

Selection of forage oat (*Avena sativa*) genotypes through GGE Biplot and BLUP

Selección de genotipos de avena forrajera (*Avena sativa*) a través de GGE Biplot y BLUP

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Recibido: 15 marzo de 2023; Revisión: 27 de abril 2023; Aceptado: 26 de mayo de 2023

doi: <https://doi.org/10.5281/zenodo.8377231>

ABSTRACT

In Bolivia, the productivity of forage oat is relatively low. To address this issue, the present study aimed to identify more productive and stable genotypes using statistical methods such as GGE Biplot and BLUP (Best Linear Unbiased Prediction). The research was conducted in three different environments in Bolivia, evaluating six commercial varieties of forage oats, including three from Instituto Nacional de Innovación Agraria (INIA-Peru) and the remaining from Centro de Investigación Forrajera (CIF) -Violeta Bolivia. The data obtained were analyzed using GGE Biplot and BLUP, resulting in an average yield of 10.29 ± 3.51 t ha⁻¹ of dry matter. BLUP exhibited a higher cumulative variance than GGE Biplot in the first two components. Both models demonstrated similar trends in terms of productivity and stability values, facilitating the selection process. Consequently, Tayco and Texas were identified as the most promising genotypes due to their exceptional dry matter yield and phenotypic stability.

Key words: *Avena sativa*, BLUP, GGE, multi-environment (MET), stability.

RESUMEN

En Bolivia, la productividad de avena forrajera es relativamente baja. Con el objetivo de abordar este problema, el estudio tuvo como objetivo identificar genotipos más productivos y estables utilizando métodos estadísticos como

el GGE Biplot y BLUP (Mejor Predicción Lineal No Sesgada). La investigación se llevó a cabo en tres ambientes de Bolivia, donde se evaluaron seis variedades comerciales de avena forrajera. Tres de ellas provienen del Instituto Nacional de Innovación Agraria (INIA-Perú) y las restantes del Centro de Investigación Forrajera (CIF) – Violeta, Bolivia. Los datos obtenidos fueron analizados a través de la metodología GGE Biplot y BLUP. El resultado en términos de rendimiento promedio fue de 10.29 ± 3.51 t ha⁻¹ de materia seca y BLUP exhibió una mayor varianza acumulada que GGE Biplot en los dos primeros componentes. Ambos modelos demostraron tendencias similares en cuanto a los valores de productividad y estabilidad, lo que facilitó el proceso de selección. En consecuencia, Tayco y Texas fueron identificados como los genotipos más prometedores debido a su excepcional rendimiento de materia seca y estabilidad fenotípica.

Palabras clave: *Avena sativa*, BLUP, GGE, multi localidad (MET), estabilidad.

INTRODUCTION

In 2018, Bolivia produced 11,363.0 tons of forage oats, resulting in a yield of 2,464.0 kg ha⁻¹ (INE & MDRYT, 2019). These statistics indicate a relatively low level of productivity compared to neighboring countries in the region. In the highlands (Altiplano) of Bolivia, approximately 95% of biomass production is stored as haylage to ensure an adequate supply

during the winter season. The availability of sufficient and timely feed plays a crucial role in the success and prosperity of livestock (Bilal et al., 2017), and insufficient forage supply can negatively impact meat and milk productivity (Ahmad et al., 2014; Rana et al., 2014).

To enhance forage oat production, plant breeding emerges as a fundamental approach for identifying genotypes with high productivity and resilience to biotic and abiotic stressors (Atlin, Cairns, & Das, 2017). This process requires conducting multi-environmental tests to select and recommend genotypes that exhibit both high yields and stability across different locations and over time (Olivoto et al., 2019; Smith & Cullis, 2018). Achieving this requires the application of statistical models with improved prediction capabilities.

In the analysis of multi-environmental trials, various statistical models are employed to study the adaptability and stability of cultivars. These methodologies encompass analysis of variance, nonparametric regressions, multivariate analysis, and mixed models. Among these, GGE Biplot and REML/BLUP (Best Linear Unbiased Prediction) methods have gained prominence due to their practicality in result interpretation and precision in genotype selection (Olivoto et al., 2017; Santos et al., 2017).

GGE Biplot graphs are constructed using the first and second principal components, which represent the genotype main effect and the genotype-by-environment interaction, respectively. This graphical approach enables accurate prediction of the average genotype yield across different environments and facilitates the identification of stable genotypes (Yan & Tinker, 2006). Conversely, Resende (2016) and Santos et al. (2019) assert that BLUP serves as an optimal selection procedure, maximizing selective accuracy and allowing the simultaneous utilization of multiple sources of information. BLUP addresses the issue of imbalance, considers the genetic relationship

among the evaluated plants, and accounts for the coincidence between the selection and recombination units.

Mixed models have gained extensive utilization in plant breeding programs due to their estimation procedures, which effectively address issues such as unbalanced design skewness, non-additive features, and scattered data arising from investigations (Bandera-Fernández & Pérez-Pelea, 2018; Hu, 2015). Furthermore, mixed models enable the estimation of crucial parameters of quantitative genetics in multi-environmental trials and facilitate the selection of superior genotypes based on criteria such as productivity, adaptability, and stability.

AMMI (Additive Main Effects and Multiplicative Interaction) and BLUP are popular methods for analyzing multi-environment trials. However, AMMI has limitations in accommodating a linear mixed-effects model (LMM) structure, while BLUP requires graphical treatment for handling random genotype-by-environment interaction (GEI) structures (Olivoto et al., 2019). In response to these challenges, WAASB (Weighted Average of Absolute Scores from Singular Value Decomposition of BLUP Matrix) was proposed as a statistical tool by Olivoto et al. (2019). WAASB offers an effective approach to identify highly productive and broadly adapted genotypes. Against this backdrop, this study aims to utilize GGE Biplot and BLUP methods to select more productive and stable genotypes of forage oats under three distinct environmental conditions in Bolivia.

MATERIALS AND METHODS

The research was conducted at three distinct locations in Bolivia, as shown in Table 1. Cuyahuani is situated in the northern region of La Paz, where the local community primarily engages in milk production. The Toralapa Innovation Center is located in the upper part of Cochabamba, along the old Santa Cruz Road.

Similarly, the National Potato Innovation Center is situated in the high valley of Cochabamba. In both these areas, agriculture serves as the predominant economic activity.

Furthermore, derived from climatic data collected from three distinct locales, specifically the Huarina, Tarata, and Toralapa weather stations, the climatic characteristics are presented

in Table 1 for the period spanning October 1, 2018, to June 2019. In addition, the soil composition varied across these locales: Tarata exhibited a sandy loam type, Toralapa featured a silty loam composition, while Huarina demonstrated a silty clay loam composition.

Table 1. Experimentation location of trials implemented in the 2018-2019 agricultural season.

Department	Municipality	Location	Temp. (°C)	PP (mm)	Georeferenced		
					Latitude (LS)	Longitude (LW)	Altitude (m.a.s.l.)
La Paz	Huarina	Cuyahuani	5-11	583	16° 13' 13.98"	68° 35' 8.75"	3839
Cochabamba	Tiraque	C.I.Toralapa	10-16	748	17°36'44.76"	66° 0'34.61"	2754
	Tarata	C.N.I. Potato	11-23	764	17°28'39.18"	65°39'14.64"	3536

C.I.= Innovation Center ; C.N.I.= National Innovation Center ; PP= precipitation

The research was conducted using a completely randomized block design (CRBD) with three repetitions in three different environments. Each experimental plot consisted of five rows, each measuring 5 m in length, with a spacing of 0.25 m between rows. The sowing was carried out using a continuous jet method at a density of 90 kg ha⁻¹. The genetic material evaluated included six varieties of forage oats, three obtained from Instituto Nacional de Innovación Agraria (INIA) Peru (INIA-Tayco, INIA-Villacanota, and INIA-Africana), while the remaining three were from the Centro de Investigación Forrajera (CIF) -Violeta, Seed Company in Cochabamba Department, Bolivia (Texas, Gaviota, and Aguila). Conventional tillage practices were employed, and the soil was rotated accordingly. Weed control was performed manually to maintain the crop free from unwanted vegetation.

Harvesting was conducted when each genotype reached 50% pasty grain stage, and the samples were weighed to determine the yield of green matter. To determine the dry matter content in percentage (%), a sub-sample weighing 500 g of fresh fodder was placed in a moisture-free kraft paper bag and dried in an oven at 105 °C until a constant minimum weight was achieved.

The obtained value was then multiplied by the fresh mass to calculate the dry matter yield of each respective experimental unit.

To obtain the results, a descriptive analysis of measures of central tendency and dispersion was performed. Additionally, skewness and kurtosis were determined as a normality test. Statistical differences were evaluated using a fixed combined analysis of variance, with a significance level set at $P < 0.01$ and 0.05 .

For the analysis of multi-environmental trials, the GGE biplot (Genotype, Genotype × Environment) and BLUP (Best Linear Unbiased Prediction) methodologies were employed. Detailed descriptions of both models can be found in Olivoto's work (2019). Data processing was carried out using the R Statistical Analysis System (R Core Team, 2019). For the multi-environment analyses, also known as Multi-Environment Trial (MET), the METAN package developed by Olivoto (2019) was utilized.

RESULTS AND DISCUSSION

Table 1 presents the average plant length for the studied varieties, which was determined to be 112.16 ± 19.87 cm. The average length of

leaves from the middle and upper thirds of the genotypes was 25.02 ± 9.46 cm, with a width of 1.62 ± 0.39 cm. In terms of biomass productivity, the national average for green matter yield is reported as 2.3 t ha^{-1} (INE & MDRYT, 2019). However, in this investigation, significantly higher averages of $41.30 \pm 17.57 \text{ t ha}^{-1}$ for green

matter yield and $10.29 \pm 3.51 \text{ t ha}^{-1}$ for dry matter yield were obtained, surpassing the national average. Moreover, the skewness and kurtosis values were found to be lower (± 1), indicating normality (Mishra et al., 2019). Therefore, the research values align with these parameters, except for the leaf length measurement.

Table 1. Descriptive statistics of six oats forage genotypes in localities of Huarina, Tarata, and Toralapa Innovation Center, during the 2018-2019 agricultural season.

Descriptive statistics	Variables					
	Plant height	Leaf length	Leaf width	Green matter yield	Dry matter percentage	Dry matter yield
Measures	--cm--	--cm--	--cm--	--t ha ⁻¹ --	--%--	--t ha ⁻¹ --
Mean	112.16	25.02	1.62	41.30	26.58	10.29
Standard deviation	19.87	9.46	0.39	17.57	6.22	3.51
Skewness	-0.80	1.05	0.01	0.12	0.40	-0.18
Kurtosis	0.14	0.32	-0.83	-0.92	-0.62	-0.67
Minimum	64.00	14.30	0.81	9.00	16.34	2.79
Maximum	148.00	50.00	2.50	74.40	41.77	16.34

Table 2. Analysis of variance for the variables PH (plant height), LL (leaf length), LW (leaf width), GMY (green matter yield), DMP (dry matter percentage) and DMY (dry matter yield).

Sources of variation	DF [‡]	Mean squares					
		AP	LL	LW	GMY	DMP	DMY
Block	2	563.33	99.84	0.33*	559.18*	22.82	27.72
Environment	2	3066.41**	1233.73**	1.26**	3129.07**	463.39**	23.61
Block (Env)	4	1054.47**	117.21*	0.18	746.30**	109.58**	27.49*
Genotypes	5	263.37	63.25	0.23*	182.21	13.53	14.77
Env×Gen	10	134.96	21.59	0.08	126.81	12.50	8.11
Exp. Error	30	225.85	35.73	0.07	127.11	14.91	9.51
CV ^{††} (%)		13.40	23.89	16.62	27.29	14.53	29.98
R ²		0.68	0.77	0.73	0.77	0.78	0.56

[‡]degrees of freedom; ^{††}coefficients of variation; R²= coefficient of determination; **P<0.01; *P<0.05

Based on the combined analysis of variance (Table 2), the genotypes did not exhibit significant differences ($P > 0.05$) except for leaf width ($P < 0.05$). However, significant differences ($P < 0.01$) were observed among the environments for plant height, leaf length, leaf width, green matter yield, and dry matter percentage. The lack of significant interaction among the evaluated variables could suggest that the genotypes displayed similar behavior across the assessed environments or indicate a potential limitation in the discriminatory capacity of the environments. Nevertheless, Fasahat et al., (2015)

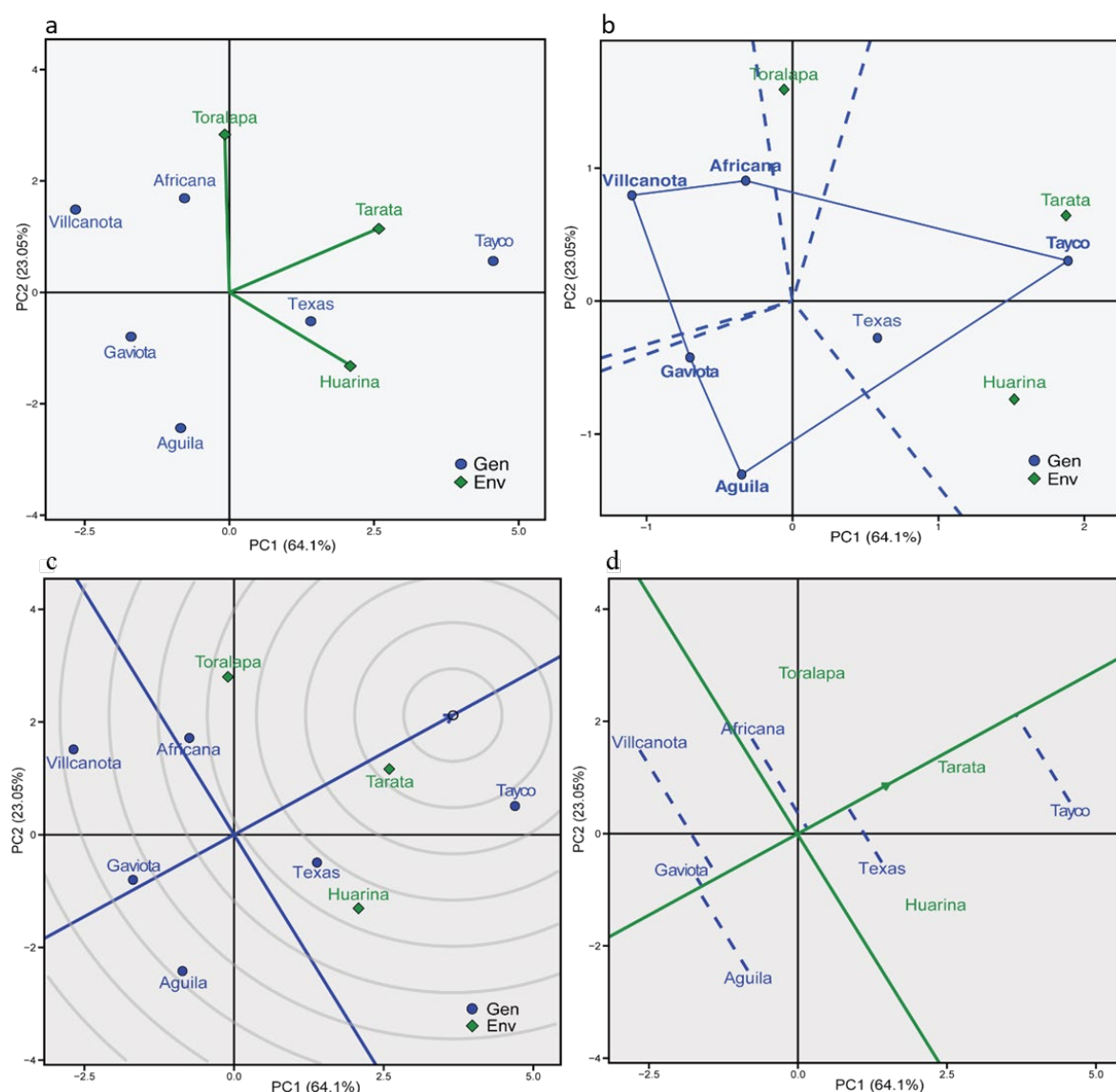
suggest that stability analyses based on various principles can enhance the identification of stable genotypes, even in the absence of interactions between parameters. Additionally, the coefficients of variation fell within acceptable ranges, and the coefficients of determination demonstrated a genetic expression above 50%.

In the GGE Biplot methodology, it was observed that the first two main components (PC1: 64.1% and PC2: 23.05%) accounted for a cumulative variance of 87.15%. This variance

was derived from the decomposition of singular genotype values (G) and interaction effects (GxE). According to Yan and Kang (2002), the first main component (PC1) represents the adaptability of genotypes, displaying a strong correlation with performance. Meanwhile, the second main component (PC2) indicates stability, with genotypes closer to zero being more stable. Based on this interpretation, the Texas variety meets the stability criteria, as it is

positioned closer to the origin on the first component. Additionally, Yan and Kang (2002) suggest that a polygonal view of a biplot is the most effective way to visualize the patterns of interaction between genotypes and environments. Therefore, in Figure 1b, the vertex genotypes—INIA-Tayco, Aguila, Gaviota, INIA-Villcanota, and INIA-Africana—exhibit higher dry matter yields in the respective environments represented by each sector.

Figure 1. GGE biplot analysis: a) basic biplot, b) who-won-where, c) genotype ranking (ideal genotype) and d) yield vs. genotype stability evaluated in three locations during the 2018-2019 agricultural season.



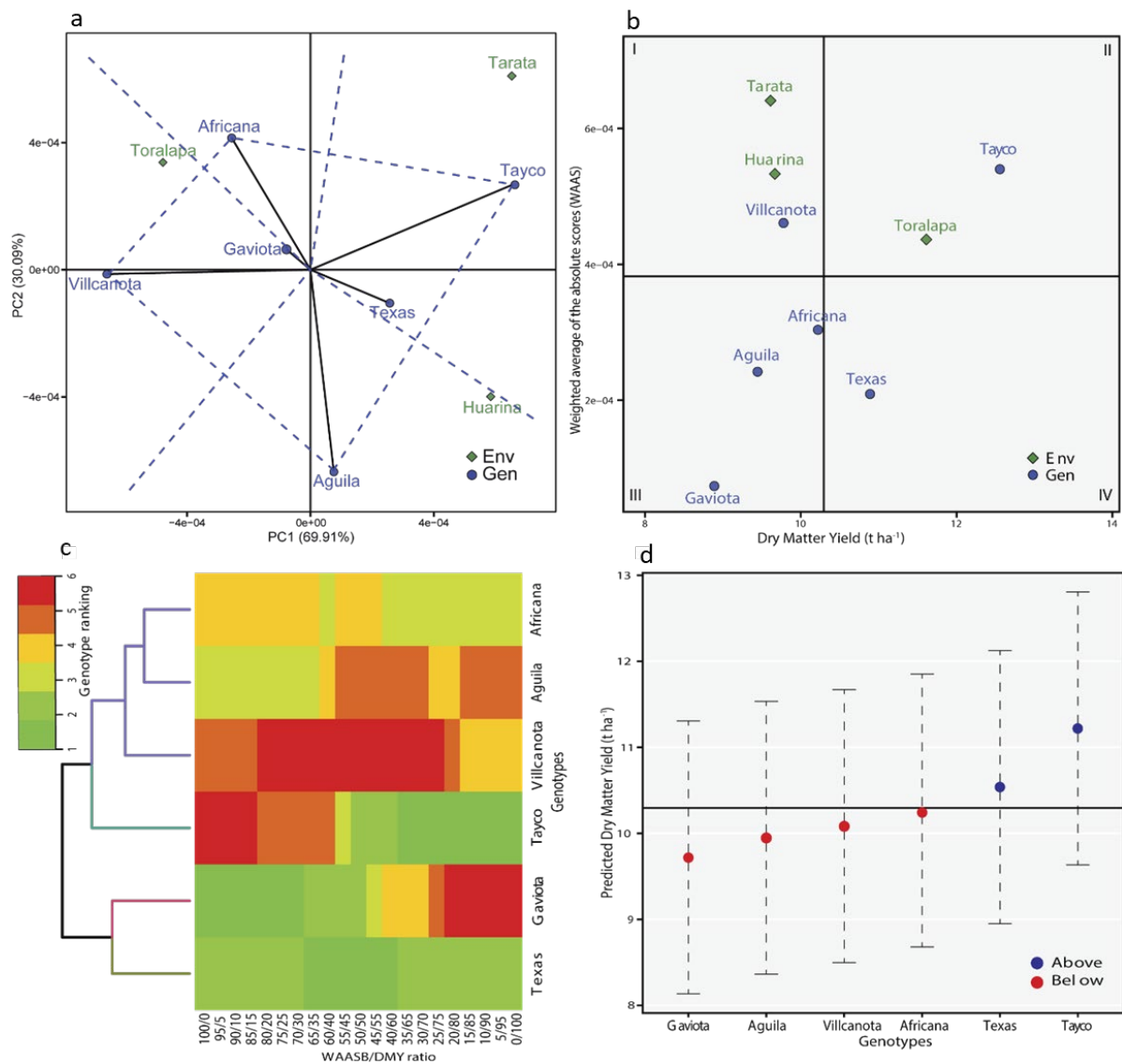
The center of the concentric circles (Figure 1c) represents the position of an ideal genotype, although such a genotype does not exist in reality. However, it serves as a reference for evaluation and allows for meaningful

inferences (Yan & Tinker, 2006). In this study, INIA-Tayco was positioned closer to one of the concentric circles, suggesting a higher dry matter yield capacity and greater stability. Therefore, INIA-Tayco can be considered relatively

closer to the ideal genotype. On the other hand, the genotypes INIA-Villcanota, Gaviota, and Aguila were positioned farther away from the ideal genotype and exhibited yields below the overall research average. In Figure 1d, INIA-Tayco demonstrated the highest average yield, while Aguila had the lowest. The average yields

followed the order of INIA-Tayco > Texas > INIA-Africana. Notably, INIA-Africana exhibited higher variability in yield (indicating lower stability), whereas INIA-Tayco and Texas demonstrated moderate stability with high dry matter yields.

Figure 2. Analysis of multi-environment trials using BLUP (Best Linear Unbiased Prediction): a) AMMI “who-won-where” b) dry matter yield \times weighted average of absolute scores (WAASB) c) classification of genotypes according to the weight considered for productivity and stability and d) BLUP plot of six varieties of forage oats evaluated in three locations during the 2018-2019 agricultural campaign.



In the AMMI biplot based on the WASSB model (Figure 2a), the cumulative variance of the first two components was 100%. This indicates that the results obtained from the mixed models provide a high level of technical and

statistical validity for the inferences of this research. Additionally, this variance value exceeds that obtained from the GGE biplot methodology (Figure 1). Based on the analysis of these two components, four varieties (INIA-Tayco, Aguila, INIA-Villcanota, and INIA-

Africana) were identified as winners, displaying consistent performance across three environments or sectors in terms of forage biomass production. These results demonstrate similarity between both analysis models.

Furthermore, Olivoto et al. (2019) propose a biplot classification into four sectors or classes (I, II, III, and IV) for joint interpretation of average performance and stability (Figure 2b). The classification in quadrant (I) refers to environments with high discrimination capacity and unstable genotypes with low productivity. Accordingly, Huarina and Tarata are considered environments with high discrimination capacity, and INIA-Villcanota is classified as an unstable variety with a yield below the overall average.

INIA-Tayco falls under quadrant II according to this analysis, indicating it is an unstable genotype. However, INIA-Tayco exhibits biomass productivity surpassing the general average, and Toralapa shows noteworthy capacity for environmental discrimination, deserving special attention. Genotypes in quadrant III could be considered stable based on their low WAASB values, but they demonstrate low biomass productivity. According to Olivoto et al. (2019), genotypes within quadrant IV are highly productive and widely adapted. Consequently, Texas emerges as the genotype meeting both productivity and stability criteria.

Figure 2c, which represents the heat map, displays the genotype classification based on the ratio of WAASB to biomass yield. According to Olivoto et al. (2019), the obtained ranges utilize a ratio of 100/0 to exclusively consider stability, and a ratio of 0/100 to solely consider productivity for genotype classification. In this context, the INIA-Tayco variety is classified as having both high productivity and stability, which aligns with the observed trends in Figure 2c and 2d, reflecting the predicted interpreta-

tion of BLUP. Furthermore, Texas is categorized as highly productive, and this designation is supported by the results depicted in Figure 2c and d.

CONCLUSIONS

Based on the application of two multi-environment analysis methodologies, namely GGE Biplot and BLUP, two genotypes (INIA-Tayco and Texas) were identified as possessing high forage productivity and stability characteristics. While INIA-Tayco and Texas hold great promise for enhancing forage productivity in Bolivia's valley and highland regions, it is imperative to conduct further evaluation and stability testing over three more seasons. Once the genotypes' stability is confirmed, an effective dissemination strategy should be implemented to ensure farmers can access and benefit from these superior forage varieties, leading to improved livestock production and agricultural sustainability in the region.

Acknowledgments

We would like to express our gratitude to Universidad Indígena Boliviana Tupac Katari-UNIBOL, Centro Nacional de Innovación de Papa, and Banco Nacional de Germoplasma for their valuable assistance and support throughout the development of this experiment.

Author contribution

Data analysis and visualization: F.S. Investigation: F.S. Writing-original draft: F.S. Review: F.M. Supervision: F.M. All coauthors reviewed the final version and approved the manuscript before submission.

Conflict of interest

The authors declare no conflict of interest.

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