

Strategies for Multi-trait Selection of Sweet Sorghum Progenies

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Abstract: *The selection of sweet sorghum genotypes is based on multiple agronomical and technological traits. The objectives of this study were to evaluate the effectiveness of inter-trait recovery information in improving the selective accuracy of the predicted genetic values of sweet sorghum progenies, and to compare several selection indices in terms of selection gains using single- and multi-trait mixed models. The trials were conducted in two sites. The traits flowering time, plant height, green mass production, total soluble solids content, and tons of Brix per hectare were assessed. Significant genetic variance was observed for all traits, except for total soluble solids content. The multi-trait analysis provided more accurate estimates of genetic parameters and predictions of the progeny genetic values, and higher selection gain than the single-trait analysis. The direct selection for TBH and the FAI/BLUP index resulted in balanced genetic gains for the assessed traits.*

Keywords: *Sorghum bicolor, selection index, genetic correlation, BLUP, REML.*

INTRODUCTION


The global demand for renewable energy sources, such as biofuels, has increased (OECD/FAO 2021). The use of these renewable sources aims to reduce greenhouse gas emissions from the use of fossil fuels (Abdullah et al. 2019). Brazil is the second-largest producer of ethanol in the world using sugarcane as feedstock (Karp et al. 2021). However, to strengthen its ethanol production chain, especially regarding filling the window in the sugarcane off-season, which runs from December to April, the ethanol plants might use other bioenergy crops, which would maximize the use of resources and contribute to low-carbon agriculture and more efficient agricultural and industrial processes.

Sweet sorghum [*Sorghum bicolor* (L.) Moench] has emerged as a complementary bioenergy crop that has favorable aspects to its exploitation, such as high green mass yield, juicy stalks, and high levels of fermentable sugars in its juice (Regassa and Wortmann 2014, Appiah-Nkansah et al. 2019). It is a crop that has been attracting attention due to its great potential for ethanol production (Wu et al. 2010, Fernandes et al. 2014, Ahmad Dar et al. 2018).

Sweet sorghum breeding programs aim to obtain cultivars with higher ethanol yield. However, this quantitative trait has some peculiarities, such as complex genetic architecture and pronounced environmental effects (Burks et al. 2015, Rocha et al. 2018, Cooper et al. 2019). The first peculiarity is that ethanol yield results from the expression of several interrelated agro-industrial traits, such as

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plant height, green mass production, flowering time, juice yield, fermentable sugar content in the juice, and tons of Brix per hectare (TBH) (Leite et al. 2017). Another aspect is the difficulty or even the infeasibility of performing phenotyping for ethanol yield, especially in the early stages of the breeding cycle, in which many genotypes must be tested, and the experimental material might be limited (e.g., seeds). Regarding the difficulty of performing direct selection for ethanol yield, an alternative is to obtain genetic gain via indirect selection. The path analysis study by Lombardi et al. (2015) found that TBH is highly correlated with ethanol yield, in addition to having a strong, positive, direct effect on this target trait of sweet sorghum breeding. Thus, TBH is suitable for indirect selection for ethanol yield.

The decision about selection of promising sweet sorghum cultivars is based on multiple traits. This task can be accomplished using single- or multi-trait mixed models. However, the selection efficiency is expected to increase by using multi-trait mixed models (Henderson and Quaas 1976) because these models make it possible to recover information on the covariance between traits and thus might significantly improve the accuracy of genetic value predictions, making the selection process more efficient (Piepho et al. 2008, Viana et al. 2010, Alves et al. 2018).

Another option for selecting genotypes for ethanol yield is the use of selection indices. In this context, some breeders have commonly used selection indices based on single-trait mixed models to predict genetic values (Meier et al. 2019), but their main disadvantage is the impossibility of exploiting the covariance (correlation) between traits. Some indices frequently used are based on the sum of the predicted genetic values (additive index) (Resende 2007), and the sum of rank index (Mulamba and Mock 1978). More recently proposed selection indices, such as the factor analysis based on ideotype-design associated with the best linear unbiased prediction (FAI-BLUP) index, are also based on univariate mixed models (Rocha et al. 2017). Although the FAI-BLUP index make it possible to exploit the covariance between traits, the use of multi-trait mixed model might increase the expected genetic gain with selection.

Given the above, the objectives of this study were to evaluate the effectiveness of inter-trait recovery information in improving the selective accuracy of the predicted genetic values of sweet sorghum progenies, and to compare several selection indices in terms of selection gains using single- and multi-trait mixed models.

MATERIAL AND METHODS

Sites

The trials were conducted in the 2016/2017 agricultural crop year in two sites in the State of Minas Gerais, Brazil (Figure 1). The first was in the experimental area of Embrapa Maize and Sorghum (lat 19° 27' 57" S, long of 44° 14' 49" W, alt 767 m asl) in the municipality of Sete Lagoas, Minas Gerais, Brazil. The region has an average temperature of approximately 23 °C and mean annual rainfall of 1,403 mm. The climate, according to Köppen's climate classification, is Cwa. The soil is classified as *Latossolo vermelho* (Oxisol) with a gently undulating relief. The second experiment was conducted at the Center for Scientific and Technological Development in Agriculture, Muquém Farm (lat 21° 14' S, long 45° 00' W, alt 932 m asl), Federal University of Lavras, Lavras, Minas Gerais. The region has an average annual temperature of 19.4 °C and a mean annual rainfall of 1,529.7 mm. The climate, according to Köppen's climate classification, is Cwa. The soil is classified as *Latossolo vermelho-amarelo* (Oxisol) with a gently undulating relief. Precipitation and average temperature data were obtained during the trials for the two sites (INMET 2021).

Progeny evaluation

A total of 196 half-sib progenies from a cycle-0 population of recurrent selection were evaluated (Leite et al. 2019). The experiments were conducted in a 14×14 lattice design with two replications in Lavras and three replications in Sete Lagoas. The plot consisted of 3-m-long rows with 60-cm spacing between rows. Sowing was performed at the end of October 2016 in Sete Lagoas and at the beginning of November 2016 in Lavras. Harvesting was performed when the grains were in the hard dough stage, at approximately 115 days after sowing.

The following traits were measured: flowering time (FLOW, days) by the number of days from sowing up to when 50% of the plants of the plot flowered; plant height (PH, m), according to the mean height (m) of eight plants taken at random from each plot, measured from the soil surface to the tip of the panicle using a measuring tape; green mass production (GMP, t ha⁻¹) at the time of each cutting, according to the weight of the whole plants from each plot, weighed

on a digital hanging scale in kg, converted to t ha⁻¹; total soluble solids content (TSS, °Brix) through a portable digital refractometer (Instrutemp - ITREFD-45), with automatic correction of temperature and maximum resolution of 0.1 °Brix; and tons of Brix per hectare (TBH), obtained by the product of GMP and TSS/100.

Statistical analyses

The data analyses were performed using the mixed-model methodology, with estimation of the fixed effects via best linear unbiased estimates (BLUEs), and prediction of the random effects via best linear unbiased predictions (BLUPs) (Henderson 1974), and the use of the restricted maximum likelihood (REML) method for estimation of variance components (Patterson and Thompson 1971). Multi-environment single-trait (Equation 1) and multi-trait multi-environment (five traits) (Equation 2) analyses were performed as described below:

$$y_t = X_t \beta_t + Z_t b_t + W_t g_t + Q_t s_t + e_t \quad (1)$$

where y_t is the vector of phenotypic data of the trait t ; β_t is the vector of fixed effects of the sites and of the replications within sites plus the overall mean of trait t ; b_t is the vector of the sub-block effects within replications at the sites of trait t , $b_t \sim N(0, I \sigma_{b_t}^2)$; g_t is the vector of the progeny effects of trait t , $g_t \sim N(0, I \sigma_{g_t}^2)$; s_t is the vector of the progeny \times site interaction effects of trait t , $s_t \sim N(0, I \sigma_{s_t}^2)$; e_t is the vector of the errors of trait t , $e_t \sim N(0, I \sigma_{e_t}^2)$; X_t, Z_t, W_t, Q_t are the incidence matrices of fixed and random effects; $\sigma_{b_t}^2, \sigma_{g_t}^2, \sigma_{s_t}^2$, and $\sigma_{e_t}^2$ are the variances of sub-blocks within replications at each site, of progenies, of progeny \times site interaction, and of the experimental error, respectively.

$$y = X\beta + Zb + Wg + Qs + e \quad (2)$$

where y is the vector of stacked phenotypic data of the traits $y' = (y'_1, \dots, y'_5)$; β is the vector of fixed effects of the sites and of the replications within each site added to the overall mean; b is the vector of the sub-block effects within replications

at each site, $b \sim NMV(0, \Sigma_b)$; Σ_b is the covariance matrix of the sub-blocks, defined as $\Sigma_b = \begin{bmatrix} \sigma_{b_1}^2 & \dots & 0 \\ \vdots & \ddots & \vdots \\ 0 & \dots & \sigma_{b_5}^2 \end{bmatrix} = \otimes I_{28'}$

where $\sigma_{b_t}^2$ is the variance of sub-blocks for the trait t ($t=1, \dots, 5$); g is the vector of the progeny effects, $g \sim NMV(0, \Sigma_g)$; Σ_g

is the matrix of genetic covariances, defined as $\Sigma_g = \begin{bmatrix} \sigma_{g_1}^2 & \dots & \sigma_{g_{15}} \\ \vdots & \ddots & \vdots \\ \sigma_{g_{15}} & \dots & \sigma_{g_5}^2 \end{bmatrix} = \otimes I_{196'}$ where $\sigma_{g_t}^2$ is the variance of progenies

for the trait t ($t=1, \dots, 5$) and $\sigma_{g_{tt'}}$ is the genetic covariance of progenies between traits t and t' ; s is the vector of the progeny \times site interaction effects, $s \sim NMV(0, \Sigma_s)$; Σ_s is the covariance matrix of the progeny \times site interactions, defined

as $\Sigma_s = \begin{bmatrix} \sigma_{s_1}^2 & \dots & \sigma_{s_{15}} \\ \vdots & \ddots & \vdots \\ \sigma_{s_{15}} & \dots & \sigma_{s_5}^2 \end{bmatrix} = \otimes I_{392'}$, where $\sigma_{s_t}^2$ is the variance of progeny \times site interaction for the trait t ($t=1, \dots, 5$) and

$\sigma_{e_{tt'}}$ is the covariance of progeny \times site interaction between traits t and t' ; e is the error vector, where $e \sim NMV(0, \Sigma_e)$;

Σ_e is the matrix of error covariance, defined as $\Sigma_e = \begin{bmatrix} \sigma_{e_1}^2 & \dots & \sigma_{e_{15}} \\ \vdots & \ddots & \vdots \\ \sigma_{e_{15}} & \dots & \sigma_{e_5}^2 \end{bmatrix} = \otimes I_n$, where $\sigma_{e_t}^2$ is the error variance for the

trait t ($t=1, \dots, 5$) and $\sigma_{e_{tt'}}$ is the error covariance between traits t and t' ; and $X, Z, W,$ and Q are the design matrices that associate the fixed and random effects with the data vector y .

Wald test was used to test the significance of the fixed effects. The likelihood ratio test was used to verify the significance of the random effects (Mrode 2014, Resende et al. 2014). From the estimates of the variance components, the correlation of the progenies across the sites was estimated by the expression $r_B = \frac{\sigma_{g_t}^2}{\sigma_{g_t}^2 + \sigma_{s_t}^2}$ for each trait t , the genetic and environmental correlations between traits t and t' by the expressions $r_{gtt'} = \frac{\sigma_{g_{tt'}}}{\sqrt{\sigma_{g_t}^2 \times \sigma_{g_{t'}}^2}}$ and $r_{ett'} = \frac{\sigma_{e_{tt'}}}{\sqrt{\sigma_{e_t}^2 \times \sigma_{e_{t'}}^2}}$.

The significance of the genetic correlations was evaluated via Bootstrap at 5% probability level by the “bias-corrected

and accelerated" (BCa) method, with 1000 bootstrap replications, using the *wBoot* R package (Weiss 2016), while the residual correlations were checked by t-test at 5% probability level. The mean selective accuracy of progenies for each trait (t) was estimated by the expression $r_{gg} = \sqrt{1 - \frac{PEV}{\sigma_{g_t}^2}}$, where PEV is the mean prediction error variance of the BLUPs of the progenies (Resende and Duarte 2007).

For multi-trait selection, the following selection indices were adopted based on the BLUPs of the progenies from the multi-environment single- and multi-trait mixed models:

1 - Direct selection based on the TBH trait, where the indirect gains obtained for the other traits were estimated.

2 - Mulamba and Mock index (I_{MMj}), estimated from the rank of the progenies as $I_{MMj} = \sum_{t=1}^5 r_{tj}$, where I_{MMj} is the value of the sum of rank index associated with progeny j and r_{tj} is the rank of progeny j on trait t ;

3 - Additive index (I_A), estimated by the expression $I_{Aj} = \sum_{t=1}^5 \hat{g}_{tj} \times w_t \times \frac{1}{\sigma_{g_t}}$ as described by Resende (2007), in which I_{Aj} is the value of the additive index associated with progeny j ; \hat{g}_{tj} is the predicted genetic value of progeny j for trait t ; w_t is the economic weight associated with trait t ; and σ_{g_t} is the standard deviation of progenies for trait t . The economic weight was assumed equal to 1, because it is hard to define them for the assessed traits. Furthermore, single- and multi-trait BLUPs were already weighed by the heritability of the traits and by the covariances between traits. The last one is just valid for multi-trait analysis.

4 - The FAI-BLUP index combines factor analysis (exploratory factor analysis) with ideotypes (confirmatory factor analysis) to explore the covariance between the traits evaluated, as proposed by Rocha et al. (2017). Factor analysis was performed on the BLUPs of the progenies. The number of ideotypes (NI) was equal to $NI = 2^n$, where n is the number of factors with eigenvalues equal to or greater than 1. From this, the distances between the progenies evaluated and the ideotypes and the relative similarity measures were calculated, which enabled the ranking of the genotypes, determined

by the following equation: $P_{jk} = \frac{1}{d_{jk} \sum_{j,k=1}^{n,m} \frac{1}{d_{jk}}}$ where P_{jk} is the relative similarity of progeny j to ideotype k and d_{jk} is the distance from progeny j to ideotype k in standardized mean Euclidean distance.

The goal of selection was to decrease FLOW and increase PH, GMP, TSS, and TBH. A selection intensity of 10% was considered and the selection gain was only investigated for global selection, that is, it was performed based on average performance across sites. The expected selection gain ($GS_{\%}$) for each strategy was estimated based on the BLUPs of the 20 best progenies by the expression $GS_{\%} = \frac{BLUP_t}{\bar{Y}_t} \times 100$, in which $BLUP$ is the mean BLUP of the progenies selected for trait t and \bar{Y}_t is the overall mean of trait t . The analyses of coincidence among the selection indices using single- and multi-trait mixed models were performed by the agreement index proposed by Cohen (1960).

The statistical analyses using the mixed-model methodology were performed in R environment (R Core Team 2018). Single-trait analysis was performed using the *lme4* package using the penalized least squares algorithm (Bates et al. 2015), and multi-trait analysis using the *sommer* package version 3.7 using the Newton-Raphson algorithm (Covarrubias-Pazarán 2016).

RESULTS AND DISCUSSION

The genetic variance among half-sib progenies was non-null for most traits in the single- and multi-trait analyses (Table 1), except for TSS. It is noteworthy that this genetic variance was exclusively related to the additive genetic effects because they were evaluated in half-sib progenies, so they exploit $\frac{1}{4}$ of the additive variance of the population. The mean selective accuracies of progenies were high for most of the traits ($r_{gg} > 0.7$), except for TSS (Table 1), which indicates high reliability to select promising progenies based on experimental data (Resende and Duarte 2007). The values ranged from 0.77 for PH to 0.95 for FLOW and TBH (Table 1). The absence of genetic variance for TSS might have occurred due to environmental fluctuations, especially rainfall during the harvest season (Figure 1), which may have led to the dilution of soluble solids of the stem juice. Unlike what was observed in this study, França et al. (2016) observed high heritability estimates for TSS.

Table 1. Estimates of genetic variance among half-sib progenies of sweet sorghum (σ_g^2), variance of the progeny \times site interaction (σ_s^2), error variance (σ_e^2), mean selective accuracy of progenies ($r_{\hat{g}}$), and correlation of progenies across sites (r_g) for the agro-industrial traits evaluated in sweet sorghum progenies using single- (ST) and multi-trait (MT) BLUP

| Parameter | FLOW | | PH | | GMP | | TSS | | TBH | |
|---------------|-------|-------|--------|--------|-------|-------|------|------|------|------|
| | ST | MT | ST | MT | ST | MT | ST | MT | ST | MT |
| σ_g^2 | 17.8* | 18.0* | 0.022* | 0.023* | 88.1* | 89.8* | 0.3 | 0.3 | 2.1* | 2.2* |
| σ_s^2 | 0.5 | 0.4 | 0.0 | 0.0 | 36.9* | 39.6* | 0.6* | 0.7* | 1.4* | 1.6* |
| σ_e^2 | 8.9 | 8.9 | 0.1 | 0.1 | 98.1 | 95.3 | 2.8 | 2.7 | 2.9 | 2.8 |
| $r_{\hat{g}}$ | 0.94 | 0.95 | 0.77 | 0.87 | 0.83 | 0.92 | - | - | 0.79 | 0.95 |
| r_B | 0.97 | 0.97 | 1 | 1 | 0.70 | 0.69 | 0.30 | 0.27 | 0.61 | 0.79 |

FLOW - flowering time (days); PH - plant height (m); GMP - green mass production ($t\ ha^{-1}$); TSS - total soluble solids content ($^{\circ}$ Brix); TBH - tons of Brix per hectare ($t\ ha^{-1}$).
* Significant at 5% probability level, respectively, by the likelihood ratio test

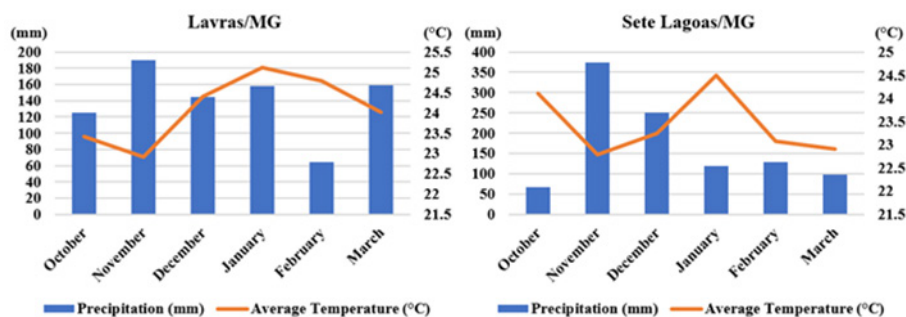


Figure 1. Precipitation and average temperature during October/2016 and March/2017 in Lavras/MG (A) e Sete Lagoas/MG (B), INMET (2021).

The variance of the progeny \times site interaction was significant for GMP, TSS, and TBH (Table 1). This indicates that the progenies showed a relatively noncoincident performance in the two sites for these traits. Studies conducted on sweet sorghum have shown the presence of genotype \times environment interactions for several traits correlated with ethanol production (Souza et al. 2013, Lombardi et al. 2018, Udoh et al. 2018). According to Murray et al. (2008) and Gutjahr et al. (2013), the TSS trait has a somewhat complex inheritance and therefore is greatly influenced by the environment. Several factors have an impact on the final TSS, including day length and radiation intensity, in addition to soil conditions, soil fertility, and the response to fertilization (Kumar et al. 2008).

In general, the multi-trait analysis provided higher estimates for several parameters (Table 1). Multi-trait analysis yielded higher estimates of the genetic variances among progenies for all traits except TSS, as well as higher mean selective accuracy estimates. According to Piepho et al. (2008) and Viana et al. (2010), multi-trait analysis should be preferred, especially when the evaluated traits are highly correlated. The selection for low heritability trait can also benefit when performed with a high heritability trait (Souza et al. 2019). Moreover, according to Schaeffer (1984), in situations where the traits have equal heritability, the selection efficiency of the multi-trait BLUP relative to the single-trait BLUP depends only on the absolute difference between the genetic and environmental correlations of the assessed traits.

Genetic correlations among the traits are of great importance for success in selections to be conducted in breeding programs. Positive correlations show that the changes of two traits are in the same direction, while negative correlations indicate their inverse relationships. High-magnitude and positive genetic correlation was observed between TBH and almost all traits (Table 2). Working with sweet sorghum progenies, França et al. (2016) also observed a high genetic correlation between the traits GMP and TBH (0.80). In a study of phenotypic correlations using path analysis, Lombardi et al. (2015) demonstrated that TBH showed a high positive correlation and a direct effect on ethanol yield. A high residual correlation was observed between GMP and TBH (0.82), which indicates that the environment affected these traits equally and in the same direction.

Table 2. Estimates of the genetic (above the diagonal) and residual (below the diagonal) correlations by multi-trait analysis for the agro-industrial traits: flowering time (FLOW, days); plant height (PH, m); green mass production (GMP, t ha⁻¹), and tons of Brix per hectare (TBH, t ha⁻¹) evaluated in Lavras and Sete Lagoas in the 2016/2017 agricultural crop year

| Correlation | FLOW | PH | GMP | TBH |
|-------------|--------|-------|-------|-------|
| FLOW | - | 0.78* | 0.83* | 0.90* |
| PH | 0.02 | - | 0.82* | 0.82* |
| GMP | -0.17* | 0.24* | - | 0.99* |
| TBH | -0.11* | 0.14* | 0.82* | - |

* Significant at 5% probability level, respectively, by the Bootstrap BCa method based on 9999 simulations for the genetic correlations, and by the t-test for the residual correlations.

In a breeding program, an issue that is always important is the multi-trait selection strategy to be adopted, since the ideotype involves a series of traits of interest. In the sweet sorghum crop, the ideotype consists of a minimum biomass yield of 60 t ha⁻¹, minimum total sugar extraction of 80 kg t⁻¹ of biomass, minimum total sugar content in the juice of 12.5%, minimum ethanol production of 60 L t⁻¹ of biomass, and minimum industrial use period of 30 days (Parrella 2011). By the single-trait mixed model, the mean estimated gains from selection were 6.2% (FLOW), 4.02% (PH), 23.3% (GMP), and 29.4% (TBH) (Figure 2). Direct selection for the TBH trait and the FAI/BLUP index were promising because they provided balanced gains for GMP and TBH, and the agreement of selected progenies was 68.75% (Table 3). The sum of rank and additive indices was not so efficient in the single-trait approach because the estimated gains were below the mean gain for GMP and TBH.

In the multi-trait approach, the expected mean gains were 10.6% (FLOW), 7.1% (PH), 29.3% (GMP) and 37.1% (TBH) (Figure 2). There was an increase in estimated gains from selection for all traits, and all strategies provided more balanced gains compared to the single-trait approach. The coincidences between the selection indices using multi-trait analysis were higher than those found with single-trait analysis (Table 3). The direct selection for TBH trait and the additive index selected the same progenies (Table 3). These two indices provided the highest gain estimate for the TBH trait (37.5%). The FAI/BLUP index provided very balanced estimates of gains from selection, which was above the mean gain obtained in the single-trait approach.

When selecting for a given trait, this will usually lead to changes in others due to genetic correlations (Ramalho et al. 2012). This fact is called the correlated response to selection, and its direction may or may not be of interest to the

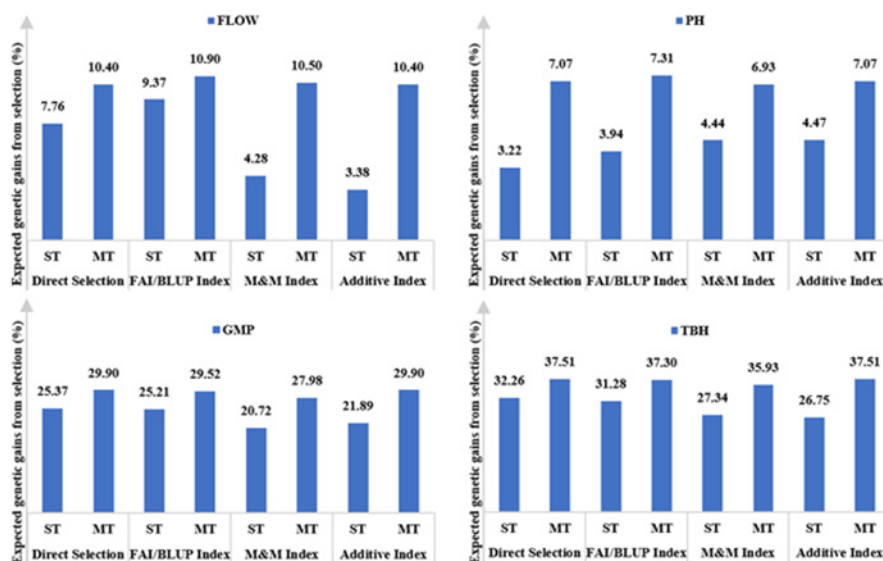


Figure 2. Expected genetic gains from selection (%) of the agro-industrial traits evaluated using different selection indices (direct selection for TBH trait, FAI/BLUP index, Sum of rank index (M&M index), and additive index) via single-trait (ST) and multi-trait (MT) analyses. PH - plant height (m); FLOW – flowering time (days); GMP - green mass production (t ha⁻¹); and TBH - tons of Brix per hectare (t ha⁻¹).

Table 3. Number of common progenies selected (between parenthesis) and agreement coefficients of top-20 best progenies between different selection indices (direct selection of TBH trait, FAI/BLUP index, Sum of rank index (M&M index), and additive index) using single- (bellow the diagonal) and multi-trait (above the diagonal) analyses

| | Direct Selection | FAI/BLUP Index | M&M Index | Additive Index |
|------------------|------------------|----------------|-------------|----------------|
| Direct Selection | - | (18) 87.50% | (18) 87.50% | (20) 100.00% |
| FAI/BLUP Index | (15) 68.75% | - | (18) 87.50% | (17) 81.25% |
| M&M Index | (12) 50.00% | (11) 43.75% | - | (18) 87.50% |
| Additive Index | (12) 50.00% | (9) 31.25% | (17) 75.00% | - |

breeder (Leite et al. 2019). In this study, direct selection for TBH trait provided satisfactory joint results in indirect gains for three of the five traits evaluated (PH, GMP, and TBH) (Figure 2). According to Hallauer et al. (2010), a high genetic correlation between traits, coupled with high heritability of secondary traits, tends to provide satisfactory genetic gains for all of them. In situations like this, the selection response is fast and very efficient, but even in the same scenario the relative selection gain is expected to be lower in highly improved populations (Hallauer et al. 2010).

The sum of rank index was not promising because it showed the lowest estimated gain from selection for the trait GMP in the single-trait approach and PH, GMP, and TBH in the multi-trait analysis (Figure 2). Inefficiency in obtaining satisfactory gains in the sweet sorghum crop by the Mulamba and Mock index was also observed by França et al. (2016) through the univariate mixed-model approach, since this index provided the lowest estimates of gains from selection for all traits measured by the authors, some of which were the same as those evaluated here (GMP and TBH).

The use of the additive index combined with the single-trait approach was not promising because it provided the lowest estimated gain in TBH among all strategies used (Figure 2). This would be a problem because TBH is directly related to ethanol production (Lombardi et al. 2015). However, its use combined with the multi-trait approach proved to be very efficient (Figure 2). This increase of selection efficiency using the multi-trait approach is due to effectiveness of inter-trait recovery information, in which the genetic covariance between traits is taken into account to obtain the predictions of genetic values of the progenies.

In corn, Mendonça et al. (2017) sought to select genotypes that combined the traits tolerance to nitrogen stress and efficient use of nitrogen. The researchers took single- and multi-trait approaches with several selection indices, including the additive index and the sum of rank index. In their multi-trait analysis, there was an increase in the estimates of gains from selection. Working with sweet corn, Entringer et al. (2016) observed that the use of the additive index combined with the multi-trait approach provided higher gain estimates and was more efficient in selecting progenies than the sum of rank index. In soybean crop, the additive and FAI/BLUP indices were efficient in selecting productive progenies associated with upright architecture (Volpato et al. 2021).

By the FAI/BLUP index, under single- and multi-trait approaches, all traits were explained by only one factor, which might be associated with the high correlations (Rocha et al. 2017). In the literature, the FAI/BLUP index has been used to select superior progenies of elephant grass (Rocha et al. 2017), common bean (Rocha et al. 2019), soybean (Woyann et al. 2019, Volpato et al. 2021), and biomass sorghum (Silva et al. 2018). Our use of the FAI/BLUP index with multi-trait BLUPs proved to be an interesting alternative, since it provided balanced gains for almost all traits, and its estimate for TBH was very close to that obtained with direct selection and by the additive index (Figure 2). Its estimate for the gain in FLOW was higher than that from any other strategy. These results indicate the efficiency of the FAI/BLUP index in providing desirable gains for a set of traits that strongly impact ethanol production but at the same time hinder the selection of earlier progenies. The FAI/BLUP original proposal (Rocha et al. 2017) uses single-trait BLUP, and it was observed that the use of FAI/BLUP based on multi-trait BLUP means provided greater increases in the estimates of gains from selection.

Multi-trait analysis provided more accurate estimates of genetic parameters and predictions of genetic values than single-trait analysis. The direct selection for TBH and the FAI/BLUP index resulted in the estimate of balanced genetic gains, both in the single-trait and in the multi-trait approaches, enabling the identification of progenies that were associated with high performance.

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REFERENCES

- Abdullah B, Syed Muhammad SAF ad, Shokravi Z, Ismail S, Kassim KA, Mahmood AN and Aziz MMA (2019) Fourth generation biofuel: A review on risks and mitigation strategies. **Renewable and Sustainable Energy Reviews** **107**: 37-50.
- Ahmad Dar R, Ahmad Dar E, Kaur A and Gupta Phutela U (2018) Sweet sorghum-a promising alternative feedstock for biofuel production. **Renewable and Sustainable Energy Reviews** **82**: 4070-4090.
- Alves RS, Carvalho Rocha JRAS, Teodoro PE, Resende MDV, Henriques EP, Silva LA, Carneiro PCS and Bhering LL (2018) Multiple-trait BLUP: a suitable strategy for genetic selection of Eucalyptus. **Tree Genetics & Genomes** **14**: 77.
- Appiah-Nkansah NB, Li J, Rooney W and Wang D (2019) A review of sweet sorghum as a viable renewable bioenergy crop and its techno-economic analysis. **Renewable Energy** **143**: 1121-1132.
- Bates D, Maechler M, Bolker B and Walker S (2015) Fitting linear mixed-effects models using lme4. **Journal of Statistical Software** **67**: 1-48.
- Burks PS, Kaiser CM, Hawkins EM and Brown PJ (2015) Genomewide association for sugar yield in sweet sorghum. **Crop Science** **55**: 2138-2148.
- Cohen J (1960) A coefficient of agreement for nominal scales. **Educational Psychological Measurement** **20**: 37-46.
- Cooper EA, Brenton ZW, Flinn BS, Jenkins J, Shu S, Flowers D, Luo F, Wang Y, Xia P, Barry K, Daum C, Lipzen A, Yoshinaga Y, Schmutz J, Saski C, Vermerris W and Kresovich S (2019) A new reference genome for Sorghum bicolor reveals high levels of sequence similarity between sweet and grain genotypes: Implications for the genetics of sugar metabolism. **BMC Genomics** **20**: 1-13.
- Covarrubias-Pazaran G (2016) Genome-Assisted prediction of quantitative traits using the R package sommer. **PLoS ONE** **11**: 1-15.
- Entringer GC, Vettorazzi JCF, Santos EA, Pereira MG and Viana AP (2016) Genetic gain estimates and selection of S1 progenies based on selection indices and REML/BLUP in super sweet corn. **Australian Journal of Crop Science** **10**: 411-417.
- Fernandes G, Braga TG, Fischer J, Parrella RAC, Resende MM and Cardoso VL (2014) Evaluation of potential ethanol production and nutrients for four varieties of sweet sorghum during maturation. **Renewable Energy** **71**: 518-524.
- França AED, Parrella RAC, Souza VF, Bastos GQ, Nunes JAR and Schaffert RE (2016) Seleção simultânea em progênies de sorgo-sacarino por meio de índices de seleção. **Pesquisa Agropecuária Brasileira** **51**: 1737-1743.
- Gutjahr S, Clément-Vidal A, Soutiras A, Sonderegger N, Braconnier S, Dingkuhn M and Luquet D (2013) Grain, sugar and biomass accumulation in photoperiod-sensitive sorghums. II. Biochemical processes at internode level and interaction with phenology. **Functional Plant Biology** **40**: 355-368.
- Hallauer AR, Carena MJ and Filho JBM (2010) **Quantitative genetics in maize breeding**. 6th edn, Springer Science, New York, 680p.
- Henderson CR (1974) General flexibility of linear model techniques for sire evaluation. **Journal of Dairy Science** **57**: 963-972.
- Henderson CR and Quaas RL (1976) Multiple trait evaluation using relatives' records. **Journal of Animal Science** **43**: 1188-1197.
- INMET - Instituto Nacional de Meteorologia (2021) Available at: <http://www.inmet.gov.br/portal/index.php?r=estacoes/estacoesAutomaticas>.
- Karp SG, Medina JDC, Letti LAJ, Woiciechowski AL, Carvalho JC, Schmitt CC, Oliveira Penha R, Kumlehn GS and Soccol CR (2021) Bioeconomy and biofuels: the case of sugarcane ethanol in Brazil. **Biofuels, Bioproducts and Biorefining** **15**: 899-912.
- Kumar SR, Shrotria PK and Deshmukh JP (2008) Characterizing nutrient management effect on yield of sweet sorghum genotypes. **World Journal of Agricultural Sciences** **4**: 787-789.
- Leite PSS, Botelho TT, Ribeiro PCO, Schaffert RE, Parrella RAC and Nunes JAR (2019) Intrapopulation recurrent selection in sweet sorghum for improving sugar yield. **Industrial Crops and Products** **143**: 1-8.
- Leite PSS, Fagundes TG, Nunes JAR, Parrella RAC, Durães NNL and Bruzi AT (2017) Association among agro-industrial traits and simultaneous selection in sweet sorghum. **Genetics and Molecular Research** **16**: 1-10.
- Lombardi GMR, Navegantes PCA, Pereira CH, Fonseca JMO, Parrella RAC, Castro FMR, Rocha MJ, Ornelas DO, Bruzi AT and Nunes JAR (2018) Heterosis in sweet sorghum. **Pesquisa Agropecuária Brasileira** **53**: 593-601.
- Lombardi GMR, Nunes JAR, Parrella RAC, Teixeira DHL, Bruzi AT, Durães NNL and Fagundes TG (2015) Path analysis of agro-industrial traits in sweet sorghum. **Genetics and Molecular Research** **14**: 16392-16402.
- Meier C, Meira D, Marchioro VS, Olivoto T, Klein LA and Souza VQ (2019) Selection gain and interrelations between agronomic traits in wheat F5 genotypes. **Revista Ceres** **66**: 271-278.
- Mendonça LF, Granato ÍSC, Alves FC, Morais PPP, Vidotti MS and Fritsche-

Strategies for Multi-trait Selection of Sweet Sorghum Progenies

- Neto R (2017) Accuracy and simultaneous selection gains for N-stress tolerance and N-use efficiency in maize tropical lines. **Scientia Agricola** **74**: 481-488.
- Mrode RA (2014) Linear models for the prediction of animal breeding values. 3rd edn, CAB International, Wallingford, 360p.
- Mulamba NN and Mock JJ (1978) Improvement of yield potential of the Eto Blanco maize (*Zea mays* L.) population by breeding for plant traits. **Egyptian Journal of Genetics and Cytology** **7**: 40-51.
- Murray SC, Rooney WL, Mitchell SE, Sharma A, Klein PE, Mullet JE and Kresovich S (2008) Genetic improvement of sorghum as a biofuel feedstock: II. QTL for stem and leaf structural carbohydrates. **Crop Science** **48**: 2180.
- OECD/FAO (2021) **OECD-FAO agricultural outlook 2021-2030**. OECD, Paris, 337p.
- Parrella RAC (2011) Melhoramento genético do sorgo sacarino. **Agroenergia em Revista Edição** **3**: 8-9.
- Patterson HD and Thompson R (1971) Recovery of inter-block information when block sizes are unequal. **Biometrika** **58**: 545.
- Piepho HP, Möhring J, Melchinger AE and Büchse A (2008) BLUP for phenotypic selection in plant breeding and variety testing. **Euphytica** **161**: 209-228.
- R Core Team (2018) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.
- Ramalho MAP, Abreu AFB, Santos JB and Nunes JAR (2012) **Aplicações da genética quantitativa no melhoramento de plantas autógamas**. Editora UFLA, Lavras, 522p.
- Regassa TH and Wortmann CS (2014) Sweet sorghum as a bioenergy crop: Literature review. **Biomass and Bioenergy** **64**: 348-355.
- Resende MDV (2007) **Matemática e estatística na análise de experimentos e no melhoramento genético**. Embrapa Florestas, Colombo, 561p.
- Resende MDV and Duarte JB (2007) Precisão e controle de qualidade em experimentos de avaliação de cultivares. **Pesquisa Agropecuária Tropical** **37**: 182-194.
- Resende MDV, Silva FF and Azevedo CF (2014) **Estatística matemática, biométrica e computacional: Modelos mistos, multivariados, categóricos e generalizados (REML/BLUP), inferência bayesiana, regressão aleatória, seleção genômica, QTL-GWAS, estatística espacial e temporal, competição, sobrevivência**. Suprema, Visconde do Rio Branco, 881p.
- Rocha JRASC, Machado JC and Carneiro PCS (2017) Multitrait index based on factor analysis and ideotype-design: proposal and application on elephant grass breeding for bioenergy. **GCB Bioenergy** **10**: 52-60.
- Rocha JRASC, Nunes KV, Carneiro ALN, Marçal TS, Salvador FV, Carneiro PCS and Carneiro JES (2019) Selection of superior inbred progenies toward the common bean ideotype. **Agronomy Journal** **111**: 1181-1189.
- Rocha MJ, Nunes JAR, Parrella RADC, Leite PSS, Lombardi GMR, Costa MLM, Schaffert RE and Bruzi AT (2018) General and specific combining ability in sweet sorghum. **Crop Breeding and Applied Biotechnology** **18**: 365-372.
- Schaeffer LR (1984) Sire and cow evaluation under multiple trait models. **Journal of Dairy Science** **67**: 1567-1580.
- Silva MJ, Carneiro PCS, Carneiro JES, Damasceno CMB, Parrella NNLD, Pastina MM, Simeone MLF, Schaffert RE and Parrella RAC (2018) Evaluation of the potential of lines and hybrids of biomass sorghum. **Industrial Crops and Products** **125**: 379-385.
- Souza NO, Alves RS, Teodoro PE, Silva LA, Tardin FD, Tardin AB, Resende MDV and Bhering LL (2019) Single and multiple-trait blup in genetic selection of parents and hybrids of grain sorghum. **Revista de la Facultad de Ciencias Agrarias** **51**: 1-12.
- Souza VF, Parrella RAC, Tardin FD, Costa MR, Carvalho Junior GA and Schaffert RE (2013) Adaptability and stability of sweet sorghum cultivars. **Crop Breeding and Applied Biotechnology** **13**: 144-151.
- Udoh D-A, Rasmussen SK, Jacobsen S-E, Iwo GA and Milliano W (2018) Yield stability of sweet sorghum genotypes for bioenergy production under contrasting temperate and tropical environments. **Journal of Agricultural Science** **10**: 42.
- Viana JMS, Sobreira FM, Resende MDV and Faria VR (2010) Multi-trait BLUP in half-sib selection of annual crops. **Plant Breeding** **129**: 599-604.
- Volpato L, Rocha JRASC, Alves RS, Ludke WH, Borém A and Silva FL (2021) Inference of population effect and progeny selection via a multi-trait index in soybean breeding. **Acta Scientiarum - Agronomy** **43**: 1-10.
- Weiss NA (2016) **wBoot: Bootstrap methods**. R package version 1.0.3. <https://CRAN.R-project.org/package=wBoot>.
- Woyann LG, Meira D, Zdziarski AD, Matei G, Milioli AS, Rosa AC, Madella LA and Benin G (2019) Multiple-trait selection of soybean for biodiesel production in Brazil. **Industrial Crops and Products** **140**: 111721.
- Wu X, Staggenborg S, Propheter JL, Rooney WL, Yu J and Wang D (2010) Features of sweet sorghum juice and their performance in ethanol fermentation. **Industrial Crops and Products** **31**: 164-170.