

Multivariate diallel analysis allows multiple gains in segregating populations for agronomic traits in *Jatropha*

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ABSTRACT. *Jatropha* is research target worldwide aimed at large-scale oil production for biodiesel and bio-kerosene. Its production potential is among 1200 and 1500 kg/ha of oil after the 4th year. This study aimed to estimate combining ability of *Jatropha* genotypes by multivariate diallel analysis to select parents and crosses that allow gains in important agronomic traits. We performed crosses in diallel complete genetic design (3 x 3) arranged in blocks with five replications and three plants per plot. The following traits were evaluated: plant height, stem diameter, canopy projection between rows, canopy projection on the line, number of branches, mass of hundred grains, and grain yield. Data were submitted to univariate and multivariate diallel analysis. Genotypes 107 and 190 can be used in crosses for establishing a base population of *Jatropha*, since it has favorable alleles for increasing the mass of hundred grains and grain yield and reducing the plant height. The cross 190 x 107 is the

most promising to perform the selection of superior genotypes for the simultaneous breeding of these traits.

Key words: *Jatropha curcas* L.; Multivariate statistics; Parental selection; Hybrid selection; Selection index; Genetic breeding

INTRODUCTION

Jatropha curcas L. is an oleaginous species with great potential for biofuel production (Laviola et al., 2013) because it does not compete against annual crops used as a source of human and animal food. In Brazil, this species is in domestication phase (Bahadur et al., 2013; Carels et al., 2013), being necessary more studies on genetic breeding to increase the productive potential of the crop, given that currently there is no commercial cultivar in Brazil (Teodoro et al., 2016a).

Thus, to install a genetic breeding program of *Jatropha* in Brazil numerous studies have been conducted in recent years, such as studies on genetic diversity (Rosado et al., 2010; Bhering et al., 2015), estimates of genetic parameters (Laviola et al., 2012; Teodoro et al., 2016d) and gains with selection (Bhering et al., 2013; Peixoto et al., 2016), repeatability (Laviola et al., 2013, Teodoro et al., 2016b), and path analysis (Teodoro et al., 2016c). However, for the formation of base populations for practicing the selection, diallel is the main genetic design used (Cruz et al., 2012).

In the literature, there are different methods for diallel analysis, among which Griffing (1956) is widely used because it allows estimating useful parameters in selecting parents and promising hybrid combinations. This method allows estimating the effects of general combining ability (GCA), which is dependent on the concentration of predominantly additive genes; specific combining ability (SCA), which is a result of the concentration of non-additive effect genes, reciprocal effects related to maternal inheritance, and extrachromosomal effects.

However, the traits measured are individually analyzed by univariate diallel analysis, which causes difficulties in the parent recommendation and superior hybrid combination, since most breeding programs aimed to obtain gain in multiple traits simultaneously. An alternative is the use of multivariate diallelic analysis, which allows analyzing multiple traits, facilitating the selection and discrimination of the most promising populations.

Therefore, this research was conducted in order to estimate the GCA and SCA using multivariate diallel analysis to recommend the most promising parents and crosses to reduce size and increase yield in *Jatropha*.

MATERIAL AND METHODS

Experimental area

Trial was conducted at the experimental area of Embrapa Cerrado in 2010-2015, in the municipality of Planaltina, Federal District. The municipality is located at 15°35'30"S, 47°42'30"W, at 1007 m of altitude. Climate is tropical with dry winter and rainy summer (Aw), according to Köppen's classification, with average annual temperature of 21°C, relative humidity of 68% and average rainfall of 1100 mm per year. Management practices were based on Dias et al. (2007), with adaptations according to the search results with *Jatropha* in Brazil and in the world (Durães et al., 2012; Bahadur et al., 2013; Carels et al., 2013; Resende et al., 2013).

Experimental design and traits evaluated

Crosses were performed in complete diallel genetic design, with three self-pollination and six hybridizations. Experimental design was randomized blocks, with five replications, three plants per plot and spacing 4 x 2 m. For performing the crosses, we used three genotypes selected in the germplasm bank with the following features: short size (CNPAE-107), high grain yield (CNPAE-190), and resistance to powdery mildew (CNPAE-259).

The traits evaluated were: plant height (PH, m), stem diameter (SD, mm), canopy projection between rows (CPR, m), canopy projection on the line (CPL, m), number of branches (NB), mass of hundred grains (MHG), and grain yield (YIE, g/planta). The traits PH, CPR, CPL, and NB were assessed in the third year; SD was assessed in the first year; MHG was quantified in the fourth year; YIE was assessed in the fifth year.

Statistical analysis

Initially, univariate diallel analysis was performed for each trait according to model 1 proposed by Griffing (1956). Multivariate diallel analysis was carried out according to the procedures performed by Ledo et al. (2003). Equality hypothesis of the genetic effects of multivariate diallel analysis was assessed with the Wilks test (Harris, 1975). Subsequently, we performed principal component analysis and selection index established by Pešek and Baker (1969), with the genotypic aggregate obtained according to the methodology described by Cruz et al. (2012). Pearson correlation was estimated between the first two principal components and the mean agronomic traits. Finally, the effects of GCA and SCA for the scores from each principal component and genotypic aggregate were estimated. All statistical analyzes were performed using Genes software (Cruz, 2013).

RESULTS AND DISCUSSION

Univariate diallel analysis showed a significant effect for the effect of genotypes (Table 1), demonstrating ability to select superior genotypes in the segregating populations with advances in generations. We found that the effects of dominance (Φ_{sc}) were higher in the genetic control for all traits, which indicates the existence of differences in progenitor's genic composition. According to Cruz et al. (2012), when larger estimates for Φ_{sc} are obtained in relation to others, the parents are different for most of the loci in dominance and heterotic manifestations exhibited by these crosses can be credited to genetic complementation between the loci that control the traits evaluated. These results were expected because Rosado et al. (2010) observed genetic diversity at molecular level among the parents used in this diallel, which were divided into different groups.

Despite the effect of dominance have greater importance on the studied features, it is important to mention the significance of the effects of GCA and reciprocal for all traits except GCA for the CPL. These results indicate that in addition to the effects of dominance, additive genetic effects and maternal and/or extrachromosomal effects should be considered in breeding program aimed at improving traits assessed in this study. Similar results were reported by Biabani et al. (2012) and Santana et al. (2013), who by using diallel crosses in *Jatropha* observed a significant effect on GCA and SCA.

For easier selection of superior parents and promising crosses based on traits of

interest, we performed diallel multivariate analysis (Table 2) by extension of the model 1 established by Griffing (1956). This analysis was significant by the Wilk test, indicating that the parents and the hybrid combinations have complementary alleles and the traits can be considered together in diallel analysis (Ledo et al., 2003; Silva et al., 2008; Benin et al., 2009).

Table 1. Univariate diallel analysis and quadratic components (Φ) for the traits plant height (PH), stem diameter (SD), canopy projection between rows (CPR), canopy projection on the line (CPL), number of branches (NB), mass of hundred grains (MHG), and grain yield (YIE) evaluated in nine *Jatropha* genotypes.

SV	d.f.	PH	SD	CPR	CPL	NB	MHG	YIE
Genotypes	8	0.55*	147.11*	0.70*	0.637*	176.32*	462.84*	1549525.65*
GCA	2	0.40*	373.17*	0.04*	0.003 ^{ns}	117.20*	756.98*	2193597.26*
SCA	3	0.94*	90.03*	1.23*	0.799*	210.57*	546.34*	2154437.05*
Reciprocal	3	0.25*	53.49*	0.61*	0.898*	181.49*	183.26*	515233.17*
Error	32	0.02	9.39	0.03	0.039	19.03	16.73	88121.92
Φ_{ge}	-	0.01	12.13	0.00	0.0	3.27	24.67	70182.51
Φ_{sc}	-	0.19	16.13	0.24	0.15	38.31	105.92	413263.02
Φ_{rc}	-	0.02	4.41	0.06	0.09	16.25	16.65	42711.12

*Significant at 5% probability by the F-test; ns: non-significant, SV: source of variation; d.f.: degrees of freedom; GCA: general combining ability; SCA: specific combining ability; Φ_{ge} , Φ_{sc} , and Φ_{rc} : quadratic components from GCA, SCA, and reciprocal effects, respectively.

Table 2. Summary of multivariate diallel analysis in nine *Jatropha* genotypes.

Sources of variation	d.f.	Matrix of SPS	Wilk test
General combining ability (GCA)	2	SPS (GCA)	<0.00
Specific combining ability (SCA)	3	SPS (SCA)	<0.00
Reciprocal	3	SPS (Reciprocal)	<0.00

SPS: sum of product square.

Analysis of the first two principal components retained 81% of the accumulated variance between the traits and, therefore, estimates of GCA and SCA were obtained on these components (Ledo et al., 2003). According to Cruz et al. (2012), for a satisfactory interpretation on the variability manifested among genotypes, it is necessary that the first two principal components have minimum estimates of 80% of total variation within the analyzed trait set.

To express graphically the Pearson correlation between the traits evaluated and the first two principal components, we used the correlation network, where green and red lines highlighted represent positive and negative correlations of high magnitude, respectively (Figure 1). We can verify the subdivision into the two groups: the first principal component (PC1) is positively associated with PH, CPL, and CPR; the second principal component (PC2) is positively related to the MHG, SD, and YIE.

Subdivision of traits into two principal groups is important because it met the group of first traits related to the size of the *Jatropha* genotypes, which must be reduced to ease the denser planting and mechanized harvesting. These traits are important in choosing the most appropriate spatial arrangement for planting because *Jatropha* can exceed 5 m high (Dias et al., 2007). Thus, efforts by breeding programs are focused on selecting plants with reduced size to facilitate the harvest. Teodoro et al. (2016b) observed that the MHG and SD have relation of cause and positive effect on grain yield in *Jatropha*, which reiterates the association of these traits with PC2.

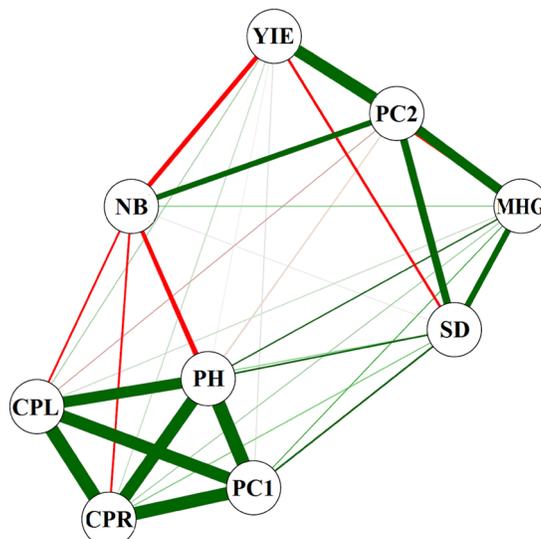


Figure 1. Network Pearson correlations between the first two principal components and agronomic traits assessed in nine *Jatropha* genotypes.

Pešek and Baker index (1969) was used to estimate the linear coefficients that allow positive gains (~10%) on the mass of hundred grains and grain yield and reduction (~10%) on plant height, which are the main goals of the *Jatropha*-breeding programs, and the other traits evaluated were considered secondary (Figure 2). After establishment of linear combinations, genotypic aggregate estimates were obtained as procedures reported by Cruz et al. (2012). It is important to mention that even the dominance effects being predominant in the genetic control for all traits, the genotypes 107 and 190 (derived from self-pollination) had genotypic aggregate estimates greater than most hybrids. These results indicate the absence of inbreeding depression for the main agronomic traits of *Jatropha*.

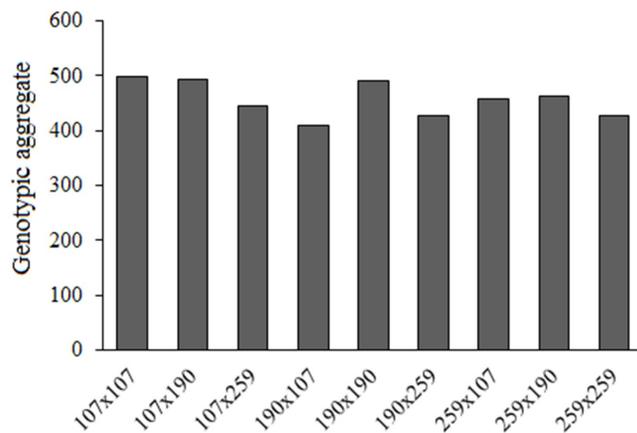


Figure 2. Genotypic aggregate obtained by the Pešek and Baker index (1969) for nine *Jatropha* genotypes.

GCA for the parents, estimated from the first two principal components and the selection index is shown in Figure 3. GCA estimates provide information on the concentration of predominantly additive genes and their effects have allowed the identification of parents used to obtain populations to practice the selection (Cruz et al., 2012). We verified that the genotypes 107 and 190 showed GCA estimates negative for PC1 and positive for PC2 and genotypic aggregate obtained by Pešek and Baker index (1969). Therefore, these results indicate that these parents are the most indicated to compose the cross blocks for having alleles to increase the YIE and MHG, and to reduce the size in *Jatropha*, which are the main objectives of the crop-breeding programs.

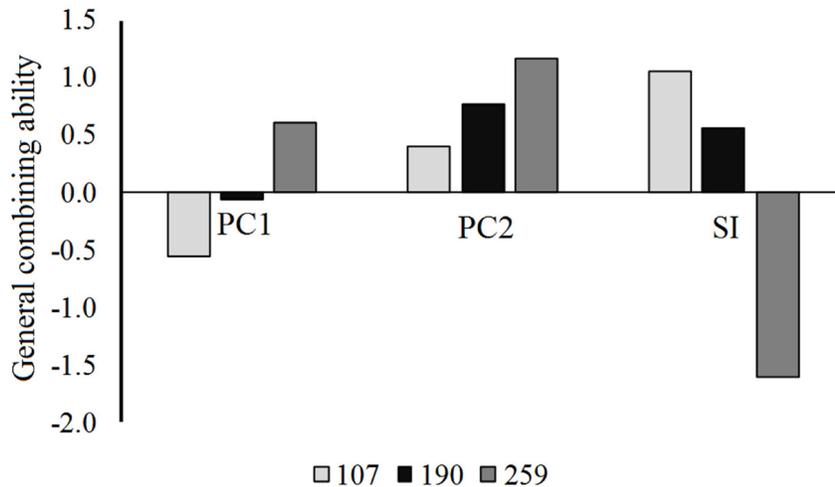


Figure 3. General combining ability of *Jatropha* genotypes obtained by the first two principal components (PC1 and PC2) and genotypic aggregate obtained by the selection index (SI) of Pešek and Baker (1969).

SCA can be defined as a deviation from the performance of the cross in relation to what would be expected based on the GCA from its parents. Its effects indicate the presence of non-additive interactions resulting from allelic complementation among the parents, allowing an improvement in the estimation of the genetic gain due to heterosis exploitation using divergent crosses (Cruz et al., 2012). Thus, from Figure 4 is possible to verify that the cross 190 x 107 obtained SCA negative estimates for CP1 and positive for CP2 and SI, being the most promising for the selection of genotypes in the segregating generations from its self-pollination. In addition, this cross gathers the two parents that have alleles more favorable for simultaneous improvement in plant height, mass of hundred grain, and grain yield.

From the results obtained in this study, it is possible to conclude that using multivariate diallel analysis allows more detailed information on the hybrid combinations with greater chances of genetic progress for the simultaneous selection of the main traits of interest. Silva et al. (2008) and Benin et al. (2009) also reported the effectiveness of this technique in identifying the parents and superior hybrid combinations aiming at gaining multiple traits in wheat.

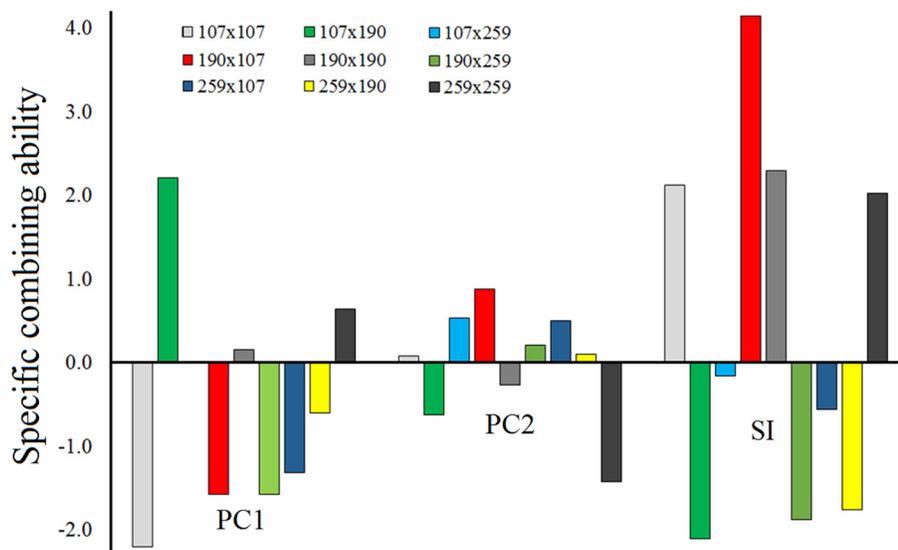


Figure 4. Specific combining ability of *Jatropha* genotypes obtained by the first two principal components (CP1 and CP2) and genotypic aggregate obtained by the selection index (SI) of Pešek and Baker (1969).

Conflicts of interest

The authors declare no conflict of interest.

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