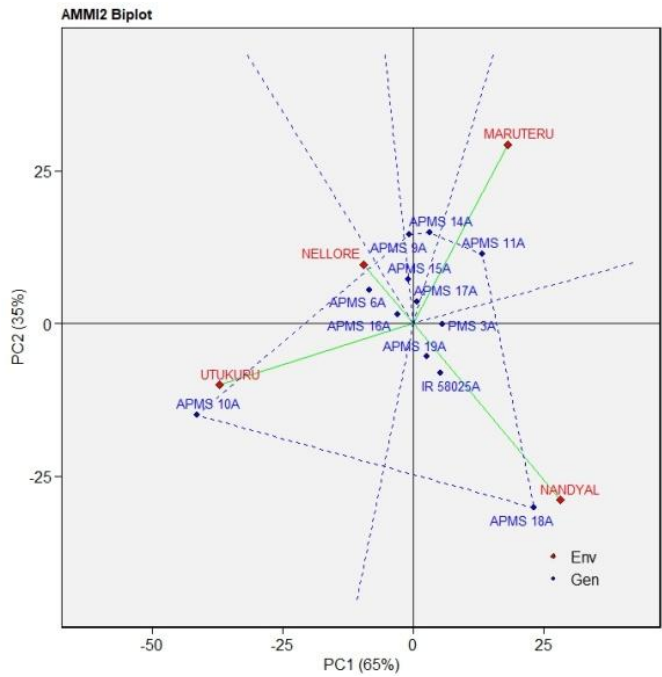


Fig 4. Proportion of phenotypic variance across four test locations among 12 CMS lines for eleven agronomic and floral traits

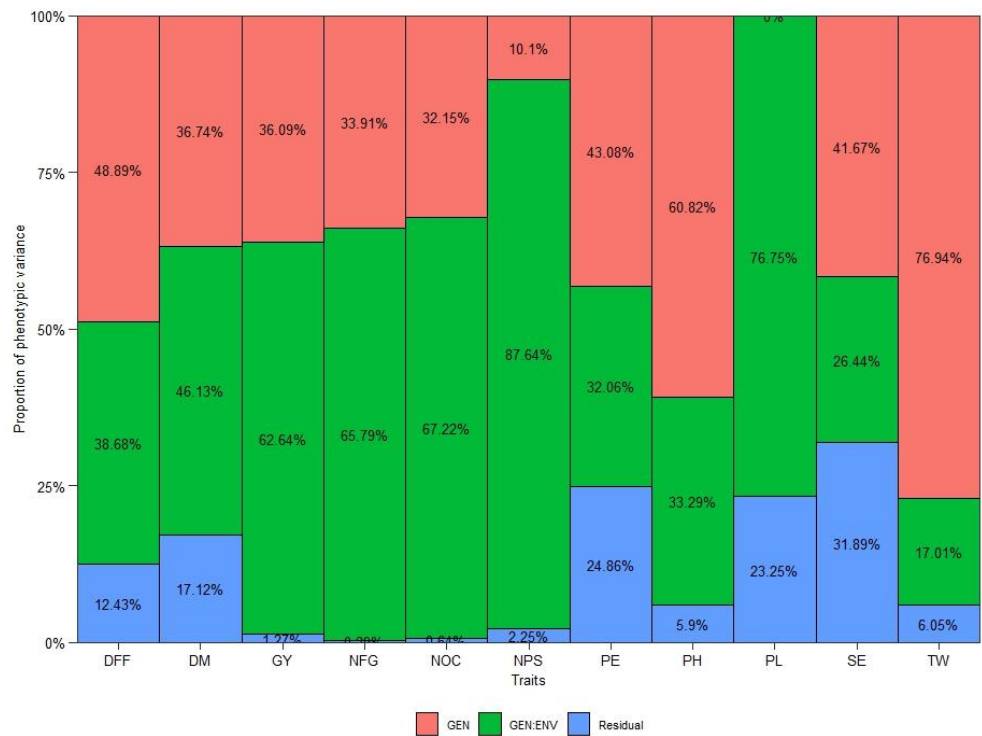


Fig 5. Estimates of the weighted average of stability (WAASB) and mean performance (WAASBY) for grain yield among 12 CMS lines across four test locations, calculated using equal weights (50:50) for grain yield and stability.

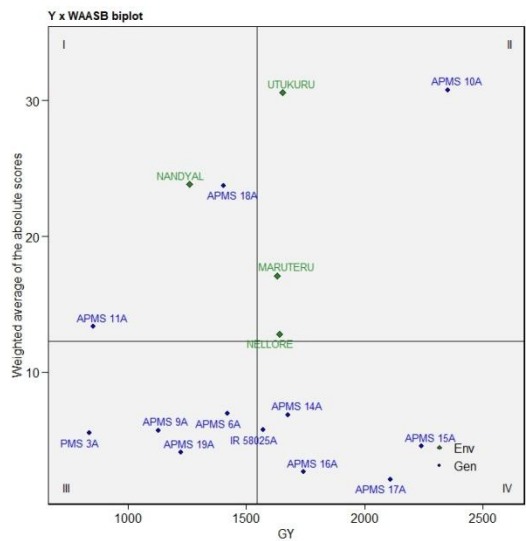


Fig 6. Best linear unbiased predilection means values of 12 CMS lines for grain yield per hectare across four test locations.

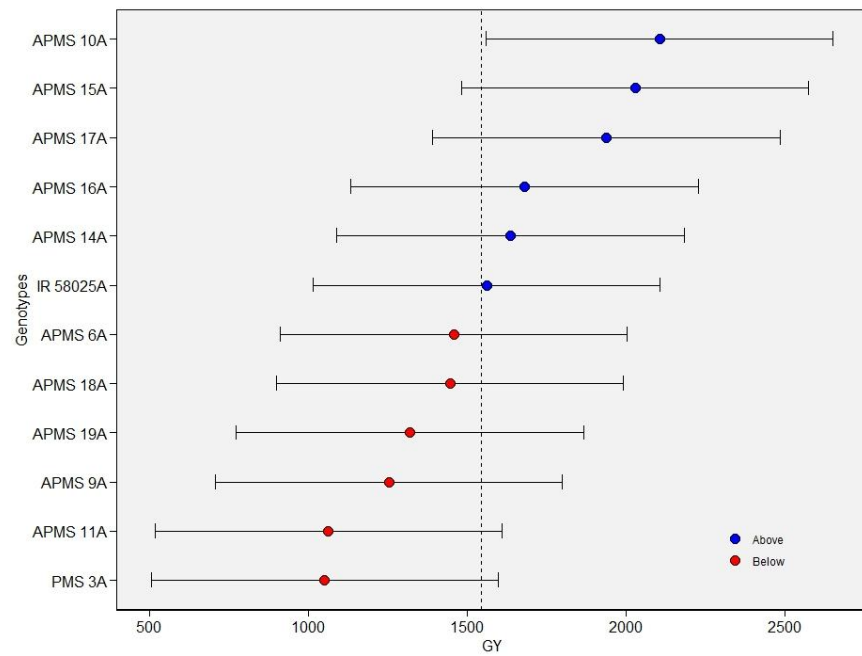


Fig 7a-b. Heatmap illustrating the ranking of 12 WA-CMS lines based on the WAASBY index under different weight combinations assigned to grain yield and stability (100:0 to 0:100).

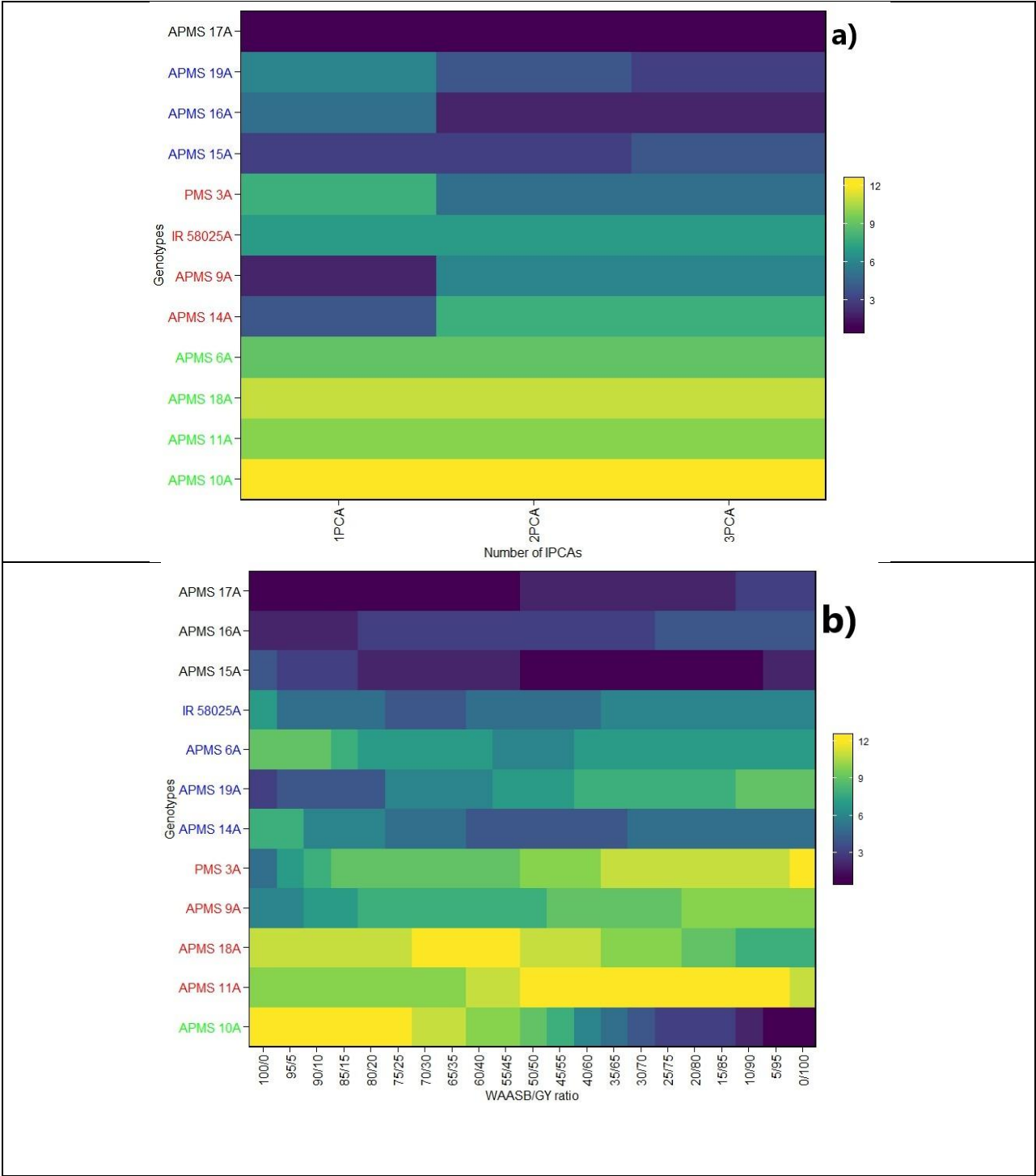


Fig. 8. Positive and negative selection differential (%) for eleven agronomic and floral traits among 12 CMS lines across four test locations

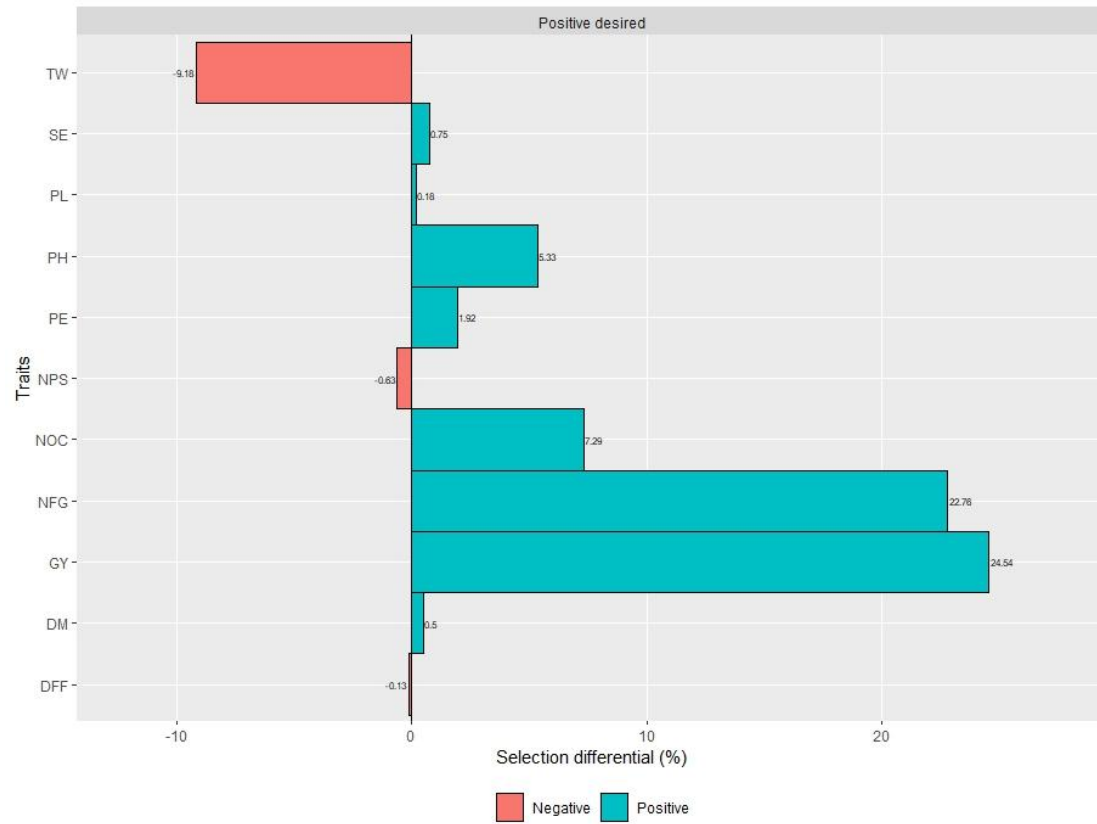


Table 1. Geographical details of test locations for evaluation of CMS lines during *kharif*, 2022

Location	Temperature, Rainfall pattern and relative humidity during rice growing season, <i>Kharif</i> , 2022																					Soil type	Latitude and Longitude
	June			July			August			September			October			November			December				
	T (°C)	R (mm)	RH (%)	T (°C)	R (mm)	RH (%)	T (°C)	R (mm)	RH (%)	T (°C)	R (mm)	RH (%)	T (°C)	R (mm)	RH (%)	T (°C)	R (mm)	RH (%)	T (°C)	R (mm)	RH (%)		
Maruteru	34.95	5.45	84.63	30.40	7.31	88.06	31.65	5.12	88.65	31.40	6.85	85.90	31.26	7.23	90.92	29.83	1.15	82.50	29.26	2.22	88.81	Vertisols	16.6268° N, 81.7389° E
Nellore	35.3	74.5	63.7	31.7	201.8	74.9	32.7	125.1	73.9	32.4	36.8	73.2	30.5	28.6	75.8	27.6	439.2	85.3	27.7	246	87.9	Vertisols	14.4426° N, 79.9865° E
Nandyal	37	2.9	76.6	32.7	6.6	84.0	33.3	3.4	80.0	32.7	4.6	82.5	31.7	7.0	80.2	31.4	0.2	78.3	30.8	1.7	84.3	Vertisols	15.4785° N, 78.4831° E
Kadapa	35.26	169	73.73	34.04	35.8	73.38	34.32	46.3	74.74	32.83	138.3	76.23	31.99	31.8	78.9	29.48	75.9	85.6	29.6	105	85.16	Alfisols	14.438 N, 78.804 E

Table 2. List of CMS lines utilized in the study

S. No	CMS line	Codes
1	APMS 6A	CMS line 1
2	APMS 9A	CMS line 2
3	APMS 10A	CMS line 3
4	APMS 11A	CMS line 4
5	APMS 14A	CMS line 5
6	APMS 15A	CMS line 6
7	APMS 16A	CMS line 7
8	APMS 17A	CMS line 8
9	APMS 18A	CMS line 9
10	APMS 19A	CMS line 10
11	PMS 3A	CMS line 11
12	IR 58025A	CMS line 12

Table 3. The joint analysis of variance for eleven agronomic and floral traits among 12 CMS lines across four test locations.

		DF		DM		PH		NPS		PL		PE	
Source	Df	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)
ENV	3	3759.06	0.00	2491.06	0.00	4969.67	0.00	1385450.30	0.00	236.21	0.00	187.73	0.00
REP(ENV)	8	4.71	0.77	13.18	0.26	6.64	0.43	147.34	0.59	0.75	0.10	6.73	0.36
GEN	11	448.76	0.00	355.77	0.00	924.08	0.00	30847.16	0.19	2.66	0.89	154.33	0.00
GEN:ENV	33	80.62	0.00	92.77	0.00	116.96	0.00	21168.92	0.00	5.37	0.00	29.28	0.00
ENV/GEN	36	387.16	0.00	292.63	0.00	521.36	0.00	134859.04	0.00	24.61	0.00	42.49	0.00
Residuals	88	7.80		10.21		6.52		179.95		0.43		6.01	
CV(%)			2.81		2.46		2.60		2.92		2.67		2.76
MSR+/MSR-			2.30		1.70		2.14		10.72		1.55		1.46
OVmean			99.44		129.85		98.23		459.94		24.57		88.93
		NFG		NOC		SE		TW		GY			
Source	Df	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)	Mean Sq	Pr(>F)		
ENV	3	13677.63	0.00	484.98	0.00	374.60	0.00	77.00	0.00	1303622.49	0.00		
REP(ENV)	8	4.59	0.70	0.22	0.46	11.50	0.05	0.08	0.96	2164.82	0.94		
GEN	11	18919.03	0.01	208.99	0.01	105.73	0.00	43.74	0.00	3061360.89	0.00		
GEN:ENV	33	18555.25	0.00	71.89	0.00	19.24	0.00	2.55	0.00	930864.22	0.00		
ENV/GEN	36	32232.88	0.00	106.32	0.00	48.85	0.00	8.75	0.00	961927.40	0.00		
Residuals	88	72.81		0.23		5.52		0.27		6244.72			
CV(%)			2.46		2.61		2.59		2.81		5.11		
MSR+/MSR-			5.07		1.86		1.67		2.00		9.86		
OVmean			36.93		18.26		90.54		18.50		1545.78		

P<0.01 indicates significance

Table 4. The mean performance for eleven agronomic and floral traits among 12 CMS lines across four test locations.

Parameters	DFF	DM	PH	NPS	PL	PE	NFG	NOC	SE	TW	GY
Mean	99.44	129.85	98.23	459.94	24.57	88.93	36.93	18.26	90.54	18.5	1545.78
SE	0.98	0.87	1.2	15.91	0.22	0.43	1.58	0.55	0.41	0.2	57.83
SD	11.67	10.36	14.33	190.23	2.58	5.14	18.86	6.53	4.93	2.39	691.57
CV	11.77	8.01	14.64	41.51	10.54	5.8	51.26	35.91	5.46	12.95	44.9
Min	76.77 (IR 58025A in Utukuru)	102.61 (IR 58025A in Utukuru)	54.74 (IR 58025A in Nellore)	218.57 (APMS 11A in Maruteru)	17.74 (IR 58025A in Nellore)	74.83 (APMS 9A in Maruteru)	10.67 (APMS 11A in Nandyal)	6.94 (APMS 11A in Maruteru)	76.51 (APMS 6A in Utukuru)	13.4 (APMS 19A in Nandyal)	200 (APMS 11A in Utukuru)
Max	124.38 (APMS 14A in Maruteru)	151.79 (APMS 14A in Maruteru)	119.87 (APMS 16A in Maruteru)	849.87 (PMS 3A in Nellore)	30.15 (APMS 14A in Maruteru)	100.43 (APMS 18A in Nandyal)	92.96 (APMS 10A in Utukuru)	32.51 (APMS 10A in Nellore)	99.58 (APMS 15A in Maruteru)	23.79 (APMS 10A in Utukuru)	4232.59 (APMS 10A in Utukuru)
MinENV	Nellore (89)	Nandyal (121.42)	Nellore (80.94)	Maruteru (275)	Nellore (21.09)	Maruteru (85.91)	Nandyal (28.63)	Maruteru (13.64)	Nandyal (88.12)	Nandyal (16.35)	Nandyal (1260.68)
MaxENV	Maruteru (110.92)	Maruteru (140)	Utukuru (107.18)	Nandyal (662.75)	Maruteru (27.08)	Utukuru (91.08)	Utukuru (53.11)	Nandyal (22.52)	Maruteru (95.28)	Utukuru (19.53)	Utukuru (1652.56)
MinGEN	IR 58025A (82.62)	IR 58025A (115.25)	IR 58025A (74.75)	APMS 6A (394.35)	IR 58025A (23.5)	APMS 9A (83.1)	APMS 11A (13.49)	PMS 3A (13.15)	APMS 6A (81.5)	APMS 18A (15.67)	PMS 3A (836.28)
MaxGEN	APMS 14A (106.62)	APMS 14A (136.62)	APMS 16A (106.22)	PMS 3A (557.7)	APMS 14A (25.25)	APMS 18A (94.47)	APMS 15A (52.46)	APMS 10A (24.67)	APMS 15A (92.85)	APMS 10A (22.49)	APMS 10A (2351.37)

Table 5. AMMI ANOVA for eleven agronomic and floral traits among 12 CMS lines across four test locations.

		DF			DM			PH			NPS			PL			PE		
Source	Df	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance
ENV	3	3759.06	0.00	50.66	2491.06	0.00	40.37	4969.67	0.00	44.61	1385450.30	0.00	70.33	236.21	0.00	62.36	187.73	0.00	11.79
REP(ENV)	8	4.71	0.77	0.17	13.18	0.26	0.57	6.64	0.43	0.16	147.34	0.59	0.02	0.75	0.10	0.53	6.73	0.36	1.13
GEN	11	448.76	0.00	22.18	355.77	0.00	21.14	924.08	0.00	30.42	30847.16	0.00	5.74	2.66	0.00	2.57	154.33	0.00	35.54
GEN:ENV	33	80.62	0.00	11.95	92.77	0.00	16.54	116.96	0.00	11.55	21168.92	0.00	11.82	5.37	0.00	15.60	29.28	0.00	20.23
PC1	13	114.26	0.00	55.80	155.47	0.00	66.00	196.17	0.00	66.10	36964.34	0.00	68.80	8.79	0.00	64.40	57.63	0.00	77.50
PC2	11	69.01	0.00	28.50	60.11	0.00	21.60	107.43	0.00	30.60	14070.06	0.00	22.20	5.19	0.00	32.20	19.75	0.00	22.50
PC3	9	46.21	0.00	15.60	42.11	0.00	12.40	14.20	0.03	3.30	7029.70	0.00	9.10	0.66	0.15	3.40	0.00	1.00	0.00
Residuals	88	7.80			10.21			6.52			179.95			0.43			6.01		
Total	176	126.47			105.19			189.89			33578.59			6.46			27.14		
		NFG			NOC			SE			TW			GY					
Source	Df	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance	Mean Sq	Pr(>F)	Proportion of variance			
ENV	3	4559.21	0.00	19.60	484.98	0.00	17.08	374.60	0.00	27.18	77.00	0.00	25.53	1303622.00	0.00	3.93			
REP(ENV)	8	0.57	0.70	0.01	0.22	0.46	0.02	11.50	0.05	2.23	0.08	0.96	0.07	2164.82	0.94	0.02			
GEN	11	1719.91	0.00	27.11	208.99	0.00	26.98	105.73	0.00	28.13	43.74	0.00	53.19	3061361.00	0.00	33.81			
GEN:ENV	33	562.28	0.00	26.59	71.89	0.00	27.84	19.24	0.00	15.36	2.55	0.00	9.29	930864.20	0.00	30.85			
PC1	13	822.16	0.00	57.60	103.94	0.00	57.00	33.07	0.00	67.70	4.73	0.00	73.20	1536887.00	0.00	65.00			
PC2	11	454.86	0.00	27.00	64.69	0.00	30.00	16.39	0.00	28.40	1.71	0.00	22.40	976252.10	0.00	35.00			
PC3	9	318.20	0.00	15.40	34.40	0.00	13.10	2.74	0.87	3.90	0.41	0.15	4.40	23.28	1.00	0.00			
Residuals	88	0.83			0.23			5.52			0.27			6244.72					
Total	176	396.50			48.41			23.49			5.14			565850.70					

Table 6. AMMI Stability Values for eleven agronomic and floral traits among 12 CMS lines across four test locations.

DFF			DM		PH		NPS		PL		PE		NFG		NOC		SE		TW		GY	
GEN	AS V	ASV_ R	ASV	ASV_ R	AS V	ASV_ R	ASV	ASV_ R	AS V	ASV_ R	AS V	ASV_ R	ASV	AS V_ R	AS V	ASV_ R	AS V	ASV_ R	AS V	ASV_ R	ASV	AS V_ R
APMS 10A	2.22	9	3.35	9	2.50	3	13.31	6	0.06	1	1.83	2	11.61	12	6.13	12	0.97	5	0.46	1	78.39	12
APMS 11A	2.67	10	2.40	5	2.82	6	6.27	4	0.88	4	3.81	8	3.48	4	2.94	9	4.45	12	0.76	5	27.25	10
APMS 14A	4.88	11	5.87	11	4.12	10	25.62	9	2.86	12	2.54	4	3.63	5	0.14	1	0.82	4	0.51	2	16.13	8
APMS 15A	1.26	5	1.77	3	3.24	8	3.41	1	1.92	9	3.79	7	2.61	3	2.19	6	0.52	3	0.66	4	7.44	4
APMS 16A	1.05	2	2.77	7	2.54	4	18.00	7	1.36	7	2.69	5	1.11	2	0.42	2	0.99	6	3.79	11	5.57	2
APMS 17A	1.08	3	1.45	2	3.18	7	27.76	12	1.10	5	1.06	1	4.98	9	0.73	3	3.34	8	1.53	9	3.96	1
APMS 18A	0.83	1	1.32	1	2.26	1	5.19	3	0.51	2	2.06	3	6.86	11	2.93	8	3.63	10	0.51	3	52.61	11
APMS 19A	1.21	4	3.09	8	4.46	11	7.76	5	0.58	3	4.52	9	3.65	6	3.39	11	0.37	1	4.90	12	7.28	3
APMS 6A	1.96	7	2.12	4	6.50	12	26.17	10	2.03	10	6.22	11	4.06	8	2.96	10	0.45	2	1.25	7	16.53	9
APMS 9A	1.56	6	3.71	10	2.38	2	5.11	2	2.09	11	4.79	10	0.37	1	1.90	5	3.76	11	1.11	6	14.54	7
IR 58025A	6.99	12	12.55	12	4.10	9	24.12	8	1.24	6	2.91	6	5.48	10	2.64	7	1.58	7	1.83	10	12.66	6
PMS 3A	2.02	8	2.45	6	2.76	5	26.95	11	1.65	8	7.39	12	3.65	7	0.86	4	3.43	9	1.52	8	10.47	5

Table 7. Estimation of variance components by restricted maximum likelihood (REML) among 12 CMS lines across eleven agronomic and floral traits over four test locations.

Parameters	DFF	DM	PH	NPS	PL	PE	NFG	NOC	SE	TW	GY
Phenotypic variance	62.75	59.65	110.60	7982.79	1.85	24.19	284.45	35.54	17.30	4.46	491992.60
Heritability	0.49	0.37	0.61	0.10	0.05	0.43	0.34	0.32	0.42	0.77	0.36
Genotype $\times$ Environment Interaction Ratio (GEI _{r2})	0.39	0.46	0.33	0.88	0.77	0.32	0.66	0.67	0.26	0.17	0.63
Broad-sense heritability on genotype mean basis (h ² _{mg})	0.82	0.74	0.87	0.31	0.00	0.81	0.67	0.66	0.82	0.94	0.70
Accuracy	0.91	0.86	0.93	0.56	0.00	0.90	0.82	0.81	0.90	0.97	0.83
Genotypic correlation (r _{ge})	0.76	0.73	0.85	0.97	0.77	0.56	1.00	0.99	0.45	0.74	0.98
Genotypic coefficient of variation (CV _g)	5.57	3.61	8.35	6.17	0.00	3.63	26.60	18.51	2.97	10.02	27.26
Residual variation (CV _r)	2.81	2.46	2.60	2.92	2.67	2.76	2.46	2.61	2.59	2.81	5.11
CV ratio (CV _g /CV _r)	1.98	1.46	3.21	2.12	0.00	1.32	10.80	7.10	1.14	3.57	5.33

Table 8. MTSI ranking among 12 CMS lines across eleven agronomic and floral traits over four test locations.

S.No	Genotype	MTSI
1	APMS 17A	1.482
2	APMS 16A	1.572
3	APMS 14A	2.268
4	APMS 15A	2.278
5	APMS 19A	3.117
6	APMS 18A	3.136
7	APMS 10A	3.193
8	APMS 9A	3.562
9	PMS 3A	3.649
10	APMS 6A	3.705
11	APMS 11A	4.062
12	IR 58025A	4.956

Table 9. Evaluation of selection differential and genetic gains for eleven agronomic and floral traits among 12 CMS lines across four test locations.

VAR	Factor	Xo	Xs	SD	SDperc	h2	SG	SGperc	sense	goal	Communalit y	Uniquenesse s
DFE	FA 1	99.44	99.31	-0.13	-0.13	0.82	-0.10	-0.10	increase	0	0.95	0.05
DM	FA 1	129.85	130.50	0.65	0.50	0.74	0.48	0.37	increase	100	0.95	0.05
PH	FA 1	98.23	103.46	5.24	5.33	0.87	4.57	4.66	increase	100	0.79	0.21
NFG	FA 2	36.93	45.33	8.41	22.76	0.67	5.66	15.32	increase	100	0.73	0.27
NOC	FA 2	18.26	19.59	1.33	7.29	0.66	0.87	4.78	increase	100	0.93	0.07
GY	FA 2	1545.78	1925.12	379.34	24.54	0.70	263.99	17.08	increase	100	0.83	0.17
PL	FA 3	24.57	24.61	0.04	0.18	0.00	0.00	0.00	increase	100	0.95	0.05
PE	FA 3	88.93	90.64	1.71	1.92	0.81	1.39	1.56	increase	100	0.88	0.12
SE	FA 3	90.54	91.23	0.68	0.75	0.82	0.56	0.62	increase	100	0.65	0.35
NPS	FA 4	459.94	457.05	-2.89	-0.63	0.31	-0.91	-0.20	increase	0	0.63	0.37
TW	FA 4	18.50	16.80	-1.70	-9.18	0.94	-1.60	-8.64	increase	0	0.88	0.12
PC		PC1	PC2	PC3	PC4	Min		Max	total	Mean		
Eigenvalues		3.75	2.78	1.53	1.10	-9.18		24.54	68.71	4.85		
Variance (%)		34.15	25.27	13.95	10.08	-8.64		17.08	43.87	3.22		
Cum. variance		34.15	59.43	73.38	83.46							

Table 10. Comparative selected stable high grain yield CMS lines across various stable indexes models

AMMI	ASV	WAASB	WASSBY	BLUP	MTSI	Common CMS lines
APMS 14A, APMS 15A, APMS 16A, and APMS 17A	APMS 17A, APMS 16A, APMS 19A, and APMS 15A	APMS 14A, APMS 15A, APMS 16A, and APMS 17A	APMS 17A, APMS 16A, APMS 19A, and APMS 15A	APMS 10A, APMS 15A, APMS 16A, and APMS 17A	APMS 16A, APMS 17A, APMS 14A, APMS 15A,	APMS 14A, APMS 15A, APMS 16A, and APMS 17A

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