ARTICLE

Comparison of methodologies (REML/BLUP vs. GGE Biplot) for soybean selection in the Brazilian Cerrado

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Abstract: *The selection of adapted and stable soybean genotypes for the Brazilian Cerrado is hindered by the complex genotype x environment (G x E) interaction. This study aimed to compare the effectiveness of the REML/BLUP and GGE Biplot. Yield data from 33 genotypes in 44 environments (2019-2020 and 2020-2021 growing seasons) were analyzed. The results indicated a significant G x E interaction and high selective accuracy (0.86), validating the selection precision. Both methodologies proved to be effective and concordant in identifying a group of elite genotypes (commercial checks G3 (BASF), G6 (BASF), G4 (BASF), and the experimental lines G17 (BASF) and G18 (BASF)). REML/BLUP provided the quantitative basis for selection, whereas the GGE Biplot elucidated the formation of mega-environments and the positioning of genotypes relative to the ideotype. It is concluded that the methodologies are complementary, enhancing decision-making and maximizing genetic gains in soybean breeding programs for the Cerrado.*

Keywords: *Stability analysis, adaptability, heritability, mixed models*

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INTRODUCTION

The genotype by environment (G x E) interaction is a factor that significantly influences the outcomes of plant breeding programs, particularly in the soybean crop (Albuquerque et al. 2023, Elmerich et al. 2023). This interaction manifests when a single genotype exhibits phenotypic variability in response to variations in environmental conditions, leading to changes in the performance ranking of this genotype across different locations and growing seasons (Fu and Wang 2023, Napier et al. 2023). This scenario complicates the identification and selection of broadly adapted varieties, as a cultivar's performance may not be consistent across diverse cultivation environments (Souza et al. 2021, Rani et al. 2023). A critical analysis of G x E effects is therefore an essential step in genetic improvement and agronomic evaluation studies, as evidenced by the pioneering works on phenotypic stability (Allard and Bradshaw 1964).

To study the G x E interaction and select superior genotypes, multi-environment trials (METs) are essential, but the resulting data are often large and unbalanced, demanding more robust statistical methodologies (Resende 2007). Although various analytical tools exist, Linear Mixed Models (LMMs), through the REML/BLUP (Restricted Maximum Likelihood / Best Linear Unbiased

Prediction) procedure, are recognized as a more robust methodology (Capistrano et al. 2021). The efficiency of REML/BLUP is particularly notable for handling complex experimental data, allowing for the accurate estimation of variance components and the reliable prediction of genetic values (BLUPs), which are crucial for genotype selection across various crops (Bezerra et al. 2023, Nascimento et al. 2024).

As a complement to the quantitative approach of REML/BLUP, GGE Biplot (Genotype main effects and Genotype × Environment interaction) analysis emerges as a multivariate method focusing on the effects of the genotype (G) and the G x E interaction, providing graphical visualizations that facilitate the identification of mega-environments and the evaluation of ideal genotypes (Enyew et al. 2021, Bomma et al. 2024). The combined use of different methods to investigate adaptability and stability is, therefore, highly desirable. By combining the information provided by REML/BLUP and GGE Biplot, it is possible to explore different parameters, offering a more comprehensive analytical perspective for decision-making (Gonçalves et al. 2020). This integrated approach, which unites the quantitative prediction of REML/BLUP with the visual interpretation of interaction patterns from the GGE Biplot, offers a more robust analytical basis for genotype selection. The combined application of these methods, in the context of an extensive trial network in the Brazilian Cerrado, not only allows for the confirmation of selection concordance but also deepens the understanding of mega-environment formation and specific genotype performance.

The genotype-environment interaction is, therefore, an important factor in the selection of soybean genotypes. Although it hinders the selection of broadly adapted cultivars, its analysis is fundamental for exploring specific adaptations, enabling the recommendation of superior genotypes for defined environmental niches. The central hypothesis is that high consistency will exist in the classification of superior genotypes by both REML/BLUP and GGE Biplot. However, it is argued that integrating quantitative information (REML/BLUP) with the visual patterns (GGE Biplot) constitutes a more robust tool for optimizing cultivar recommendation strategies. Given this scenario, the objective of this study was to compare the REML/BLUP and GGE Biplot methodologies for selecting soybean genotypes with high stability and adaptability, in order to identify the most effective approach for breeding programs across the diverse environments of the Brazilian Cerrado.

MATERIAL AND METHODS

Characterization of genotypes and environments

This study included 33 soybean genotypes, comprising 9 commercial cultivars used as checks and 24 lines. The genotypes were provided by different companies in the sector, including BASF, TMG, Bayer, and Genética Dom Mário. The experimental network consisted of 44 environments, resulting from the combination of different locations and years, derived from Value for Cultivation and Use (VCU) trials conducted in the Brazilian Cerrado during the 2019/2020 and 2020/2021 growing seasons. The trials spanned multiple Edaphoclimatic Regions, distributed across the states of Goiás, Mato Grosso, Tocantins, Bahia, and Pará (Figure 1). The response variable evaluated was grain yield, determined from the harvest of the experimental plots, with the values subsequently adjusted to 13% moisture content and expressed in kilograms per hectare (kg ha^{-1}).

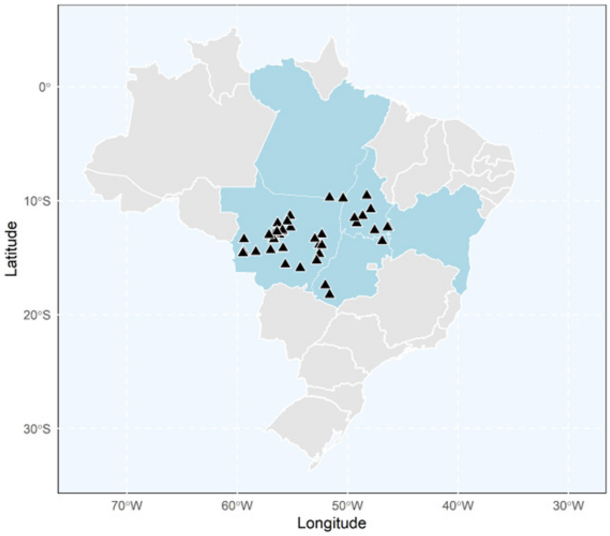

Figure 1. Geographic distribution of the 44 environments used in the study, located in the Brazilian Cerrado.

Experimental design

In each of the 44 environments, the trials were established using a randomized complete block design with three replications. Each experimental plot consisted of four 5-meter-long rows, spaced 0.5 meters apart. For yield evaluation, the two outer rows were considered as borders; only the two central rows were harvested and assessed, resulting in a net plot area of 5 m^2 per plot. The agronomic management adopted in each trial followed the technical recommendations for the soybean crop in the respective region, with weed, pest, and disease control carried out as necessary and at the technical discretion of the local manager.

Statistical analysis via REML/BLUP methodology

To estimate the variance components and predict the genotypic values, the following linear mixed model was fitted to the yield data, according to the REML/BLUP methodology:

$$\gamma = X\beta + Zg + Wi + \epsilon$$

where γ is the vector of phenotypic data; β is the vector of fixed effects, comprising the intercept, the effects of environments, and replications within environments; g is the vector of genotypic effects (random); i is the vector of the G x E interaction effects (random); and ϵ is the vector of errors or residuals (random). X, Z, and W are the incidence matrices associated with the fixed effects of replications, random genotypic effects, and random G x E interaction effects, respectively. The random effects were assumed to follow multivariate normal distributions with zero means and variance-covariance matrices defined as $g \sim N(0, I\sigma^2_g)$, $i \sim N(0, I\sigma^2_i)$ and $e \sim N(0, I\sigma^2_e)$.

All statistical analyses were conducted in the R software (version 4.5.1) (R Core Team 2025), using the 'Metan' package (version 1.19.0) (Olivoto et al. 2019) and the SELEGEN-REML/BLUP software. Specifically, the REML/BLUP analysis was performed using the waasb() function. The significance of the random effects was evaluated using the Likelihood Ratio Test (LRT). The genotypic (σ^2_g), G x E interaction (σ^2_i), and residual (σ^2_e) variance components were estimated via REML. Based on these components, the following genetic parameters were calculated: genotype mean-based heritability ($h^2_{mg} = \frac{\sigma^2_g}{\sigma^2_g + \frac{\sigma^2_{ge}}{e} + \frac{\sigma^2_e}{re}}$), selective accuracy ($r_{gg} = \sqrt{h^2_{mg}}$), and the genotypic correlation of genotype performance across environments ($r_{ge} = \frac{\sigma^2_g}{\sigma^2_g + \sigma^2_{ge}}$), where '$e$' is the number of environments and 'r' is the number of replications. Selection for yield, adaptability, and stability was based on the Harmonic Mean of the Relative Performance of Genotypic Values (HMRPGV).

Statistical analysis via GGE biplot methodology

The GGE biplot analysis was performed using the predicted genotypic values for each genotype in each environment, obtained from the previous analysis. The input matrix for the biplot was formed by the sum of the Best Linear Unbiased Estimator (BLUE) of the overall mean and the Best Linear Unbiased Predictions (BLUP) of the genotypic and G x E interaction effects ($\hat{y}$ = BLUE(μ) + BLUP(g) + BLUP(i)).

This multivariate method, which focuses on the portion of variance corresponding to the genotype and G x E interaction effects, was used to visually interpret the interaction patterns. The analysis was conducted using the gge() function from the 'Metan' package (Olivoto et al. 2019) in the R software (R Core Team 2025).

The following biplots were generated and interpreted: the "Which-won-where" plot to identify possible mega-environments and the winning genotypes in each; the "Mean vs. Stability" plot to evaluate the mean performance and stability of the genotypes; the "Ranking Genotypes" plot to rank genotypes relative to an ideotype (high yield and high stability); and the "Discriminativeness vs. Representativeness" plot to assess the discriminating ability and representativeness of the test environments.

Comparison between REML/BLUP estimators and GGE biplot

The comparison between the REML/BLUP and GGE biplot approaches was performed qualitatively. The criterion consisted of analyzing the ranking of the superior genotypes identified by each method (selection via HMRPGV in the REML/BLUP analysis versus positioning in the GGE Biplot), looking for consistencies and discrepancies. Additionally, the complementarity of the insights provided by each methodology for elucidating patterns of adaptability and stability was assessed. Quantitatively, the concordance between the methods was evaluated using Spearman's rank correlation coefficient (r_s). This correlation was calculated between the HMRPGV index values (from REML/BLUP) and the scores of the first principal component (PC1) of the GGE biplot, which relates to yield performance.

RESULTS AND DISCUSSION

Deviance analysis and genetic parameters via REML/BLUP

The analysis of grain yield of the 33 soybean genotypes across the 44 environments, according to the LRT, revealed significant effects ($p < 0.01$) for the environment, genotype, and the G x E interaction factors. The highly significant difference observed among environments highlights the distinct edaphoclimatic conditions of the experimental network, which is essential for a robust evaluation of adaptability and stability. These results not only confirm the magnitude of the challenge posed by this interaction for cultivar selection in the Cerrado region but also establish the validity of the data presented.

Table 1 presents the variance components and genetic parameters estimated from the linear mixed model. In this model, the residual variance (σ^2_e) represents the uncontrolled experimental variation within each environment. The coefficient of determination for the G x E interaction (R^2_i) was 0.4065, indicating that this interaction accounted for approximately 40.65% of the phenotypic variation. Furthermore, the low genotypic correlation across environments (r_{ge} = 0.09) highlights the predominant nature of the complex interaction. This magnitude indicates substantial changes in genotype rankings across environments, reinforcing the challenge of selecting broadly adapted cultivars and justifying the use of multivariate tools like the GGE biplot. The genotype mean-based heritability (h^2_{mg}) was estimated at 0.7519, resulting in a selective accuracy (r_{gg}) of 0.8671. This indicator of experimental quality confirms that the observed complexity in the genotypes' responses is not a statistical artifact or noise, but a real biological and environmental phenomenon. The coefficient of experimental variation ($CV_{e\%}$) was 9.77%, while the coefficient of genetic variation ($CV_{g\%}$) was 2.62%.

It is crucial to highlight the robustness provided by the experimental network. While the single-plot heritability (h^2_g = 0.01603) reflects the magnitude of environmental noise and G x E interaction, the high genotype mean-based heritability ((h^2_{mg} = 0.7519) demonstrates the effectiveness of evaluating 44 environments. This contrast confirms that averaging performance across multiple replications and environments successfully minimized random error, allowing for a precise estimation of genetic values. This is practically reflected in the high selective accuracy (r_{gg} = 0.8671), which validates the selection process.

Table 1. Likelihood Ratio Test and genetic parameters for grain yield of 33 soybean genotypes evaluated across 44 environments during the 2019 and 2020 growing seasons in the Brazilian Cerrado

Effect	**LRT Statistic**	**Pr (>Chisq)**
Gen	44.017088	3.25522×10^{-11}
Rep(env)	24.925835	5.95786×10^{-07}
Env	249.97571	2.62867×10^{-56}
Gen x Env	506.35471	3.9385×10^{-112}
Estimates of variance components and genetic parameters		
Genotypic variance	σ^2_g	15207.2
G x E interaction variance	σ^2_i	152660.3
Residual variance	σ^2_e	209459
Phenotypic variance	σ^2_p	377326.5
Plot-level heritability	h^2	0.01603
Coefficient of determination of the interaction	R^2_i	0.4065
Genotype mean-based heritability	h^2_{mg}	0.7519
G x E correlation	r_{ge}	0.0906
Selective accuracy	r_{gg}	0.8671
Coefficient of genetic variation	$CV_{g\%}$	2.62
Coefficient of experimental variation	$CV_{e\%}$	9.772
$CV_{g\%}/CV_{e\%}$ Ratio	$CV_{g\%}/CV_{e\%}$ Ratio	0.2681

Selection of superior genotypes via REML/BLUP

Table 2 summarizes the performance and ranking of the top five selected genotypes based on the HMRPGV index. The commercial checks G3 (BASF) and G6 (BASF) ranked first and second, respectively, exhibiting the highest mean genotypic values (BLUP). This superiority of commercial checks validates the high quality of the genetic standard currently used in the region. However, a significant finding was the performance of the experimental lines G17 (BASF) and G18 (BASF), which, along with the check G4 (BASF), comprised the group of the top five elite materials.

The classification of these experimental lines among the top rank indicates their potential to become new commercial cultivars, as they showed productive levels competitive with the best market standards. Furthermore, by using the HMRPGV index, the selection penalized instability, ensuring that these five genotypes combine high yield with the necessary resilience to cope with the environmental variations of the Cerrado.

By outperforming several commercial checks, the experimental lines G17 (BASF) and G18 (BASF) show promising genetic potential for the specific stress conditions of the Cerrado. These genotypes represent valuable germplasm for inclusion in hybridization blocks, potentially contributing to the accumulation of favorable alleles for high yield and stability in the breeding population.

Table 2. Performance of the top soybean genotypes selected via REML/BLUP, based on the Harmonic Mean of the Relative Performance of Genotypic Values (HMRPGV) and the Predicted Mean Genotypic Values (Mean BLUP), and via GGE Biplot based on the First Principal Component score (PC1 - Performance) and the Average Environment Coordination (AEC - Stability) for grain yield (kg ha^{-1}) in the Brazilian Cerrado

Overall Rank	**Genotype**	**HMRPGV** (kg ha^{-1})	**Mean BLUP** (kg ha^{-1})	**PC1 Score** (Performance)	**AEC Stability**
1st	G3	1.0556	4980.43	0.39	139.76
2nd	G6	1.0523	4966.72	0.40	270.14
3rd	G17	1.0296	4870.51	0.18	303.06
4th	G18	1.0294	4871.98	0.21	350.69
5th	G4	1.0274	4854.56	0.23	315.74

Analysis of G x E interaction and overall performance via GGE biplot

The first two principal components (PC1 and PC2) jointly explained 33.37% of the total variation from the genotype plus G x E interaction (G + GE) effects, with PC1 accounting for 20.21% and PC2 for 13.16%. The interaction biplot (Figure 2a) displays the scatter of the 33 genotypes and 44 environments, illustrating the presence and magnitude of the G x E interaction. In the 'Mean vs. Stability' analysis (Figure 2b), the commercial checks G3 (BASF), G4 (BASF), and G6 (BASF) were identified as superior, as they combined high mean yield (higher PC1 scores) with good stability (a smaller perpendicular projection to the average environment axis).

Formation of mega-environments and genotype-specific performance

The 'Which-won-where' analysis, shown in Figure 3, divided the test environments into six distinct mega-environments based on the best-performing genotype in each sector. The experimental line G11 (BASF) was the winner for ME1 and the experimental line G12 (BASF) for ME2, while the commercial checks G6 (BASF) and G3 (BASF) were the winners in the remaining four groups, showing broad adaptation. The delineation of mega-environments goes beyond recommending a single, general cultivar, enabling the targeting of specific genotypes, such as the experimental lines G11 (BASF) and G12 (BASF), to the microregions where their superior performance has been confirmed. This strategic recommendation approach has the potential to increase the rate of regional yield gains.

Ranking relative to the ideotype and evaluation of test environments

The commercial checks G3 (BASF), G6 (BASF), G4 (BASF), and the experimental lines G17 (BASF) and G18 (BASF) were closest to the center of the concentric circles, thus being considered the most desirable according to the ranking of genotypes relative to an ideal profile of high yield and stability (Figure 4a). The evaluation of test environments (Figure 4b) identified environment X38 (Formosa do Rio Preto, BA - season 2020/21) as the most discriminating and

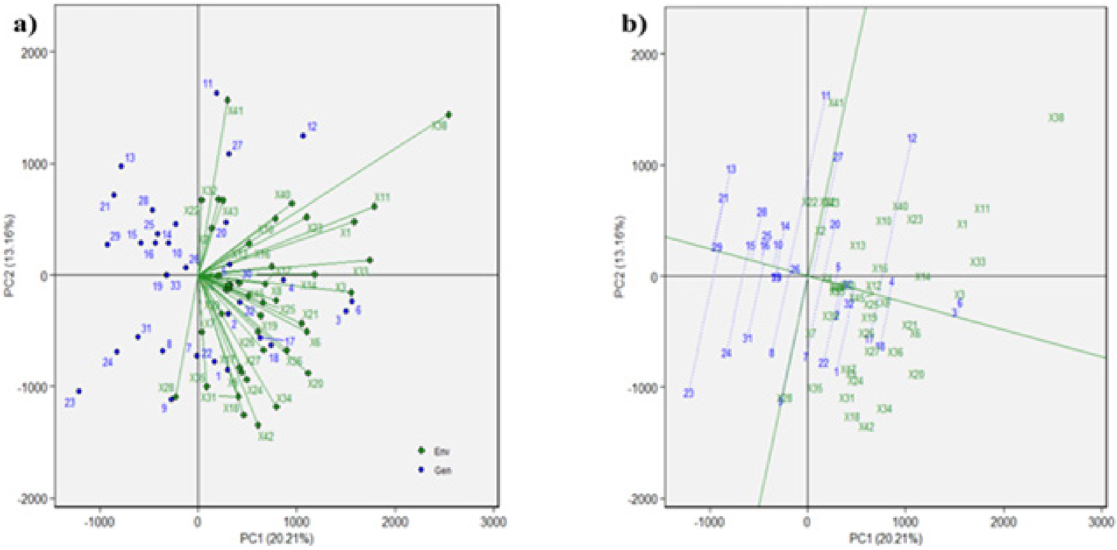

Figure 2. Analysis of the genotype x environment (G x E) interaction and yield stability using the GGE biplot methodology in multi-environment trials. Blue points/numbers represent the genotypes and green diamonds/labels represent the environments. **(a)** Interaction pattern and relationship among genotypes and environments. **(b)** Classification of genotypes according to mean yield and stability ('Mean vs. Stability').

environment X8 (Querência, MT - season 2019/20) as the most representative of the trial network. The practical implications of this methodological complementarity are straightforward. The identification of ideal environments, such as X38 (Formosa do Rio Preto, BA - season 2020/21) for its discriminating ability and X8 (Querência, MT - season 2019/20) for its representativeness, allows for a more strategic and efficient allocation of resources, optimizing the final stages of line evaluation.

Quantitative concordance between methodologies

Spearman's correlation analysis confirmed a strong agreement between the statistical approaches. The HMRPGV selection index (REML/BLUP) showed a high and significant correlation with the GGE biplot performance scores (PC1) ($r_s = 0.9188$; $p < 0.0001$). Similarly, the mean genotypic values (BLUP) were strongly correlated with PC1 ($r_s = 0.9168$; $p < 0.0001$). These results quantitatively validate the effectiveness of both methodologies in identifying the same group of elite genotypes for the Cerrado.

This indicates that the first principal component (PC1) of the GGE biplot was efficient in capturing the variation related to yield performance, behaving similarly to the quantitative indices of REML/BLUP. Consequently, the complementarity of the GGE biplot lies mainly in the second principal component (PC2), which explained 13.16% of the variation. While PC1 and REML/BLUP identify the most productive genotypes, the visual analysis of PC2 adds distinct

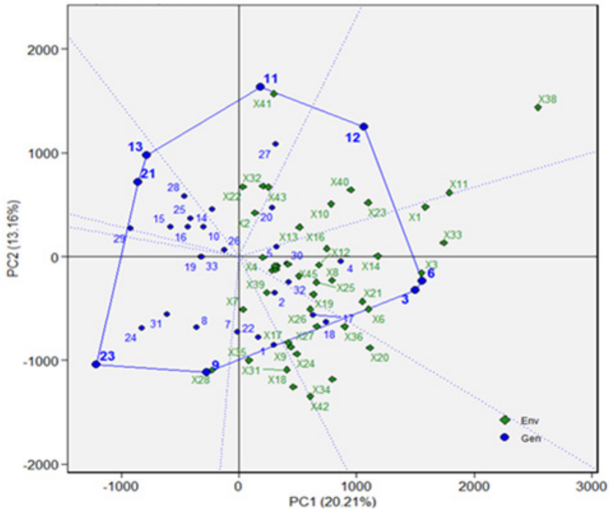

Figure 3. 'Which-Won-Where' GGE biplot analysis of the genotype x environment (G x E) interaction for soybean genotypes evaluated in multi-environment trials. The polygon is drawn connecting the vertex genotypes (blue labels) and environments (green labels) are positioned within sectors.

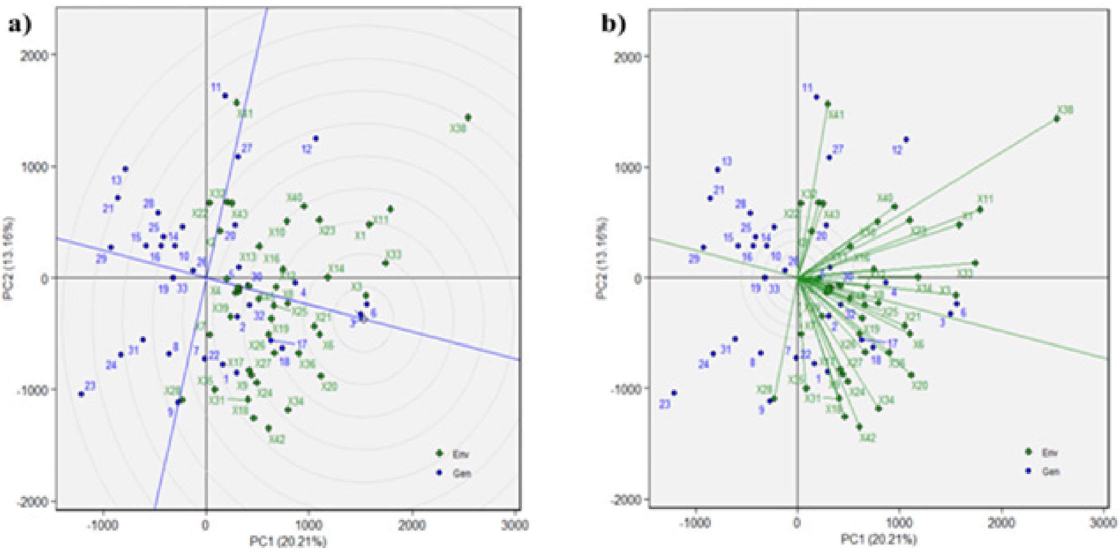

Figure 4. Evaluation of genotypes and environments using GGE biplot analysis. Blue points/numbers represent the genotypes and green diamonds/labels represent the environments. **(a)** Ranking of genotypes based on proximity to the ideotype ('Ranking Genotypes'). **(b)** Evaluation of test environments for their discriminating power and representativeness ('Discriminativeness vs. Representativeness').

information about the stability and specific interactions of these materials, which are not immediately visible in a purely quantitative ranking.

CONCLUSIONS

Given the validity of the data, the main finding of this study was the high level of agreement between the REML/BLUP and GGE Biplot methodologies. Both approaches consistently identified a cohesive group of superior genotypes (the commercial checks G3 (BASF), G6 (BASF), G4 (BASF), and the experimental lines G17 (BASF) and G18 (BASF)) and delineated six distinct mega-environments. This result reinforces the findings of other studies in the field, such as those by Gonçalves et al. (2020) and Pradebon et al. (2023), who also observed a strong correlation in the ranking of soybean genotypes when employing both approaches. This convergence validates the central hypothesis of this research: that the methods, although conceptually distinct, are complementary.

DATA AVAILABILITY

The datasets generated and/or analyzed in this study, as well as the supplementary tables and figures, are available from the corresponding author upon reasonable request.

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