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Genetic variability among *Cnidoscolus phyllacanthus* (Mart.) Pax. et K. Hoffm. mother trees in nursery conditions

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ABSTRACT - The genetic variability among 36 Cnidoscolus phyllacanthus mother trees was estimated to select higher quality genotypes for seedling production. The experiment was structured in a randomized complete-block design with 36 open-pollinated families in four replications. Each plot consisted of four 280 cm³ plastic containers with one seedling each. Data of the seedling collar diameter (CD) and height, number of thorns cm⁻¹ of the main vein of the leaf, and root (RDM), stem, leaf, aboveground and total dry matter were collected after six months. The seedling collar diameter and dry matter generally presented significant and high genetic correlations. The expected genetic gains resulting from the selection among families ranged from 7.91 to 23.48% to CD and RDM, respectively. Selection based on CD maximized the genetic gains (66.10%).

Key words: tree improvement, genetic parameters, genotype correlation, phenotype correlation, semi-arid.

INTRODUCTION

Cnidoscolus phyllacanthus (Mart.) Pax. et K. Hoffm. (faveleira) is an allogamous, xerophilous tree that grows in the northeastern semi-arid region of Brazil. The monoecious and, mainly, the protogynous characteristics are responsible for the predominance of cross-fertilization in this species. Its dry leaves and bark are eaten by cattle, sheep, goats and donkeys. The seeds can be fed to chicken and pigs and produce edible oil and flour, rich in mineral and protein.

The species is widely used as medicinal plant by people living within the area of occurrence. Ohta et al. (1994) in Japan, Ho and Chen (1995) in China and Ng and

Wege (1996) in Australia report its importance for the production of antibiotics and analgesics. It may also be used as firewood and low quality wood or in the recovery of degraded areas (Lorenzi 1998). In view of the high economic potential the faveleira tree has for the semi-arid region in northeastern Brazil, the lack of information on silvicultural management and product extraction are calling for more studies on these issues.

Efficient strategies of plant improvement depend on knowledge about the genetic properties of the populations that can be visualized by their genetic parameters (Resende 2002). This kind of knowledge has resulted in great advances in the forestry sector, especially for *Eucalyptus* and *Pinus* species.

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The easiest and most practical way of obtaining genetic gains for a single trait is to select it directly. This may however affect other traits in a direction and magnitude that depends on each one of them and the associations they have with each other. In general, genetic improvement does not aim at the development of a single, but of sundry traits (Paula et al. 2002). The study of the genetic correlation, which expresses the degree of genetic association among the traits or the influences they exercise on others, is therefore very important (Resende 2002).

When data obtained at different ages are considered, correlations account for important information in the process of early selection of high quality genotypes. Early selection is especially useful when dealing with perennial species because it reduces the length of each selection cycle (Gonçalves et al. 1998). However, for an effective early selection, a significant, high magnitude correlation between the juvenile and mature values of the traits of interest is required. Additionally, knowledge on the association among traits becomes fundamental when selection is difficult for a particular trait due to its low heritability and/or owing to problems in measurement and identification (Cruz and Carneiro 2003). Thus, correlations are also important to estimate the gains correlated to the selection and for the establishment of the selection index (Vencovsky and Barriga 1992, Ramalho et al. 2000a).

High-quality seedlings produced from seeds or vegetative parts of high quality trees are essential at establishing a forest stand. Such trees can be identified by studying genetic parameters of the population of interest. The present study had the objective to assess the genetic variability among faveleira mother trees from a natural population and select high quality genotypes for seedling production.

MATERIAL AND METHODS

Seeds from open pollination were collected from 36 mother trees in a natural stand where faveleira trees predominate in the Núcleo de Pesquisa para o Semi-árido (NUPEARIDO), an experimental station of the Centro de Saúde e Tecnologia Rural, Universidade Federal de Campina Grande (UFCG), in Patos, Paraíba, Brasil. The tree density in this 16-ha stand has an average of 22 faveleira trees ha⁻¹, with a mean of 5.29 + 0.41 m (minimum and maximum heights: 4.2 and 8.1 m, respectively), and a mean DBH 11.54 + 0.84 cm (1.3 m above ground) (minimum and maximum DBH: 5.4 and 20.7 cm). The 36 faveleira mother trees were chosen for their high flowering and fruit

production capacity, as well as good health conditions. The tree trunks were numbered and spaced at least 20 m apart from each other. The collected seeds were sent to the Faculdade de Ciências Agrárias e Veterinárias, Universidade Estadual Paulista (FCAV/UNESP), in Jaboticabal, São Paulo, Brasil, soon after harvesting, drying, and removal from the fruit shell. The seed dormancy was broken by manual scarification with sandpaper # 120. The seeds were then placed on a washed and sterilized sand substratum in transparent 11 x 11 x 4 cm plastic boxes and covered. The temperature was kept at 20 or 35 °C, corresponding, respectively, to an eight hours light and 16 hours darkness regime. After 10 days, the germinated seeds were transferred to 280 cm³ plastic containers (tubes).

The tubes were maintained 3 cm above the bench level when placed in 60 x 40 cm polypropylene trays holding 48 units. The seedlings in these tubes were grown in a greenhouse. In the first two weeks, seedlings were watered (1250 mL tray⁻¹) daily by submerging the open bottom tip of the tubes in water. Thereafter, the tubes were irrigated thrice a week.

Infesting plants were removed from the tubes monthly. White acari (*Polyphagotarsonemus latus*) were controlled by spraying abamectina (3 mL 10 L⁻¹ of water) 75 and 85 days after seedling transplantation. A nutritive solution (30 mg of N dm⁻³; 30 mg of P dm⁻³; 30 mg of K dm⁻³; 10 mg of S dm⁻³; 0.5 mg of Zn dm⁻³ and 0.5 mg of B dm⁻³) was applied 120 days after transplantation. Three weeks later, the double amount of nutrients was applied likewise.

Six months after seedling transplantation (June 2002), the following data were collected: seedling collar diameter (CD) (mm plant⁻¹) and height (H) (cm plant⁻¹), number of thorns cm⁻¹ on the main vein of the third fully developed leaf from the top (NT), and the dry matter of the seedling's root (RDM), stem (SDM), leaf (LDM), as well as the aboveground (AGDM) and total dry matter (TDM) (g plant⁻¹). Diameter and height were measured by means of a plastic ruler (rounded to the nearest half cm) and a digital paquimeter (accurate to one hundredth of a mm), respectively. Dry matter was obtained at 65 ± 3 °C in a forced air circulation oven until the plant material reached constant mass.

The experiment was conducted in a randomized block design, with 36 treatments (families) replicated in four 4-tube plots. Data on a per plant basis were considered for the analysis of variance and the treatments (families) as random factor. It was assumed that the open-pollinated families were composed by half-sib individuals, and the additive genetic variance ($\hat{\sigma}_{A}^{2}$) was estimated by $\hat{\sigma}_{A}^{2} = 4\hat{\sigma}_{p}^{2}$, where $\hat{\sigma}_{a}^{2}$ is the genetic variance between families. ANOVA,

variances, coefficients of variance, heritabilities, genetic correlations, and genetic gains from direct and indirect selections (correlated response) were estimated by software Genes (Cruz 2001). A selection intensity of 25% among families was adopted. Selection was simulated based on increments in all traits, except for NT. Significance tests (Student t test) of the genetic and phenotypic coefficient of correlations were run according to Gomes (2000) recommendations.

RESULTS AND DISCUSSION

There were significant differences among the openpollinated families for all traits considered in this study (Table 1). Significant differences among families indicate the existence of genetic variability among them and, consequently, the possibility of successful selection.

Paula et al. (2003) found significant differences for seedling collar diameter and height and leaf dry matter among 18 open-pollinated families of *Eucalyptus grandis* 120-day old seedlings under greenhouse conditions. They found no significant differences for root, stem and aboveground dry matter (P > 0.05).

Compared to the above mentioned literature data, the results obtained in the present study corroborate a more favorable situation for selection, possibly because faveleira has so far not been submitted to genetic improvement and because a higher number of trees was considered.

Data in Table 1 show that the narrowest range was observed for collar diameter (CD), as the maximum value

exceeds the minimum by 48%, while the largest difference was observed for root dry matter (RDM) (152%). Except for height (H), the differences of all traits surpassed 100%. Both growth (CD and H) and dry matter trait differences between the highest and lowest-performance families were smaller than those reported by Paula et al. (2003).

The coefficients of variation (CV) of the experiment were low for CD, intermediate for H, RDM and TDM, and high for the other traits, according to Gomes (2000)'s classification.

Heritability is a parameter frequently used by crop breeders, as it makes the prediction of the possible success of selection possible (Ramalho et al. 2000b). Estimates of family heritability in the strict sense (h_m^2) ranged from 0.43 to 0.83, respectively, for leaf dry matter (LDM) and RDM (Table 1). Reported values of for open-pollinated families of *Eucalyptus grandis* (Paula et al. 2003) were similar for LDM and aboveground dry matter (AGDM), and lower for the other traits.

The heritability (h_m^2) value was high for TDM, and intermediate for AGDM and the number of thorns (NT) (Table 1). Heritability values for D and H in the present study (Table 1) were in the mean higher than those found for other tree species in the phases of seedling and initial establishment in the field (Farias Neto and Bianchetti 2001, h^Paiva et al. 2001). In general, heritability was high for most of the studied traits (Table 1). This fact, along with the great genetic variability among the families, makes it clear that selection on these traits is promising. It should be pointed out that the families derived from non-selected genotypes and the experimental precision were satisfactory, which surely contributed to the high heritability estimates.

Table 1. Summary of analysis of variance and heritability (h_m^2) estimate for collar diameter (CD), height (H), number of thorns (NT), and root (RDM), stem (SDM), leaf (LDM), aboveground (AGDM) and total (TDM) dry matter of 36 open-pollinated *Cnidoscolus phyllacanthus* families of 180-day-old seedlings growing under greenhouse conditions

Sources of variation	df	Mean Squares									
		CD mm	H cm	NT	RDM g	SDM g	LDM g	AGDM g	TDM g		
Blocks	3	0.49	71.53	0.37	0.95	0.18	0.29	0.92	2.50		
Families	35	0.84**	14.30**	0.22**	2.04**	0.08**	0.04*	0.17**	2.81**		
Error	105	0.19	4.45	0.10	0.34	0.04	0.02	0.09	0.62		
Mean		5.45	19.36	1.48	3.41	0.84	0.53	1.38	4.79		
Range		4.25 to 6.29	14.63 to 23.06	1.07 to 2.21	1.89 to 4.76	0.51 to 1.18	0.35 to 0.76	0.87 to 1.76	2.76 to 6.22		
CV (%)		8.05	10.90	21.60	17.04	22.35	29.50	21.95	16.50		
		0.77	0.69	0.54	0.83	0.55	0.43	0.46	0.78		
$CV_{g}(\%)$		7.43	8.10	11.64	19.13	12.30	12.96	10.12	15.45		
CV /CV		0.92	0.74	0.54	1.12	0.55	0.44	0.46	0.93		

* P < 0.05, ** P < 0.01

The estimates of the coefficients of genetic variation among families (CVg) and the estimates of the ratio CVg/ CV are in Table 1. The highest estimates were obtained for RDM. Heritability and CVg/CV ratio allow a better estimation of the expected genetic gain from selection. Vencovsky and Barriga (1992) underscored that heritability and CVg/CV estimates close to or higher than the unity express a favorable situation for genetic selection. Thus, CD, RDM and TDM are traits with higher and CVg/CV values than NT, LDM and AGDM. Paula et al. (1996) however emphasized that the magnitude of CVg and CV should also be considered. For instance, CVg/CV is similar for CD and TDM while the expected gain from selection for TDM should be higher due to its higher CVg. Thus, RDM and TDM family means are expected to show higher increases during the selection process.

Most of the phenotypic and genotypic correlations were significant (Table 2). The highest phenotypic correlation was observed between RDM x TDM and CD x TDM, while the genotypic correlation was intermediate between CD x stem dry matter (SDM) and RDM x TDM. In general, the coefficients of genotypic correlation were higher than the coefficients of phenotypic correlation (Table 2). Paula et al. (2002) pointed out that these results show that the genetic factors are more important than the environmental ones in the expression of at least one of the traits. The selection process on one or a few traits may have little or no effect on other not less important traits, be it positively or negatively (Paula et al. 1998). The study of the correlation between traits allows the use of more suitable strategies to avoid the negative effect the selection of one trait may have on others.

Most of the genotypic correlation estimates were high, positive and statistically significant (Table 2), probably due to pleiotropic genes or genic linkage. Genic linkage is of transitory nature, as it can be broken in

0.99**

0.87**

0.95**

0.27

0.77**

0.67**

0.36*

0.21

following segregating generations (Falconer and Mackay 1996). When the objective is to improve all traits in one and the same direction, positive correlation is desirable, as the increase of one of them implies a concomitant augmentation of other positively correlated traits. However, the positive correlations observed between NT and the other traits are undesirable, as the natural objective to decrease the number of thorns involves a decrease in CD, SDM and AGDM. Paula et al. (2002) mentioned that a high genotypic correlation between certain types of traits may mean that they are controlled by pleiotropic genes. High genotypic correlation values are observed between CD and RDM, CD and SDM, CD and AGDM, CD and TDM, RDM and TDM, and SDM and AGDM (Table 2). If these authors' statements are true, the positive correlations involving NT would suggest that these traits are controlled by linked genes, and, consequently, the NT-positivecorrelation problem could certainly be overcome by link breakage in future segregating generations. In this case, plant breeders should use selection strategies to identify appropriate recombinants (i.e.: plants with less or no thorns that show desirable magnitude in the other traits of interest).

Height and collar diameter are non-destructive traits, easy to measure, especially under nursery conditions. Collar diameter is significantly (P < 0.01) correlated with almost all plant DM traits (Table 2). This is a very important result. Consider CD x RDM correlation. The root system plays a fundamental role in nutrient and water storage in faveleira trees, especially in the initial phase of seedling establishment in the field under high water stress, so common in its habitat. Thus, a seedling that develops a strong root system has its advantages. However, to evaluate the root system, it is necessary to destroy the plant. Collar diameter evaluation can be a non-destructive alternative, because of the high correlation between CD

Traits	CD	Н	NT	RDM	SDM	LDM	AGDM	TDM
CD		0.47**	0.38*	0.85**	0.83**	0.32	0.73**	0.91**
H	0.44**		0.03	0.30	0.75**	0.33	0.68**	0.42*
NT	0.52**	0.06		0.14	0.26	0.04	0.20	0.17
RDM	0.90**	0.26	0.20		0.65**	0.13	0.51**	0.98**

0.76**

0.56**

0.99*

0.27

0.88**

0.84**

0.02

0.45**

0.04

0.35*

0.24

Table 2. Estimates of phenotypic (above) and genotypic (bellow the diagonal) correlations between collar diameter (CD), height (H), number of thorns (NT), and R (RDM), stem (SDM), leaf (LDM), aboveground (AGDM) and total (TDM) dry matter of 36 open-pollinated *Cnidoscolus phyllacanthus* families of 180-day-old seedlings growing under greenhouse conditions

* P < 0.05, ** P < 0.01 by the Student t test

SDM

LDM

AGDM

TDM

0.88**

0.78**

0.68**

0.77**

0.68**

0.30

0.40*

0.70**

0.11

and RDM and other biomass traits. This is not true in the case of height, so this trait is not recommended as a referential for RDM selection.

The expected gains in direct and indirect selections are presented in Table 3. The expected gain is a minimum for CD and H and maximum for RDM and TDM. Expected gains for CD and H were similar to those estimated by Farias Neto and Castro (1998), in 21 Sclerolobium paniculatum open-pollinated families at the age of 96 months. With increasing tree age, Gonçalves et al. (1998) observed an increase in the estimates of genetic parameters of rubber trees (Hevea brasiliensis). Borges et al. (1980) also verified this same trend of increase in height and diameter heritability in E. grandis open-pollinated families. This increase can be explained by the stronger effect the environment exerts on young plants. As the trees age, the genotype surpasses the environment in the phenotypic expression. However, Sebbenn et al. (2003) observed that height heritability values in Araucaria angustifolia trees decreased from the first to the third year, and appeared to be constant from the fifth to the 18th year. Nevertheless, assuming an increase of heritability as the faveleira ages, an increase in genetic gain values is to be expected compared to the values obtained for seedlings in the present study.

The gain estimation among families is very important for seedling production because it allows the identification of high quality trees for seed collection. On the other hand, within-family selection can be practiced by means of a stratified selection, by early selection of the best seedlings of each family. Traditionally, this is done by visual selection of the best seedlings, a not very precise procedure.

High expectations of direct gains are not necessarily related to high h_m^2 and CV/CVg ratio values. This agrees with the findings of Paula et al. (1996) in that the magnitudes of the genetic (CVg) and experimental (CV)

coefficients of variation should also be considered. For example, SDM, LDM and AGDM showed higher expected gains than CD and H (Table 3), although their h_m^2 and CVg/CV were lower than the respective values for CD and H. (Table 1). This can be attributed to the superior CVg values of the DM traits.

Paula et al. (2002) pointed out that when genetic gain is estimated based on differential selection, indirect selection will be, at the most, equal to and never higher than direct selection. This can be verified in Table 3, as the indirect gain estimates of every trait were lower throughout than those of direct selection gains. For example, RDM direct selection gain estimate totaled 23.48%, while the highest indirect gain estimate for this trait (23.23%) was obtained with the direct selection in TDM.

Considering the entire genetic gain obtained directly from the selection on a particular trait and indirectly from the selection on other traits, selection for collar diameter was the one that maximized total genetic gains (66.10%) (Table 3). Paula et al. (2003) highlighted that the most important traits in seedling production are collar diameter, height, and stem, leaf and aboveground dry matter. Thus, favela trees number 17, 54, 42, 58, 01, 52, 03, 57, and 04 are the best ones, because their progenies performed best for these and the entire set of traits considered in this study (Table 4).

CONCLUSIONS

1. Due to the presence of high genetic variability among the 36 open-pollinated families of *Cnidoscolus phyllacanthus* (Mart.) Pax. et K. Hoffm. (Euphorbiaceae) seedlings grown in nursery conditions, it can be concluded that it is possible to select superior quality genotypes successfully.

Table 3. Estimates of the percentage of genetic gains from direct (diagonal) and indirect (out of diagonal) selections for collar diameter
(CD), height (H), number of thorns (NT), and root (RDM), stem (SDM), leaf (LDM), aboveground (AGDM) and total (TDM) dry matter
based on 36 open-pollinated Cnidoscolus phyllacanthus families of 180-day-old seedlings growing under greenhouse conditions

Selected	Expected gain (%)									
trait	CD	Н	NT	RDM	SDM	LDM	AGDM	TDM	gains	
CD	7.91	2.31	4.40	18.77	9.39	2.29	5.76	15.27	66.10	
Н	3.23	8.47	- 0.67	8.66	9.15	1.51	5.31	8.34	44.00	
ΝT	3.22	2.06	- 9.29	- 5.86	- 0.78	- 0.01	- 0.41	- 4.09	- 21.60	
RDM	6.13	- 0.57	- 1.61	23.48	5.27	- 0.66	2.44	16.78	51.26	
SDM	6.50	5.92	3.03	14.28	11.36	3.35	7.20	12.99	64.63	
LDM	1.74	1.46	- 0.58	- 1.00	3.60	10.91	6.31	2.41	24.85	
AGDM	4.81	4.26	2.03	6.47	9.66	8.77	8.55	8.46	53.01	
TDM	7.29	1.62	0.28	23.23	8.44	0.08	4.37	17.56	62.87	

Table 4. Mean collar diameter (CD), height (H), number of thorns (NT), and root (RDM), stem (SDM), leaf (LDM), aboveground (AGDM) and total (TDM) dry matter of open-pollinated *Cnidoscolus phyllacanthus* families of 180-day-old seedlings growing under greenhouse conditions

Families	CD mm	H cm	NT	RDM	SDM	LDM g	AGDM	TDM
01	6.00	18.60	1.50	4.76	0.86	0.54	1.41	6.17
02	5.74	16.75	1.53	4.41	0.80	0.55	1.34	5.75
03	5.88	18.22	1.47	4.20	0.85	0.46	1.30	5.51
04	5.84	19.81	1.59	3.81	0.90	0.56	1.45	5.26
05	5.33	17.10	1.40	3.85	0.74	0.50	1.24	5.09
06	5.61	20.50	1.37	3.31	0.88	0.76	1.64	4.96
07	5.34	19.28	1.34	3.13	0.77	0.62	1.39	4.53
08	5.81	20.06	1.86	3.44	0.93	0.40	1.33	4.77
09	5.80	21.57	1.24	4.65	0.95	0.39	1.34	6.00
10	4.90	21.38	1.28	2.83	0.86	0.44	1.30	4.13
11	5.32	16.88	1.51	3.69	0.71	0.37	1.08	4.77
12	5.07	18.72	1.25	3.16	0.71	0.46	1.17	4.33
14	5.23	19.38	1.43	3.07	0.70	0.50	1.20	4.27
15	5.59	20.94	1.48	3.59	0.83	0.58	1.41	5.00
17	6.29	20.78	1.42	4.56	1.05	0.51	1.57	6.13
18	4.79	18.06	1.50	2.48	0.67	0.37	1.04	3.52
19	5.61	20.35	1.62	3.10	0.88	0.51	1.39	4.50
20	5.19	20.60	1.29	2.94	0.76	0.48	1.24	4.18
22	5.00	17.72	1.49	2.20	0.66	0.35	1.00	3.20
23	4.77	19.31	1.28	2.46	0.67	0.73	1.40	3.86
29	5.44	22.25	1.75	3.55	0.96	0.58	1.54	5.09
30	4.82	17.91	1.21	2.72	0.75	0.49	1.23	3.96
32	4.94	17.07	1.35	2.66	0.68	0.49	1.17	3.83
41	4.25	14.63	1.57	1.89	0.51	0.37	0.87	2.76
42	6.07	22.62	1.87	3.76	1.18	0.58	1.76	5.52
43	5.46	17.72	1.88	2.94	0.81	0.60	1.42	4.35
44	5.59	21.97	1.22	2.92	0.96	0.70	1.67	4.58
45	5.28	17.41	1.49	3.35	0.76	0.53	1.29	4.65
46	4.99	19.72	1.07	2.96	0.85	0.50	1.34	4.30
52	5.99	23.06	1.41	4.58	1.11	0.52	1.64	6.22
53	5.40	18.75	1.32	3.13	0.91	0.66	1.57	4.70
54	6.09	20.25	2.21	3.61	0.98	0.62	1.61	5.22
57	5.87	18.31	1.17	3.90	0.96	0.62	1.58	5.48
58	6.03	18.44	1.78	4.42	0.99	0.63	1.62	6.04
59	5.40	19.82	1.63	3.32	0.78	0.59	1.37	4.68
61	5.43	21.13	1.48	3.45	0.94	0.66	1.60	5.05

2. Based on all studied traits, direct selection on collar diameter maximizes the genetic gains, showing that collar diameter is a good alternative for the selection of the studied traits.

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Variabilidade genética entre matrizes de *Cnidoscolus phyllacanthus* (Mart.) Pax. et K. Hoffm. na fase de mudas

RESUMO - Avaliou-se a variabilidade genética entre 36 matrizes de Cnidoscolus phyllacanthus (faveleira), visando a seleção de genótipos superiores para a produção de mudas. O delineamento utilizado foi o de blocos casualizados, com 36 famílias de polinização aberta e quatro repetições. As parcelas foram constituídas de quatro plantas, uma em cada tubete de 280 cm³. Seis meses após a repicagem, foram coletados os seguintes dados nas mudas: diâmetro do coleto (D), altura, número de espinhos/cm da nervura principal da folha e matéria seca de raiz (MSR), de caule, de folhas, da parte aérea e total. O Diâmetro do coleto apresentou correlações genéticas significativas e de altas magnitudes com quase todos os componentes da matéria seca da planta. As expectativas de ganhos com a seleção entre famílias variaram de 7,91 a 23,48% para D e MSR, respectivamente. A seleção praticada no diâmetro do coleto foi a que maximizou o total dos ganhos genéticos (66,10%).

Palavras-chave: melhoramento florestal, parâmetros genéticos, correlação genotípica, correlação fenotípica, semi-árido.

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