

## CORRELATIONS AND PATH ANALYSIS ON OIL CONTENT OF CASTOR GENOTYPES

### *CORRELAÇÕES E ANÁLISE DE TRILHA SOBRE O TEOR DE ÓLEO DE GENÓTIPOS DE MAMONA*

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**ABSTRACT:** With the aim of to evaluate the correlation and the direct and indirect effects of morphological descriptors on oil content of castor (*Ricinus communis* L.) genotypes, an experiment was conducted in the agricultural years 2010/2011 and 2011/2012 at Universidade Estadual de Mato Grosso do Sul – Unit of Aquidauana (UEMS/UUA). The experimental design used was randomized blocks with three replications. The treatments consisted of seven castor genotypes (BRS Energia, IAC-2028, IAC-Guarani, BRS Nordestina, BRS Paraguaçu, IAC-80 and IAC-226). The following descriptors were evaluated: plant height (PH), stem diameter (SD), number of bunches (NB), number of fruits (NF), mass of fruits (MF), mass of hundred seeds (MHS) and oil content (OC). Initially, it was determined the following genetic parameters: genotypic variance, environmental variance, coefficient of experimental variation, coefficient of genotypic variation, heritability, environmental correlations, phenotypic correlations and genetic correlations. The phenotypic correlations were deployed by path analysis in estimates of direct and indirect effects, and preceded the diagnosis of multicollinearity in explanatory variables and independent explanatory variables on the OC (primary dependent variable). The results indicated that the direct and indirect selection of genotypes with plant height, stem diameter, number of bunches and mass of hundred seeds is promising to select genotypes with high oil content in castor.

**KEYWORDS:** Biodiesel. Genetic breeding. *Ricinus communis* L. Vegetable oil.

### INTRODUCTION

Castor (*Ricinus communis* L.) is a valuable oilseed crop that provides almost the entire world's supply of hydroxy fatty acids (TORRES et al., 2015). It is used in the production of lubricants, paints, soaps and pharmaceuticals. India is the largest producer of castor seed and the United States imports 44.000 mega grams of castor oil annually (GHANDI et al., 1994, ADEYANJU et al., 2010; TORRES et al., 2014, 2015).

The castor belongs to the family Euphorbiaceae. It is an essentially tropical/subtropical species and it grows naturally over a wide range of geographical regions including temperate areas. Exploitation of castor ranges from the simple harvesting of beans from wild plants through the cultivation of hybrid varieties and the use of improved cultivation methods. Wild plants are perennials, but where it is deliberately cultivated an annual production cycle is possible (GOGOI et al., 2009; DHEDHI et al., 2010).

One of the primary objectives of castor breeders is to increase the oil content, therefore

several researches to identify which major genetic and environmental factors that are interrelated with this variable have recently been conducted (JIN-QUIN et al., 2004; YADAV et al., 2004; RAMU et al., 2005; GOGOI et al., 2009; ADEYANJU et al., 2010; DHEDHI et al., 2010; TORRES et al., 2013). Generally, oil content represents the final character resulting from many developmental and biochemical processes which occur between germination and maturity. Before yielding improvements can be achieved, the breeder needs to identify the causes of variability in oil yield in any given environment. Since fluctuations in environment generally affects yielding through its components.

Ramu et al. (2005) suggested that individual yield components may contribute with valuable information to many researchers who have analyzed breeding through yielding components. In as much as the determination of the correlation coefficient of yield and its components are undoubtedly helpful to breeders in selecting suitable plant types based on simultaneous selection of two or more characters, a better approach of character association is the path

analysis (GOGOI et al., 2009; ADEYANJU et al., 2010; DHEDHI et al., 2010; TEODORO et al., 2014, 2015). Whereas correlation is simply a measurement of mutual association, with no regards to its cause, path-coefficient analysis specifies the cause and measures their relative importance. According to Cruz et al. (2004), this technique is most useful when conditions allow for its application.

In this context, with the purpose of contributing to the genetic improvement of castor, the present study aimed at evaluating the correlation and the direct and indirect effects of morphological traits on the oil content of genotypes of this crop.

## MATERIAL AND METHODS

The experiment was installed in the agricultural years 2010/2011 and 2011/2012 in the experimental area of the Universidade Estadual de Mato Grosso do Sul, (State University located in the city of Aquidauana, Mato Grosso do Sul - Brazil) - (UEMS/UUA). Is it specifically located in the Brazilian Savanna, whose coordinates comprise 20°27'S and 55°40'W with an average elevation of 170 m.

The soil was classified as Ultisol sandy loam texture (TORRES et al., 2014), with the following chemical features in the layer 0 – 0.20 m: pH (H<sub>2</sub>O) = 6.2; Al exchangeable (cmol<sub>c</sub> dm<sup>-3</sup>) = 0.0; Ca+Mg (cmol<sub>c</sub> dm<sup>-3</sup>) = 4.31; P (mg dm<sup>-3</sup>) = 41.3; K (cmol<sub>c</sub> dm<sup>-3</sup>) = 0.2; Organic matter (g dm<sup>-3</sup>) = 19.7; V (%) = 45.0; m (%) = 0.0; Sum of bases (cmol<sub>c</sub> dm<sup>-3</sup>) = 2.3; cation exchange capacity (CEC) (cmol<sub>c</sub> dm<sup>-3</sup>) = 5.1. The climate of the region, according to the classification described by Köppen-Geiger, is Aw (Savanna Tropical) with a cumulative rainfall during the experiment of 450 mm and the maximum and the minimum temperatures were 19 and 33°C, respectively.

The experimental design was a randomized block with three replications, and the treatments consisted of seven genotypes of castor (BRS Energia, IAC-2028, IAC-Guarani, BRS Nordestina, BRS Paraguaçu, IAC-80 e IAC-226), acquired from Empresa Brasileira de Pesquisa Agropecuária, (Embrapa), a Brazilian Enterprise of Brazilian Reseach, and from Instituto Agrônômico de Campinas (IAC), an Agronomic Institute of Campinas.

Sowings were made on December 10<sup>th</sup>, 2010 and 2011. The preparation of the area consisted of a mechanical mowing and disking, and placed two seeds per hole, with a depth of 15 cm, open manually with the assistance of a mattock. All

other cultural practices were performed as described in Torres et al. (2013, 2015).

At harvest the following descriptors were evaluated: plant height (PH), stem diameter (SD), number of bunches (NB), number of fruits (NF), mass of fruits (MF), mass of a hundred seeds (MHS) and oil content (OC). The OC was determinate according to the procedure described in Li (2004).

In the analysis of results was used the analysis of variance, adopting the following statistical model (CRUZ et al., 2004):  $Y_{ij} = \mu + g_i + b_j + e_{ij}$ , wherein:

$Y_{ij}$  = characteristic value of the i-nth genotype on the j-th block;

$\mu$  = overall average of test;

$g_i$  = effect of i-nth genotype (i = 1,2, ... , g);

$b_j$  = effect of j-th block (j = 1,2 ... , r); e

$e_{ij}$  = random error associated with the genotype i and block j  $e_{ij} \sim NID(0, s^2)$ .

Genetic parameters and their estimators were analyzed for each characteristic using the following expressions (CRUZ et al., 2004):

a) Genotypic variance:

$$\hat{\sigma}_G^2 = \frac{QM_g - QM_r}{k};$$

b) Environmental variance:

$$\hat{\sigma}_E^2 = \frac{QM_r}{k};$$

c) Phenotypic variance:  $\hat{\sigma}_F^2 = \frac{QM_g}{k}$

d) Coefficient of genotypic variation:

$$CV_g = \left( \frac{\sqrt{\hat{\sigma}_G^2}}{m} \right) \times 100;$$

e) Coefficient of experimental

$$\text{variation: } CV_e = \left( \frac{\sqrt{QM_e}}{m} \right) \times 100;$$

f)  $b = \frac{CV_g}{CV_e};$

g) Heritability:  $\hat{h}^2 = \frac{\hat{\sigma}_G^2}{\hat{\sigma}_E^2};$

In the estimates of the correlations were employed in mentioned expressions by Ramalho et al. (1993):

h) Phenotypic correlation:

$$r_F = \frac{COV_{F(xy)}}{\sqrt{\hat{\sigma}_{F_x}^2 \times \hat{\sigma}_{F_y}^2}};$$

i) Genotypic correlation:

$$r_G = \frac{\text{COV}_{G(xy)}}{\sqrt{\hat{\sigma}_{Gx}^2 \times \hat{\sigma}_{Gy}^2}};$$

j) Environmental correlation:

$$r_E = \frac{\text{COV}_{E(xy)}}{\sqrt{\hat{\sigma}_{Ex}^2 \times \hat{\sigma}_{Ey}^2}}$$

wherein:  $r_{xy}$  = correlation between characters X and Y;  $\text{COV}_{xy}$  = covariance between characters X and Y; and  $\hat{\sigma}_x^2$  and  $\hat{\sigma}_y^2$  = variance of characters X and Y, respectively.

Phenotypic correlations were deployed by path analysis in estimates of direct and indirect effects considering the following statistical model:  $Y = p_1x_1 + p_2x_2 + \dots + p_nx_n + p_eu$ , in which: y: basic variable or dependent dry matter productivity;  $x_1, x_2, \dots, x_n$ : explanatory variable;  $p_1, p_2, \dots, p_n$ : path coefficients.

Path coefficients were estimated through system proposed by Li (1975). Thus, the correlation between the dependent variable and the explanatory ( $r_{xy}$ ) is given by:

$$r_{1y} = p_1 + p_2 r_{12} + \dots + p_n r_{1n}$$

$$r_{2y} = p_1 r_{12} + p_2 + \dots + p_n r_{2n}$$

$$\dots = \dots$$

$$r_{ny} = p_1 r_{1n} + p_2 r_{2n} + \dots + p_n$$

The determination coefficient was calculated by:  $R^2 = p_{1y}^2 + p_{2y}^2 + \dots + 2p_{2y}p_{2n}r_{2n}$ .

The diagnosis of multicollinearity in the explanatory variables was performed based on the condition number (CN) established by Montgomery and Peck (2001), with  $NC < 100$  weak (not constituting a serious problem);  $100 < NC < 1000$ , moderate to strong, and  $> 1000$ , severe.

With the effect of multicollinearity detected, the analysis was performed as suggested by Cruz et al. (1994), being carried out with the assistance of a computer application Genes (CRUZ, 2006).

## RESULTS AND DISCUSSION

Table 1 gives an overview of the genetic parameters evaluated in this study. The experimental coefficient of variation ranged between (CVe) 7.7 (OC) and 25.0 (NF), which according to Lúcio et al. (1999) classifies experimental high precision. The genotypic coefficient of variation (CVg) varied between 0.4 (OC) and 35.4 (MHS), which indicates that among the descriptors evaluated MHS and NF are the ones who show greater variability, highly promising for the direct selection of superior genotypes. Considering that the existence of genetic variability in a population is crucial to any breeding program (RAMALHO et al., 2000), the germplasm under study proves in principle, promising to selection or hybridization with potential for development of new genotypes.

**Table 1.** Estimates of genetic parameters for seven morphological descriptors evaluated in seven castor genotypes.

Parameter	PH <sup>1</sup>	SD	NB	NF	OC	MF	MHS
Minimum	1.04 m	0.02 m	1.0	32.0	40.1 %	56.6 g	31.3 g
Maximum	2.94 m	0.06 m	3.3	177.4	55.3 %	759.93	127.9 g
CVg (%) <sup>2</sup>	11.0	14.9	28.3	32.0	0.4	23.5	35.4
CVe (%)	19.6	15.9	23.7	35.0	7.7	63.5	32.5
b	0.6	0.9	1.2	0.9	0.0	0.4	1.1
$\hat{\sigma}_F^2$	657.7	32.4	0.3	694.1	2.5	3,502.6	357.7
$\hat{\sigma}_G^2$	430.0	27.4	0.3	578.3	0.0	1,579.9	313.6
$\hat{\sigma}_E^2$	227.7	5.2	0.0	115.8	2.4	1,922.7	44.1
$\hat{h}^2$ (%)	65.4	84.0	89.5	83.3	1.95	45.1	87.7

<sup>1</sup>PH: plant height; SD: stem diameter; NB: number of bunches; NF: number of fruits; OC: oil content; MF: mass of fruits; MHS: mass of hundred seeds. <sup>2</sup>CVg: coefficient of genotypic variation; CVe: coefficient of experimental variation; b: relation between CVg and CVe;  $\hat{\sigma}_F^2$ : phenotypic variance;  $\hat{\sigma}_G^2$ : genotypic variance;  $\hat{\sigma}_E^2$ : environmental variance;  $\hat{h}^2$ : heritability.

The b quotient was greater than 1.0 only for NB and MHS, indicating favorable situations for the selection of all other descriptors according to the interpretation of this value recommended by

Vencovsky (1978). The descriptors PH, SD, NB, NF and MHS showed heritability ( $h^2$ ) greater than 60%, indicating that it less influenced by the environment (CRUZ et al., 2004). On the other hand, the low  $h^2$

of CB and MF descriptors occurred due to the higher estimate of the  $\hat{\sigma}_E^2$  in relation to  $\hat{\sigma}_G^2$ , indicating that these descriptors are highly influenced by the environment (CRUZ et al., 2004).

Table 2 shows significant phenotypic correlations ( $r_P$ ) ( $p < 0.05$ ) between descriptors PH x SD, PH x OC, PH x MHS, SD x OC, SD x MHS, OC x MHS and NF x MHS resembling the results obtained by Yadav et al. (2004), Gogoi et al. (2009), Adeyanju et al. (2010) and Dhedhi et al., (2010). Therefore, despite low genetic indications ( $CV_e$ ,  $CV_g$ ,  $b$  and  $h^2$ ) of the descriptor OC, the selection

of genotypes with high PH, SD and MHS will consequently result in genotypes with high oil content due to the fact that  $r_P$  is positive. This is probably due to the occurrence of pleiotropism or genetic linkage disequilibrium between pairs of characters that favor the simultaneous selection of two or more characters by selection in only one of these (FALCONER, 1987; GOLDENBERG, 1968). Moreover, the selection of a character can cause an undesirable selection of another character, for example, in case  $r_P$  be negative between NF x MHS (CORREA et al., 2015).

**Table 2.** Estimates of phenotypic ( $r_P$ ), genotypic ( $r_G$ ) and environmental ( $r_E$ ) correlations between six morphological descriptors evaluated in seven castor bean genotypes.

Descriptors		PH <sup>1</sup>	SD	NB	NF	OC	MF	MHS
PH	$r_G$	---	0.9709	-0.0216	-0.3393	0.4176	0.0491	0.9502
	$r_E$	---	0.7073	0.3001	0.0924	0.0117	-0.095	-0.0469
	$r_P$	---	0.8860*	0.0406	-0.2282	0.6185*	0.5283*	0.7097*
SD	$r_G$		---	-0.4048	-0.4740	0.8966	0.8136	0.8846
	$r_E$		---	0.3175	0.0718	0.0208	-0.0635	0.0077
	$r_P$		---	-0.3101	-0.3849	0.6350*	0.4820	0.7580*
NB	$r_G$			---	-0.1622	-0.4418	0.0017	-0.0124
	$r_E$			---	0.5228	0.4041	-0.2469	0.1186
	$r_P$			---	-0.0711	-0.3254	-0.0581	0.0025
NF	$r_G$				---	-0.0984	0.842	-0.8603
	$r_E$				---	0.3772	-0.0242	-0.2533
	$r_P$				---	-0.3699	0.5089	-0.7716*
OC	$r_G$					---	0.8809	0.4532
	$r_E$					---	-0.1676	0.0094
	$r_P$					---	-0.0403	0.5855*
MF	$r_G$						---	0.0023
	$r_E$						---	-0.0863
	$r_P$						---	-0.021
MHS	$r_G$							---
	$r_E$							---
	$r_P$							---

<sup>1</sup>PH: plant height; SD: stem diameter; NB: number of bunches; NF: number of fruits; OC: oil content; MF: mass of fruits; MHS: mass of hundred seeds. \*Significant at 0.05 probability by t-test.

Genotypic correlations ( $r_G$ ) showed the same signal, in most cases, superior to their corresponding  $r_P$  values, indicating that the phenotypic expression is decreased given the influences of environment, corroborating the results of Yadav et al. (2004), Gogoi et al. (2009), Adeyanju et al. (2010) and Dhedhi et al., (2010). Thus, the  $r_P$  may be useful in the absence of estimates of  $r_G$ .

Environmental correlations ( $r_E$ ) between characters with differences in magnitude and sign, in relation to the respective correlations  $r_G$  revealed that the environment favored a character over another and that the genetic and environmental causes of variation have different physiological

mechanisms, hindering the indirect selection. But for descriptors related significantly with OC, the signals were similar, which accords to Adeyanju et al. (2010) result in concomitant increase in one or more components.

Although the normally used as indicators of association between correlations ( $r_P$ ,  $r_G$  or  $r_E$ ), it does not always demonstrate cause and effect (FALCONER, 1987). In this context, the path analysis is a very useful technique to demonstrate the true correlations of cause and effect (CRUZ et al., 2004). In order for the evaluation of the association between characters to have an estimate and generate a biologically safe and appropriate interpretation, it is of fundamental importance that

the degree of collinearity among the independent variables be tested (COIMBRA et al., 2005).

The  $r_p$  between OC and other agronomic descriptors was split into direct and indirect effects. This analysis initially showed severe collinearity ( $NC > 1000$ ) based on the classification proposed by Montgomery and Peck (2001). In this case, it is advisable for the elimination of inter-related variables (REZENDE, 2007) or to conduct the analysis considering the effects of collinearity (CRUZ et al., 2004; COIMBRA et al., 2005).

Problems relating to multicollinearity were already expected, since they are involved in the analysis descriptors biologically dependent. However, this result does not invalidate the realization of the path analysis, because there are already appropriate methodologies to study the direct and indirect effects on multicollinearity (CARVALHO, 1995; CARVALHO et al., 1999; COIMBRA et al., 2005). Therefore, the

methodology proposed by Carvalho (1995), denominated path analysis in crest for parameter estimation as an alternative form to methodology of the least squares. The constant  $k$  added to the diagonal of the matrix  $X'X$  was determined by examining the trace crest, through the construction of a graph where the path coefficients based on values of  $k$  were plotted in the range of  $0 < k < 1$ .

Table 3 delineates the direct and indirect descriptors reviews about OC with  $k$  value equal to 0.90 effects. Analyzing only the direct effects, there was a positive influence of descriptors PH, SD, NB and MHS on the variable OC. By analyzing the indirect effects, an association of the very same descriptors was found, in other words, their selection can influence directly or indirectly the increase in OC. These results confirm the reports of Li et al. (2004), Ramu et al. (2005) and Adeyanju et al. (2010) that the selection of genotypes with high MHS increase the OC.

**Table 3.** Estimates of the direct and indirect effects of the plant height (PH), stem diameter (SD), number of bunches (NB), number of fruits (NF), mass of fruits (MF) and mass of hundred seeds (MHS) on oil content (OC) of seven castor bean genotypes.

\*Significant by t-test at 5% probability.

Effect	PH	SD	NB	NF	MF	MHS
Direct over OC	0.2258	0.1688	0.2258	-0.0540	-0.1155	0.1334
Indirect through PH	-	0.2000	0.0092	-0.0515	0.1193	0.1280
Indirect through SD	0.1496	-	-0.0523	-0.0650	0.0814	0.5855
Indirect through NB	-0.0062	0.0478	-	0.0109	0.0090	-0.0004
Indirect through NF	0.0123	0.0208	0.0038	-	-0.0275	0.0416
Indirect through MF	-0.0610	-0.0557	0.0067	-0.0588	-	0.00
Indirect through MHS	0.0947	0.1011	0.0003	-0.1029	-0.0028	-
Total (Pearson)	0.6185*	0.6350*	-0.3254	-0.3699	-0.0403	0.5855*
Coefficient of determination = 0.8998						

Coefficient of determination of the path analysis model ( $R^2$ ) was equal to 0.8998, featuring that 89.98% of the variation in the dependent variable (OC) in the crest model, which is explained by the variables used in the causal diagram. The coefficient of determination is an indicator for the analysis of the model adopted. In the case of  $R^2$  close or equal to unity (1), it is accepted that the variations in the dependent variable are explained by variations of the explanatory variables. The magnitude of the coefficient of determination is higher than that found in other oilseed crops such as soybean (TEODORO et al. 2015) and canola (COIMBRA et al., 2005).

Therefore, both the direct and the indirect selection of descriptors PH, SD, NB and MHS are effective in the increase of OC because the direct effects also contributed through indirect ways to increase this parameter.

## CONCLUSION

The direct and indirect selection of genotypes with plant height, stem diameter, number of bunches and mass of hundred seeds is the fastest, most economical and most effective way to select genotypes with high oil content in castor.

**RESUMO:** Com o objetivo de avaliar a correlação e os efeitos diretos e indiretos de caracteres morfológicos sobre o teor de óleo de genótipos de mamona (*Ricinus communis* L.), um experimento foi conduzido nos anos agrícolas 2010/2011 e 2011/2012 na Universidade Estadual de Mato Grosso do Sul, Unidade Universitária de Aquidauana (UEMS/UUA). O delineamento experimental utilizado foi o de blocos casualizados com três repetições. Os tratamentos consistiram de sete genótipos de mamona (BRS Energia, IAC-2028, IAC-Guarani, BRS Nordestina, BRS Paraguaçu, IAC-80 e IAC-226). Foram avaliados os descritores: altura da planta (PH), diâmetro do colmo (SD), número de cachos (NB), número de frutos (NF), massa de frutos (MF), massa de cem sementes (MHS) and teor de óleo (OC). Inicialmente foram determinados os seguintes parâmetros genéticos: variância genotípica, variância ambiental, coeficiente de variância experimental, coeficiente de variação genotípica, herdabilidade, correlações ambientais, correlações fenotípicas e correlações genotípicas. As correlações fenotípicas foram desdobradas pela análise de trilha em estimativas de efeitos diretos e indiretos, e procedeu-se ao diagnóstico da multicolinearidade das variáveis independentes explicativas sobre o OC (variável dependente principal). Os resultados indicam que a seleção direta e indireta de genótipos com altura da planta, diâmetro do colmo, número de cachos e massa de cem sementes são promissoras para se selecionar genótipos com maior teor de óleo em mamona.

**PALAVRAS-CHAVE:** Biodiesel. Melhoramento genético. *Ricinus communis* L. Óleo vegetal.

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