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## PLANT BREEDING - Article

# Genotype by environment interaction and adaptability of photoperiod-sensitive biomass sorghum hybrids

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**ABSTRACT:** Biomass sorghum is an alternative feedstock to cogenerate energy and produce second-generation ethanol. The aim of this study was to evaluate the Genotype by Environment Interaction (GEI) in biomass sorghum and to identify the hybrids that associate high adaptability and stability using the Toler nonlinear regression, the Genotypes plus Genotype by Environment (GGE) biplot, and the Annicchiarico recommendation index. Thirty-three experimental photoperiod-sensitive single-cross hybrids and three checks were evaluated in relation to the traits: flowering time, plant height, moisture content, green mass yield, and dry mass yield. It

was observed that the effects of hybrids, environment, and GEI were expressive. The GEI was predominantly complex for the traits related to the biomass yield. The Toler, GGE biplot and Annicchiarico methods show complementarity. The experimental hybrids 1, 8, 22, 31 and 33 are promising because of associating stability and lower recommendation risk. The hybrids 1 and 8 present broad adaptability, while the hybrids 22, 31 and 33 exhibit specific adaptability to high quality environments.

**Key words:** *Sorghum bicolor*, single-cross, Toler nonlinear regression, recommendation index, GGE biplot.

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## INTRODUCTION

Currently, there is a strong world demand for energy, and there is great concern with countries whose energy matrix is heavily based on nonrenewable energy sources, such as oil and oil products. Brazil is in a prominent position compared to some countries, since renewable energy sources supply 43.5% of the Brazilian energy matrix (MME 2017). Brazilian geographic and agroclimatic characteristics make possible to explore several renewable sources of energy, such as bioenergy crops, which is the production of biomass in the process of energy cogenerated by burning in high pressure boilers (Naik et al. 2017).

Biomass sorghum presents a promising feedstock for energy cogeneration because it exhibits productivities up to 150 Mg·ha<sup>-1</sup> of fresh mass in a cycle of only five months with entirely mechanized cultivation process (Rooney et al. 2007; Mullet 2017). Public or private breeding programs for biomass sorghum are relatively recent. Brazilian Agricultural Research Corporation (Embrapa) for Maize and Sorghum has conducted a biomass sorghum breeding program focused on obtaining hybrids with high energetic potential from Multi-Environment Trials (MET) for assessing the value of cultivation and use (VCU) of new hybrids prior to recommendation (Parrella et al. 2010).

There are few studies with biomass sorghum hybrids based on MET conducted in a wide diversity of environments in terms of geographic, climate and soil conditions. Nevertheless, some of these studies have detected pronounced Genotype by Environment Interaction (GEI) effect (Souza et al. 2014; Castro et al. 2015; Andrade et al. 2016). Biomass sorghum is characterized by its wide adaptability to different growing environments provided by evolutionary process. Thus, more studies on GEI based on MET can help to discriminate the hybrids by environmental sensitivity, and also allow to better describe the interrelationship among environments, and specific GEI. In this regard, the hybrid's characterization according to adaptability and stability for energetic biomass yield is essential to release commercially superior cultivars.

There are several methodologies to study adaptability and stability. A frequent question of the breeder is about which method to choose. Studies have showed that some methods traditionally used for evaluating phenotypic stability are not complementary, because they are based on similar concepts of stability (Bornhofen et al. 2017).

Rono et al. (2016) suggested that this choice can be made according to the profile and the characteristics of the data set to be analyzed. For instance, nonparametric methods might be used when data do not follow clearly any probability distribution. On the other hand, some studies have proposed to apply complementary methods (Ferreira et al. 2006; Borges et al. 2000; Figueiredo et al. 2015). Borges et al. (2000) suggested to use simultaneously the Toler method (Toler and Burrows 1998) and the Annicchiarico reliability index (Annicchiarico 1992). The Toler method provides information about genotype response patterns, while the Annicchiarico method provides an easy interpretation about adaptability and phenotypic stability (Borges et al. 2000; Carvalho et al. 2016). Ferreira et al. (2006) suggested to cope with GEI by applying a multivariate approach, such as the Additive Main Effects and Multiplicative Interaction (AMMI) and the Genotypes plus Genotype by Environment (GGE) Biplot, complemented by the Toler method. In general, all authors emphasize that the use of complementary methods simultaneously might help agronomists and breeders to identify promising genotypes that associate desirable response pattern and low recommendation risk across environments.

The aim of this study was to evaluate the GEI in biomass sorghum and to identify hybrids that associate high adaptability and stability using the Toler and Annicchiarico methods.

## MATERIAL AND METHODS

### Environments and Genotypes

Data from 10 environments of the VCU trials of biomass sorghum in the 2014/2015 agricultural crop, coordinated by Embrapa Maize and Sorghum, were used. The description of the environments where the experiments were set up regarding geographic aspects, climate and dates of sowing and harvesting are presented in Table 1.

In these VCU trials, 36 genotypes were evaluated, being 33 experimental photoperiod-sensitive single-cross hybrids of biomass sorghum (201429B001 to 201429B033), developed by the biomass sorghum breeding program of Embrapa Maize and Sorghum, and three checks: a biomass sorghum cultivar 'BRS 716' (34), and two forage sorghum cultivars ['Volumax' (35) and 'BRS 655' (36)]. The cultivars 'BRS 716' e 'BRS 655' belongs to Embrapa Maize and Sorghum, while 'Volumax' belongs to Monsanto Company. →

**Table 1.** Environment description of the value for cultivation and use (VCU) trials of biomass sorghum according to the geographical aspects latitude (Lat), longitude (Long), altitude (Alt), climate (Cl); and to the cultivation aspects planting dates (PD) and harvest dates (HD) in the 2014/2015 agricultural crop year.

Environments	Lat	Long	Alt	PD	HD	Cl <sup>1</sup>
Nova Porteirinha – MG	15° 48' S	43° 18' O	85 m	22/11/14	20/05/15	Aw
Dracena – SP	21° 28' S	51° 31' O	421 m	15/11/14	28/04/15	Aw
Uberlândia – MG	18° 55' S	48° 16' O	863 m	06/12/14	24/05/15	Aw
Sete Lagoas – MG	19° 27' S	44° 14' O	761 m	06/11/14	12/05/15	Aw
Lavras – MG	21° 14' S	45° 00' O	919 m	22/11/14	12/05/15	Cwa
Goiânia – GO	16° 40' S	49° 15' O	823 m	18/12/14	23/05/15	Aw
Dourados – MS	22° 13' S	54° 48' O	430 m	11/11/14	13/05/15	Aw
Sinop – MT	11° 50' S	55° 38' O	384 m	04/12/14	11/05/15	Aw
Pelotas – RS	31° 46' S	52° 20' O	7 m	06/12/14	25/03/15	Cfa
Guaira – SP	24° 04' S	54° 15' O	220 m	26/11/14	14/04/15	Cfa

<sup>1</sup>Koppen climate classification.

## Experimental Planning and conducting

The experiments were laid out in a 6 × 6 triple lattice design. The plots were constituted by four 5.0 m long rows, spaced 0.7 m apart, considering only the two central lines as useful.

The experiments were set up and conducted following the same directions for VCU trials of biomass sorghum coordinated by Embrapa Maize and Sorghum. The furrowing of the area and simultaneous fertilization of the planting was done by application of NPK 8-28-16 formulation, according to soil analyses, and recommendation for the crop. On this occasion, 1/3 of nitrogen was applied. The seeding was carried out manually at a depth of 3 to 4 cm. The thinning was performed about 10 to 15 days after emergence, leaving 10 plants per linear meter. The population density was 140,000·ha<sup>-1</sup> plants. Cover fertilization of the remaining 2/3 of nitrogen was applied 30 to 35 days after emergence. The control of weeds was carried out by the application of herbicides, especially atrazine base with dosage of 3 kg active ingredient per hectare, complemented by mechanical weeding. Applications of insecticides and fungicides for control of insect pests and diseases were carried out, when the incidence was observed.

The harvest was done manually with variable time among the environments, from 109 (Pelotas) to 187 (Sete Lagoas), days after sowing or planting (DAP), with a mean of 162 DAP. The harvest was accomplished when the grains reached the farinaceous or hard dough stage.

Morphoagronomic traits evaluated were: a) Flowering time (FLOW): counting the number of days elapsed from planting to the point where 50% of the plants in the plot were flowering, that is, 50% of the flowers of the panicle of each plant with release of pollen; b) Plant height (PH): measured in meters from the soil surface to the apex of the panicle. Data were taken from five plants randomly selected from the plot area and averaged; c) Total green mass yield (GMY): determined by weighing in kg·plot<sup>-1</sup> of all whole plants of the area of each plot using suspension scale, having been cut 10 cm from the soil surface. GMY data were then converted to tons per hectare; d) Total dry mass yield (DMY): five whole plants without panicles were randomly taken at each plot, which were passed in an electric forage chopper. Afterwards, the material was homogenized and a sample was taken for oven drying with forced ventilation at 60 °C. The dry mass expressed as a percentage (% DM) was determined by the difference between the fresh and dry matter weights. Subsequently, the DMY (tons per hectare) was calculated for the product of the GMY and the % DM; e) Moisture content (MC): calculated by the difference between the weights of the fresh and dry matter and then expressed as percentage.

The PH and GMY traits were assessed in all environments, while the FLOW was evaluated in only six environments (Sete Lagoas-MG, Sinop-MT, Lavras-MG, Nova Porteirinha-MG, Uberlândia-MG and Goiânia-GO). The DMY and MC traits were evaluated in Sete Lagoas-MG, Sinop-MT, Lavras-MG, Nova Porteirinha-MG, Uberlândia-MG and Dourados-MS.

## Statistical Analysis

Individual (per environment) and multi-environment analyses with recovery of interblock information were performed using the lme4 R package (Bates et al. 2015) by the univariate mixed model approach. For the statistical models, we assumed blocks within replications, and errors as random effects following a normal distribution, common and independent variance. Selective accuracy was estimated for each environment for evaluating the experimental precision according to Resende and Duarte (2007):  $\hat{r}_{gg} = \sqrt{(1 - 1/F_c)}$ , where  $F_c$  is the value of the statistics  $F$  of Fisher-Snedecor associated with the genotype effect in the analysis of variance. The coefficient of experimental variation per environment was also estimated to measure the experimental precision according to Garcia (1989) and Pimentel-Gomes (2009):  $CV_e = s_e/\bar{y} \times 100$ , where  $s_e$  is the square root of the error mean square, and  $\bar{y}$  is the overall mean.

Previously to multi-environment analysis, the Hartley's homogeneity test of the residual variances was carried out as suggested by Pimentel-Gomes (2009). The variation due to GEI was partitioned into simple and complex parts, based on the differences of variation among genotypes, and lack of correlation between the phenotypic performances of genotypes across environments (Robertson 1959; Cruz and Castoldi 1991).

The analyses of phenotypic stability were performed by the Toler nonlinear regression method (Toler and Burrows 1998), and by the recommendation index proposed by Annicchiarico (1992) using the Stability software (Ferreira 2015). The GGE Biplot method (Yan and Tinker 2006) was also performed using the GGEbiplots R package (Dumble 2017).

The response pattern of each genotype in the evaluated environments was described by the Toler method, which was adjusted according to the following nonlinear models (Toler and Burrows 1998):

$$\bar{y}_{ij} = \alpha_i + \beta_i \mu_j + \delta_{ij} + \varepsilon_{ij}, \text{ and}$$

$$\bar{y}_{ij} = \alpha_i + [Z_j \beta_{1i} + (1 - Z_j) \beta_{2i}] \mu_j + \delta_{ij} + \varepsilon_{ij},$$

where  $\bar{y}_{ij}$ : adjusted mean of the genotype  $i$  in the environment  $j$ ;  $\alpha_i$ : intercept value at  $\mu_j = 0$  associated with genotype  $i$ ;  $\beta_{1i}$  and  $\beta_{2i}$ : regression coefficients related to response sensitivity of the genotype  $i$  in environments of lower and higher quality, respectively;  $\mu_j$ : environmental index that denotes

the effect of the environment  $j$ ;  $\beta_i$ : regression coefficient quantifying the response sensitivity of the genotype  $i$  in different environments;  $\delta_{ij}$ : deviation of the regression of the genotype  $i$  in the environment  $j$ ;  $\varepsilon_{ij}$ : average experimental error;  $Z_j = 1$ , if  $\mu_j \leq 0$  and,  $Z_j = 0$ , if  $\mu_j > 0$ .

As for the Toler method, the genotypes were classified in five groups, according to their response patterns over environments (Table 2): Group A - Criterion: reject  $H_0$ :  $\beta_{1i} = \beta_{2i}$ , with  $\beta_{1i} < 1 < \beta_{2i}$ ; Group B - Criterion: does not reject  $H_0$ :  $\beta_{1i} = \beta_{2i}$ , reject  $H_0$ :  $\beta_{1i} = 1$ , but common  $\beta_i$  is higher than 1; Group C - Criterion: does not reject  $H_0$ :  $\beta_{1i} = \beta_{2i}$ , accept  $H_0$ :  $\beta_{1i} = 1$ ; Group D - Criterion: does not reject  $H_0$ :  $\beta_{1i} = \beta_{2i}$ , reject  $H_0$ :  $\beta_{1i} = 1$ , but common  $\beta_i$  is lower than 1; Group E - Criterion: reject  $H_0$ :  $\beta_{1i} = \beta_{2i}$ , with  $\beta_{1i} > 1 > \beta_{2i}$ . Additionally, some results of the Toler method for the trait DMY were plotted in scatter plots using the ggplot2 R package (Wickham 2016).

**Table 2.** Grouping of genotypes by the Toler method for the traits flowering time (FLOW), plant height (PH), moisture content (MC), green mass yield (GMY), and dry mass yield (DMY)

Group	FLOW and MC	PH, GMY and DMY
A	Convex response and doubly undesirable	convex response and doubly desirable, i.e., consistent performance in below average environments and responsiveness in above average environments
B	Simple linear response and undesirable	simple linear response and desirable only in above average environments
C	Simple linear response not deviating from the average response	simple linear response not deviating from the average response
D	Simple linear response and desirable	simple linear response and desirable only in below average environments
E	Concave response and doubly desirable	concave response and doubly undesirable, i.e., sensitivity in below average environments and unresponsive to above average environments

The recommendation index proposed by Annicchiarico was estimated based on the relative average response of the environments from the following expression:  $I_i = p_i - z_{(1-\alpha)} s(p_i)$ , where:  $p_i$ : relative average response (%) of the genotype  $i$ ;  $z_{(1-\alpha)}$ : upper quantile of the standard normal distribution for a confidence level  $1-\alpha$ , in this study,  $\alpha = 0,25$  was pre-established, and  $s_{p_i}$ : standard deviation of the values of the relative means of the genotype  $i$  in the different environments. Moreover, we

computed the Annicchiarico index of the genotype  $i$  based on the relative average response to the check 'BRS 716' ( $I_{i(BRS716)}$ ), because this check is a single biomass sorghum cultivar.

The analysis of GGE Biplot method was carried out for the trait DMY, according to the following model (Yan and Tinker 2006):

$$\bar{y}_{ij} = \lambda_1 \gamma_{i1} \delta_{j1} + \lambda_2 \gamma_{i2} \delta_{j2} + \rho_{ij},$$

where  $\lambda_1$  and  $\lambda_2$  are singular values of the first and second Principal Components (PC) associated with the matrix of the effects of genotypes added to effects of genotype x environment interactions;  $\gamma_{i1}$  and  $\gamma_{i2}$  are eigenvectors of the first and second PC associated with the effect of the genotype  $i$ ;  $\delta_{j1}$  and  $\delta_{j2}$  are eigenvectors of the first and second PC associated with the effect of the environment  $j$ ;  $\rho_{ij}$  is the residual of the model associated with the genotype  $i$  in the environment  $j$ .

Biplots of the scores associated with two first PC were generated to better understanding the interrelationship

among genotypes and/or environments, as proposed by Yan and Tinker (2006).

## RESULTS AND DISCUSSION

The experimental precision was evaluated by observing the estimates of the selective accuracy ( $\hat{r}_{gg}$ ) and the coefficient of the experimental variation ( $CV_e$ ) (Pimentel-Gomes, 2009; Resende and Duarte, 2007). Overall, accuracy was high for all traits measured, indicating high reliability of experimental data for selective purposes. The values of  $\hat{r}_{gg}$  ranged from 28.17% (GMY, Nova Porteirinha) to 99.89% (FLOW, Lavras), while  $CV_e$  values ranged from 0.80% (FLOW, Lavras) to 22.94% (DMY, Sete Lagoas). Therefore, the estimates of  $\hat{r}_{gg}$  and  $CV_e$  that have shown the traits GMY and DMY were more influenced by environmental factors than the FLOW, PH and MC (Table 3).

The effect of environment was expressive to the phenotypic variation for all traits (Tables 3 and 4). The differences among

**Table 3.** Estimates of the parameters general mean ( $\bar{y}$ ), selective accuracy ( $\hat{r}_{gg}$ , %), and experimental coefficient of variation ( $CV_e$ , %) for flowering time (FLOW), plant height (PH), moisture content (MC), green mass yield (GMY) and dry mass yield (DMY) for the evaluation of genotypes of biomass sorghum in ten environments in the 2014/15 agricultural crop year.

Parameter	Environments <sup>1</sup>									
	SL	SI	LA	NP	UB	GO	DO	DR	PE	GU
<b>FLOW (days after sowing)</b>										
$\bar{y}$	143	116	142	141	125	115	-	-	-	-
$\hat{r}_{gg}$ (%)	99.45	99.70	99.88	99.25	98.60	99.51	-	-	-	-
$CV_e$ (%)	2.36	1.24	0.80	2.80	1.58	1.35	-	-	-	-
<b>PH (m)</b>										
$\bar{y}$	4.85	5.11	4.95	4.44	4.71	4.33	4.04	3.70	2.53	4.99
$\hat{r}_{gg}$ (%)	95.23	97.03	95.89	95.63	96.60	59.75	97.35	96.12	72.72	98.07
$CV_e$ (%)	6.40	5.17	6.15	6.08	4.88	12.95	4.73	6.49	8.64	4.05
<b>MC (%)</b>										
$\bar{y}$	66.66	69.12	66.77	66.95	68.76	-	75.25	-	-	-
$\hat{r}_{gg}$ (%)	80.99	82.75	28.48	80.94	74.93	-	90.91	-	-	-
$CV_e$ (%)	3.84	3.69	6.64	2.91	3.83	-	2.12	-	-	-
<b>GMY (ton·ha<sup>-1</sup>)</b>										
$\bar{y}$	42.23	65.48	82.61	43.08	51.52	51.75	102.98	55.30	36.57	87.42
$\hat{r}_{gg}$ (%)	86.27	94.26	92.27	71.94	82.34	86.32	88.69	89.98	81.95	75.64
$CV_e$ (%)	21.11	10.75	13.24	17.21	21.40	15.75	17.43	18.26	14.95	18.25
<b>DMY (ton·ha<sup>-1</sup>)</b>										
$\bar{y}$	14.18	20.32	27.24	14.17	16.14	-	25.22	-	-	-
$\hat{r}_{gg}$ (%)	86.62	92.61	82.66	28.17	87.58	-	87.56	-	-	-
$CV_e$ (%)	22.94	13.71	19.43	19.55	20.44	-	17.74	-	-	-

<sup>1</sup>SL (Sete Lagoas), SI (Sinop), LA (Lavras), NP (Nova Porteirinha), UB (Uberlândia), GO (Goiânia), DO (Dourados), DR (Dracena), PE (Pelotas), and GU (Guaíra).

**Table 4.** Summary of the analysis of variance and percentages of the genotype x environment interaction in the simple and complex types for flowering time (FLOW), plant height (PH), moisture content (MC), green mass yield (GMY), and dry mass yield (DMY) for the evaluation of genotypes of biomass sorghum in the 2014/15 agricultural crop year.

Trait	Genotypes			Environment			Genotype x Environment				
	DF	FC	% SQ	DF	FC	% SQ	DF	F <sub>c</sub>	% SQ	% Simple	% Complex
FLOW (days)	35	425.39**	44.37	5	721.17**	43.05	175	24.29**	12.57	42.51	57.49
PH (m)	35	74.00**	26.84	9	188.80**	65.70	315	2.29**	7.46	26.85	73.15
MC (%)	35	5.60**	16.94	5	15.31**	59.27	175	1.54**	23.79	2.18	97.82
GMY (t·ha <sup>-1</sup> )	35	20.11**	16.33	9	124.69**	77.35	315	2.49**	6.31	25.14	74.86
DMY (t·ha <sup>-1</sup> )	35	12.47**	34.19	5	40.56**	56.51	175	2.18**	9.30	15.10	84.90

DF: degree of freedom. SQ: sum of squares. %Simple. %Complex: percentages of the SQ due to simple and complex parts of the genotype by environment interaction. respectively. \*\* Significant at the 1% probability level by the Fisher-Snedecor's F test (F<sub>c</sub>).

environments are related to macro-environmental factors, such as latitude, altitude, climate and soil (Table 1). This has been highlighted in other studies with biomass sorghum (Castro et al. 2015), as well as other types of sorghum, as sweet sorghum (Figueiredo et al. 2015), forage sorghum (Mullet 2017) and grain sorghum (Batista et al. 2017).

The amplitude of variation of the means among the environments was 28 days, 2.58 m, 8.59%, 66.41 t·ha<sup>-1</sup> and 13.07 t·ha<sup>-1</sup> for FLOW, PH, MC, GMY and DMY, respectively (Table 3). It is important to stand out that the genotypes presented low relative performance in Pelotas environment, which might be explained by the high latitude, since the genotypes are photoperiod sensitive (Table 3). The daylength in regions of high latitudes is less than 12 h and 20 min, which contributes to initiate early floral development and, therefore, decreasing the PH, GMY and DMY traits (Parrella et al. 2010; Rooney and Aydin 1999).

The variation among the genotypes was expressive for all traits (Tables 4 and 5). For FLOW, the cultivars 'Volumax' (35) and 'BRS 655' (36) were the earliest, because they are photoperiod nonsensitive. The photoperiod-sensitive genotypes of biomass sorghum ranged from 120 days (28) to 143 days (34), with emphasis on the later genotypes 34, 13, 6, 15, 1 and 8. For PH, the means ranged from 2.35 m (36) to 4.88 m (22), standing out the genotypes 22, 26, 27, 32, 23, 29, 33, 25 and 20, with a mean height of 4.73 m.

Of the requirements highlighted by the thermoelectric power plants for biomass burning and energy cogeneration, it is estimated that the genotypes must present a biomass moisture content of around 50% or 55% (May et al. 2013). The experimental photoperiod-sensitive hybrids presented MC higher than 60%, ranging from 65.65% (28) to 72.30%

(19) (Table 4). These values might be considered high for burning. Edaphoclimatic and crop management factors might influence the MC in the plant at harvest time and biomass processing (Milar 2009).

In terms of the ideal genotype for genetic improvement and commercial exploitation, the hybrids must also combine high biomass production. For the variable GMY, the hybrids 33, 31 and 13 were the most productive with a mean of 74.77 t·ha<sup>-1</sup> (Table 5). In relation to DMY, the most promising hybrids were 31, 33, 1, 22, 34 and 8 (Table 5). However, they presented on average 23.37 t·ha<sup>-1</sup> of DMY, below the desired level of around 50 t·ha<sup>-1</sup> (Parrella et al. 2010). This low performance of biomass sorghum hybrids is also found in the literature (Rooney et al. 2007; Silva et al. 2015; Mullet 2017), what reinforce the need of improvement of this crop to obtain better genotypes.

Another point that must be taken into account in the selection of genotypes of biomass sorghum is that they must present high performance in different growing environments. In this case, the GEI may difficult recommendation of the hybrids. For the MC, the GEI contributed with 23.79% of the phenotype variation, and for FLOW this contribution was 12.57%. For PH, GMY and DMY, the relative contribution was lower, with values of 7.46%, 6.31% and 9.30%, respectively (Table 4).

It can be observed that in all the evaluated traits there was greater participation of the GEI of the complex type (Table 4). This indicates a lack of correlation in the average performance of the genotypes evaluated in the tested environments (Robertson 1959; Cruz and Castoldi 1991) and a possibility of the presence of genotypes adapted to specific environments (Ramalho et al. 2012). In order

**Table 5.** Adjusted means and groups by the Toler method of biomass sorghum genotypes (ID), 33 experimental hybrids [201424B001 (1) to 201424B033 (33)] and the cultivars 'BRS 716' (34), 'Volumax' (35) and 'BRS 655' (36), for flowering time (FLOW, days after sowing), plant height (PH, m), moisture content (MC, %), green mass yield (GMY, ton/ha), and dry mass yield (DMY, ton/ha) in the 2014/15 agricultural crop year.

ID	FLOW		PH		MC		GMY		DMY	
	Mean	Toler	Mean	Toler	Mean	Toler	Mean	Toler	Mean	Toler
1	142	E	4.3	A	67	B	70	B	24	C
2	127	E	4.5	B	67	B	64	E	20	C
3	130	A	4.5	B	69	B	63	B	20	B
4	126	D	4.3	B	69	B	59	D	20	C
5	140	E	4.3	C	70	B	66	E	20	C
6	142	E	4.3	C	71	B	65	B	19	B
7	125	A	4.3	C	67	B	57	C	18	C
8	141	B	4.4	B	68	C	70	B	23	C
9	130	A	4.4	C	69	C	67	D	20	C
10	130	B	4.3	B	71	B	61	D	19	C
11	128	A	4.3	C	69	C	55	D	17	C
12	127	D	4.3	C	68	A	64	B	20	B
13	142	E	4.3	A	71	B	73	E	21	B
14	132	B	4.2	C	68	D	63	B	21	B
15	142	E	4.4	E	70	B	66	B	19	C
16	132	A	4.3	C	70	C	63	D	19	C
17	138	A	4.3	D	70	B	62	D	19	B
18	133	A	4.4	B	72	D	61	D	17	D
19	125	A	4.1	D	72	D	58	D	17	C
20	135	B	4.7	B	67	B	62	B	20	C
21	125	A	4.5	C	67	D	55	A	20	C
22	136	A	4.9	B	70	D	72	B	23	B
23	134	A	4.7	B	69	D	64	E	20	C
24	122	A	4.6	B	66	D	62	D	21	C
25	123	E	4.7	C	68	C	57	E	17	C
26	137	B	4.8	B	68	A	65	E	21	C
27	127	A	4.8	A	69	B	61	B	20	C
28	120	A	4.5	D	66	E	58	A	22	C
29	138	A	4.7	A	69	D	68	D	21	C
30	123	D	4.5	C	66	C	60	A	20	B
31	138	B	4.6	B	68	B	75	B	25	B
32	133	E	4.7	C	68	C	64	A	21	C
33	139	B	4.7	B	69	C	77	B	24	B
34	143	B	4.6	C	68	A	70	B	23	C
35	93	E	2.8	D	70	D	34	D	11	D
36	82	E	2.4	D	71	E	24	D	7	D

to study more clearly the influence of GEI on adaptability and stability of the biomass sorghum genotypes in question, it is necessary to adopt additional biometric procedures, such as regression methods, multivariate approaches, and the recommendation index proposed by Annicchiarico

(1992). However, before to present the analyses using these methods is important to observe that this study was based on a single agricultural crop year. Thus, this model is unable to dissociate the genotype x environment x year interaction caused by unpredictable factors. Evaluations of MET across

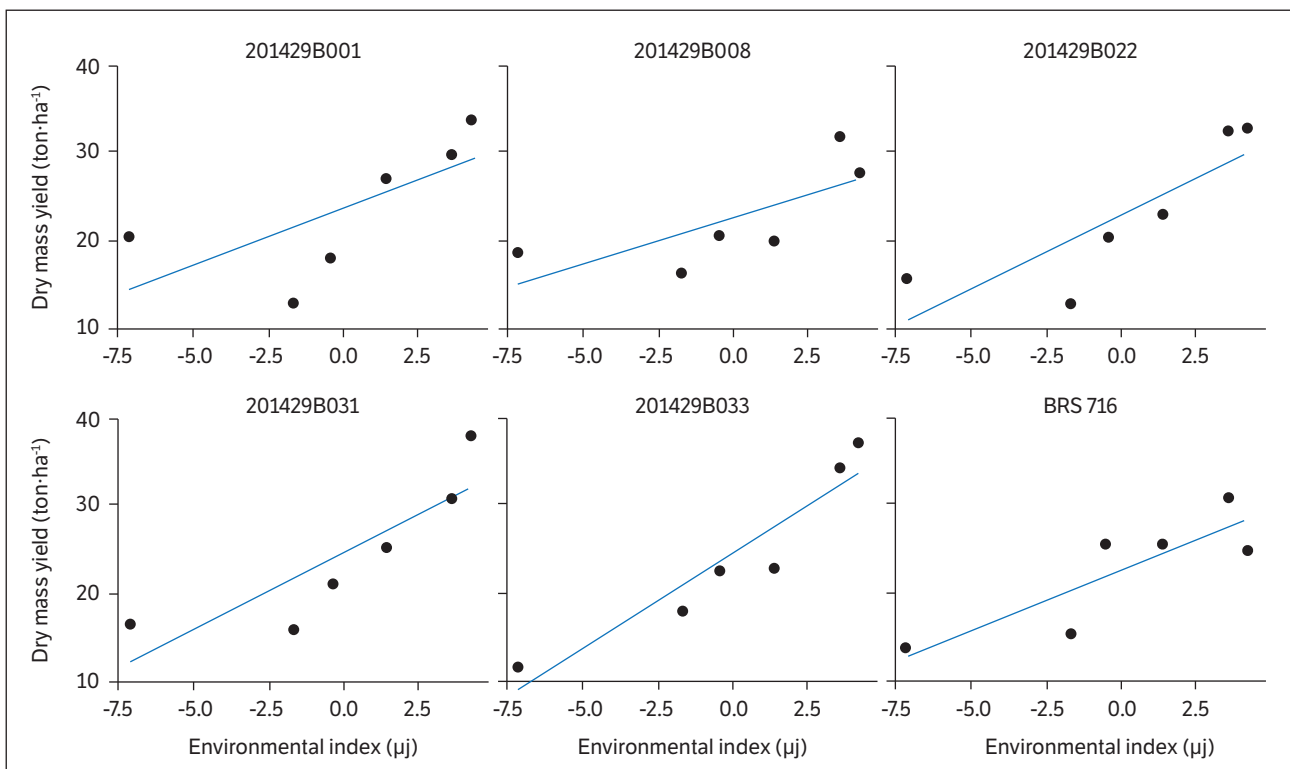
years are indispensable to dissociate the repeatable part of the GEI, and eventually it can be explored for the definition of mega-environments, and the safer recommendation of hybrids (Yan 2016). Despite this limitation, some important results in a single agricultural year can be obtained and might help breeders in a breeding program.

For all the traits, the genotypes showed variable response patterns by the Toler method (Table 5). For FLOW and MC, we adopted a particular interpretation of the groups classified by Toler (Table 2), where D and E response patterns describe the behavior that is closest to the desirable. These traits might be considered components of the general adaptability. In this case, breeders desire to reduce the flowering time without compromise the accumulation of biomass. Moisture in the biomass is directly linked to DMY and burning efficiency. It is suggested MC not superior to 55% at the harvest. The genotypes 1, 6, 13 and 15 stood out as the later ones with concave response pattern (E) and high predictability (correlation between observed and fitted means –  $r \geq 0.87$ ). For MC, genotypes 21 and 24 (pattern D) and 28 (pattern E) were highlighted with lower mean MC ( $MC \leq 67\%$ ) and predictability above 80%.

For PH, GMY, and DMY, we adopted a conventional interpretation of the groups classified by Toler (Table 2). In

the case of PH, the hybrids 27 and 29 associated high mean and desirable doubly response pattern (A) (Table 5). As for traits GMY and DMY, there is not any genotype with convex response pattern, and high mean. The hybrids with higher GMY (13, 31 and 33) were more adapted to high quality environments or Toler group B (31 and 33), while the hybrid 13 presented an undesirable doubly behavior (Table 4). For the DMY, the experimental hybrids 1, 8, 22, 31 and 33, and the check 34 showed the highest average yield of dry biomass, but with different response pattern across environments. The hybrids 1, 8 and ‘BRS 716’ presented broad adaptability and were classified in the Toler group C, while the hybrids 22, 31 and 33 were more adapted to above average environments – Toler group B (Fig. 1). These results show a problem often faced by breeders to identify productive genotypes with a desirable double response pattern (Rosse and Vencovsky 2000).

The use of two or more methods to study adaptability and phenotypic stability is only justified if there is complementarity (Borges et al. 2000; Ferreira et al. 2006). The use of the Annicchiarico (1992) method to complement the Toler (1990) method is justified by its ease of analysis and interpretation, as well as to associate in a single parameter the description of the genotype for its adaptability and phenotypic stability (Annicchiarico 1992).



**Figure 1.** Observed (black dots) and fitted means by Toler nonlinear regression (blue line) of biomass sorghum genotypes for dry matter yield.



For the PH, GMY and DMY traits, 39.40%, 18.18% and 18.18% of the experimental hybrids had a high reliability index (above 100%) as a function of the average of the environments, respectively, that is, they had a lower risk of adoption

(Table 6). In the case of FLOW and MC traits, the interpretation of the Annicchiarico index must be performed contrary, once that reduced flowering time and low moisture is desired. Considering a threshold for the reliability index less than

**Table 6.** Estimates of the Annicchiarico reliability index ( $I$ ) of the biomass sorghum genotypes (ID), 33 experimental hybrids [201424B001 (1) to 201424B033 (33)] and the cultivars 'BRS 716' (34), 'Volumax' (35) and 'BRS 655' (36), based on the mean of the environments ( $I_i$ ) and the check 'BRS716' ( $I_{i(BRS716)}$ ) for flowering time (FLOW, days), plant height (PH, m), moisture content (MC), green mass yield (GMY, ton/ha), and dry mass yield (DMY, ton·ha<sup>-1</sup>) in the 2014/15 agricultural crop year.

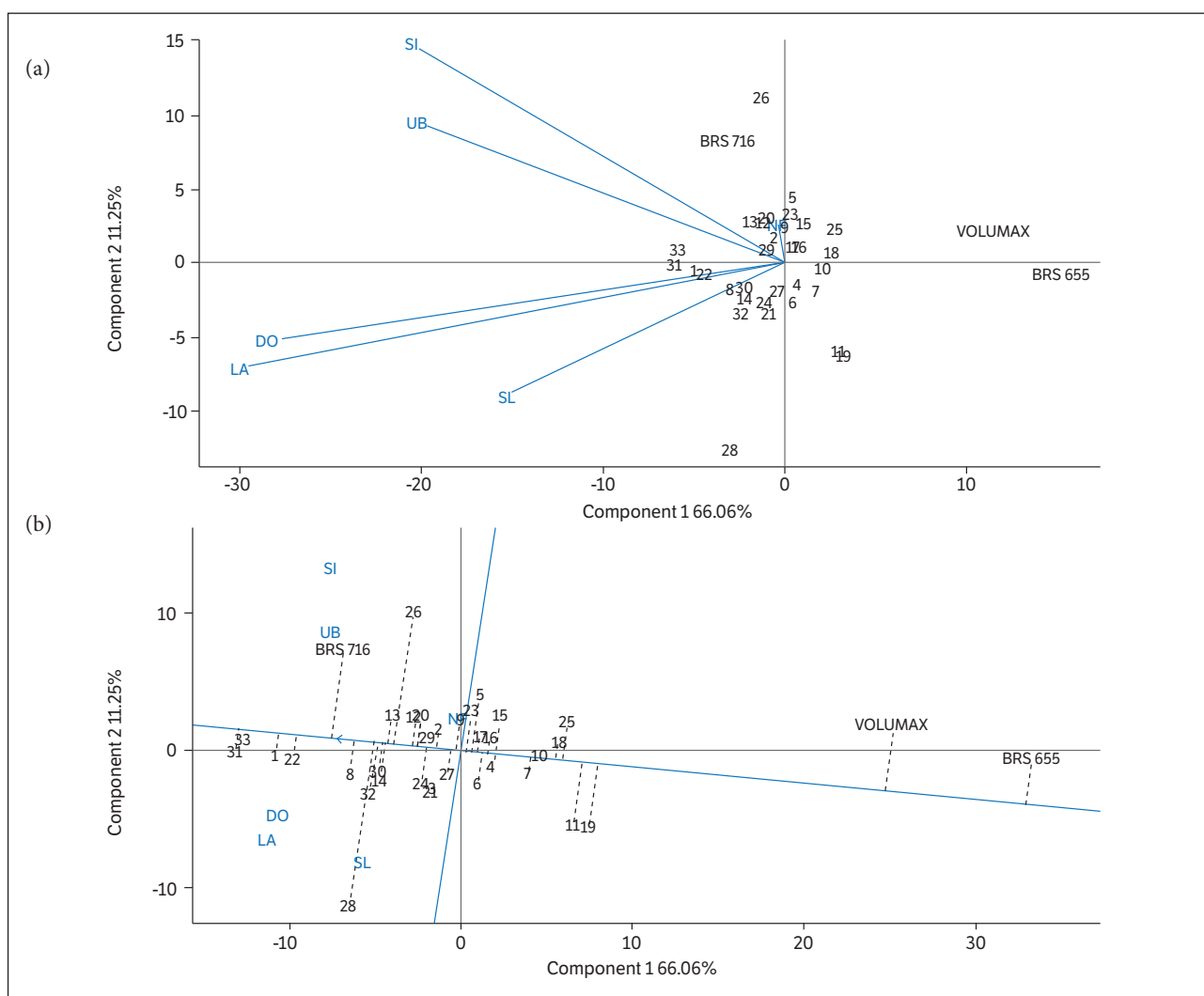
ID	FLOW		PH		MC		GMY		DMY	
	$I_i$	$I_{i(BRS716)}$	$I_i$	$I_{i(BRS716)}$	$I_i$	$I_{i(BRS716)}$	$I_i$	$I_{i(BRS716)}$	$I_i$	$I_{i(BRS716)}$
1	107.28	96.88	94.05	88.78	96.27	95.66	99.90	86.09	107.63	86.68
2	93.53	84.67	99.07	92.68	95.01	95.21	91.69	79.84	88.52	71.52
3	98.25	89.28	100.82	94.15	99.21	99.55	93.74	78.91	91.20	71.14
4	95.48	85.98	94.26	88.62	99.63	99.34	86.26	72.14	90.96	72.18
5	102.24	92.83	95.84	90.82	99.87	100.45	98.99	87.53	94.17	79.80
6	106.21	96.23	96.53	92.16	101.33	101.89	90.16	77.81	81.68	66.18
7	94.84	86.23	94.72	89.33	95.74	95.80	83.02	71.38	85.23	68.98
8	106.18	97.56	96.84	90.49	96.67	98.42	102.41	87.42	106.59	88.21
9	96.42	88.26	99.13	93.30	99.50	100.05	102.59	88.07	95.59	80.96
10	98.98	89.34	96.38	90.56	101.20	100.28	90.70	78.04	86.40	70.00
11	94.15	86.09	94.66	89.10	99.03	97.84	83.39	70.16	76.55	60.53
12	94.57	85.35	93.88	88.40	95.66	96.68	91.74	84.21	92.60	81.69
13	107.47	97.11	93.01	89.66	101.33	101.77	106.18	94.46	93.07	76.44
14	100.31	91.82	93.11	88.04	96.33	95.96	90.04	78.40	95.39	78.01
15	107.12	97.37	97.69	92.70	99.18	98.55	96.81	87.56	89.41	76.73
16	98.59	90.55	96.16	91.18	100.59	99.64	97.50	83.15	94.62	75.07
17	104.30	95.40	96.62	91.68	100.54	100.04	90.14	77.65	84.35	68.40
18	101.09	92.31	97.93	91.80	101.86	101.58	92.63	79.13	80.60	65.71
19	94.68	85.64	91.58	86.33	103.36	103.52	84.44	70.45	75.76	60.39
20	102.11	93.39	103.95	97.76	95.83	95.09	90.05	79.26	95.11	79.43
21	91.49	83.51	99.25	95.47	96.08	95.05	78.75	67.55	92.08	74.81
22	101.18	92.90	109.05	103.18	99.79	99.28	106.64	92.19	105.66	87.55
23	99.88	91.33	105.14	98.88	97.86	98.14	97.47	85.22	95.87	80.68
24	92.40	83.59	102.29	95.82	94.37	94.61	88.77	75.92	98.57	78.43
25	92.70	83.57	103.20	96.56	97.79	96.83	80.12	72.53	79.45	65.92
26	102.41	93.83	107.17	100.79	97.07	97.98	93.36	83.64	91.71	83.25
27	95.17	86.43	103.76	97.82	97.89	96.99	90.29	77.52	96.54	77.39
28	89.83	81.46	100.91	95.11	92.33	91.75	80.35	67.53	89.70	72.24
29	103.12	94.16	103.43	97.46	98.36	98.59	99.06	86.54	102.54	81.05
30	91.69	82.66	96.97	91.48	94.13	94.81	83.10	72.91	85.32	72.14
31	104.92	95.42	100.28	95.03	97.83	97.90	108.01	93.26	117.80	93.45
32	96.43	87.48	105.99	100.36	97.38	97.88	95.44	81.39	98.80	79.98
33	104.32	95.69	103.80	97.46	99.95	99.08	108.84	96.82	107.04	90.46
34	108.38	100.00	101.88	100.00	95.85	100.00	103.46	100.00	99.51	100.00
35	64.56	57.97	57.93	55.89	98.32	98.37	37.49	33.02	39.62	33.74
36	55.16	49.59	44.90	42.98	99.04	98.73	21.84	19.26	18.72	16.06

95%, ten and three experimental hybrids were highlighted FLOW and MC, respectively, where the genotypes 24, 28 and 30 were coincident (Table 6).

Another approach was to determine the reliability index of the experimental genotypes in relation to a commercial check widely adopted by farmers. For this, we used the hybrid 'BRS 716' (genotype 34), because it is a biomass sorghum cultivar, while the checks 'Volumax' and 'BRS 655' are forage sorghum cultivars. According to the analyses of adaptability and stability by the method of Annicchiarico, the experimental photoperiod-sensitive hybrids that presented the lowest risk of adoption in relation to 'BRS 716' ( $I_{i(BRS716)}$ ) were 22, 26 and 32 for PH and 24, 28 and 30 for MC. For FLOW, 25 of the 33 experimental hybrids associated lower risk of recommendation relative to 'BRS 716', while

for GMY and DMY all hybrids presented higher risk. (Table 6). Although high correlation values ( $\geq 0.92$ ) were observed between the reliability indexes, in relation to the average of the environment ( $I_i$ ) and the check ( $I_{i(BRS716)}$ ), it was detected a divergence in the classification of the genotypes (Table 6). This fact may be associated to differences in the 'BRS 716' response patterns and experimental hybrids in the tested environments for the different traits (Table 5).

According to Ferreira et al. (2006), the GEI is better described by multivariate approaches. There are some multivariate methods applied to investigate GEI, among them stands for the GGE biplot method, which allow to characterize the interrelationship among environments and genotypes (Yan and Tinker 2006). In Fig. 2 there were presented two biplots related to the trait DMY. The



**Figure 2.** Biplots showing the interrelationship among environments and genotypes (a) and mean versus stability of genotypes (b) for dry mass yield (ton/ha). Abbreviations in blue color represent the environments (NP-Nova Porteirinha, Dracena-DR, Uberlândia-UB, SL-Sete Lagoas, LA-Lavras, GO-Goiânia, DO-Dourados, SI-Sinop, PE-Pelotas, GU-Guaíra), and numbers in black color are genotypes (experimental hybrids).

emphasis on this trait is because it might be considered a natural selection index, once it takes into account the other traits. Furthermore, DMY is more closely related to energy cogeneration yield (Castro et al. 2015). The biplot A (Fig. 2) showed some environments were highly positive correlated, highlighting Uberlândia and Sinop, and Dourados, Lavras and Sete Lagoas. The environment Nova Porteirinha stands out for its low discrimination of genotypes as for DMY.

The biplot B (Fig. 2) highlights the performance and stability of the genotypes. The same experimental hybrids 31, 33, 1, 22 and 8 were pointed out in terms of adaptability as aforementioned and also associated high stability. However, GGE biplot does not inform appropriately on genotype response pattern across environments, what was done by Toler method. Additionally, the cultivar BRS 716 also had high mean, but less stable.

## CONCLUSION

The genotype by environment interaction is expressive in biomass sorghum, mainly for the traits related to the biomass yield, which was predominantly complex. The Toler, GGE biplot and Annicchiarico methods present complementarity for describing the differential relative response of genotypes across environments. The experimental photoperiod-sensitive hybrids 1, 8, 22, 31 and 33 are promising because associate stability and lower recommendation risk. Moreover, the hybrids 1 and 8 presented response pattern for broad adaptability, while the hybrids 22, 31 and 33 for specific adaptability to high quality environments.

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Conceptualization, Parrella, R. A. C.; Methodology, Nunes J. A. R.; Investigation, Delgado I. D.; Writing – Original Draft, Delgado I. D.; Writing – Review and Editing, Castro F. M. R., Gonçalves F. M. A., Nunes J. A. R., Parrella, R. A. C.; Supervision, Gonçalves F. M. A., Nunes J. A. R.

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