# GENETIC PARAMETERS IN SEED CHARACTERS OF Ormosia discolor UNDER DIFFERENT AMBIENT CONDITIONS

Helinara Lais Vieira Capucho<sup>1\*</sup>, Maria Teresa Gomes Lopes<sup>2</sup>, Manuel de Jesus Vieira Lima Junior<sup>3</sup> Mágno Sávio Ferreira Valente<sup>4</sup> Angela Maria da Silva Mendes<sup>5</sup> Graciela Ines Bolzon Muniz<sup>6</sup>

<sup>1\*</sup>Federal University of Amazonas, Graduate Program in Forestry and Environmental Sciences, Manaus, Amazonas, Brazil –

laisrick21@gmail.com

<sup>2</sup>Federal University of Amazonas, College of Agrarian Science, Department of Animal and Planta Production, Manaus, Amazonas, Brazil – mtglopes@hotmail.com

<sup>3</sup>Federal University of Amazonas, College of Agrarian Science, Department of Forest Science, Manaus, Amazonas, Brazil – mjlimajunior@gmail.com

<sup>4</sup>Federal Institute of Education, Science and Technology of Amazonas, Presidente Figueiredo, Amazonas, Brazil -

<sup>5</sup>Federal University of Amazonas, College of Agrarian Science, Department of Animal and Planta Production, Manaus, Amazonas, Brazil – 45amendes@gmail.com

<sup>6</sup>Federal University of Paraná, Graduate Program in Forestry Engineering – gbmunize@ufpr.br

Received for publication: 21/11/2019 – Accepted for publication: 08/10/2020

#### Resumo

Parâmetros genéticos em caracteres de sementes de Ormosia discolor sob diferentes condições ambientais. Ormosia discolor Spruce ex Benth. é uma árvore nativa da Amazônia, de rápido crescimento, e capacidade de nodulação, as sementes desta espécie são destinadas ao artesanato. O trabalho teve como objetivo estimar parâmetros genéticos em caracteres de sementes de O. discolor sob diferentes condições ambientais e indicar matrizes superiores para compor lotes de sementes. Foram coletadas sementes de 20 progênies selecionadas em uma população no estado do Amazonas. Foram realizados três experimentos em diferentes condições ambientais, no laboratório do Centro de Sementes/UFAM. O ambiente 1 foi realizado em temperatura constante de 30 °C. No ambiente 2, as sementes foram submetidas à temperatura de 35 °C. No ambiente 3, após 5 meses de congelamento, as sementes foram submetidas à temperatura de 30 °C. O delineamento experimental foi o inteiramente casualizado, constituído de 20 progênies, 4 repetições e 25 sementes por parcela. Os caracteres de germinação avaliados foram porcentagem, índice de velocidade, tempo médio e índice de sincronização. Foram estimadas as variâncias e correlações genéticas e fenotípicas, herdabilidade no sentido amplo e o ganho genético por seleção direta. As progênies apresentaram variabilidade genética significativa para a maioria dos caracteres avaliados. Altos valores de herdabilidade e acurácia (>90%) foram encontrados para a porcentagem de germinação em todas as condições ambientais. As matrizes 2, 7 e 16 foram superiores para os caracteres de germinação em todas as condições testadas, sendo indicadas para produção de mudas. Dezoito sementes de O. discolor são suficientes para a realização das análises biométricas com acurácia de 95% na seleção de genótipos. Palavras-chave: espécie nativa, tecnologia de sementes, seleção.

#### Abstract

*Ormosia discolor* Spruce ex Benth. is a native tree of the Amazon with has fast growth and nodulation capacity, whose seeds are used for craftwork. The objective of this study was to estimate genetic parameters in seed characters of *O. discolor* at different ambient temperatures and to indicate superior matrices to compose seed lots. Seeds of 20 progenies selected from a population in the state of Amazonas were collected. Three experiments were performed under different ambient temperatures at the Seed Center laboratory/UFAM. Experiment 1 was performed at constant temperature of 30 °C. In experiment 2, seeds were submitted to a temperature of 35 °C. In experiment 3, after five months of freezing, the seeds were submitted to a temperature of 30 °C. The experimental design was completely randomized, consisting of 20 progenies, four replicates, and 25 seeds per plot. The germination characters evaluated were germination rate, mean germination time, and synchronization and speed germination indexes. Genetic and phenotypic variances and correlations, broadsense heritability, and genetic gain through direct selection were estimated. *O.discolor* progenies showed significant genetic variability for most of the characters evaluated. High heritability and accuracy values (> 90%) were found for germination rates at all ambient temperatures. Matrices 2, 7, and 16 were superior for gered sere sufficient for biometric analysis with accuracy of 95% in genotype selection.

*Keywords*: native species, seed technology, selection.

### INTRODUCTION

*O.discolor* Spruce ex Benth. is a pioneer species belonging to the Fabaceae family, popularly known in Portuguese as tento flamenguista or tento vermelho e preto. The species is typical of upland forests and has confirmed occurrence in the north of the state of Amazonas, Brazil (REFLORA, 2018). The tree has rapid growth, reaches approximately 18 meters in height, has leaves with five to seven leaflets, small flowers with 6-8 mm,

dehiscent, leathery fruits, usually with one to two red-colored seeds, or red with irregular black marks (REFLORA, 2018)

Studies reported that the species presents nodulations when grown in a nursery, a desirable characteristic for reforestation in degraded soils (MOREIRA *et al.*, 1992). However, the most common uses of this species in the state of Amazonas are biojewellery making and craftwork because of the vibrant colors of seeds.

Most studies with the species of the genus *O*. Jackson are focused on the validation of methods for overcoming seed dormancy as their tegument is impervious, a hereditary characteristic common among species of the genus (OLIVEIRA *et al.*, 2016). This condition can be overcome with mechanisms of mechanical or acid scarification applied to impervious-tegumented seeds (BASQUEIRA *et al.*, 2011). However, such information is not available in the literature for *O. discolor*, with more information on pre-germinaton treatments that would facilitate seedling production and standardize the germination process of the species being required.

Temperature is among the main factors affecting seed germination, as it acts directly on the rate of water absorption and on the biochemical reactions that determine the process (OLIVEIRA; PEREIRA, 2014). For most seeds in the Amazon biome, the most common temperature range for the standard germination test has been approximately 25 to 30 °C because of the physiological adaptation of the species to the hot climate of the region during the natural process of germination (LIMA JUNIOR, 2016).

In the Amazon, one of the main discussions on climate change regards the negative impacts it may have on forest ecosystems and how natural resources can be regulated and conserved in face of these effects. In the case of the seeds, storage at temperatures below 0 °C has been essential to enable regulation and conservation, provided that the physiological quality of seeds is not reduced under these conditions. Thus, it is necessary to check what are the effects of high temperatures after a period of freezing on the germination of *O. discolor* seeds in order to identify superior matrices that meet the demand for seeds resistant to future environmental conditions.

The genetic quality of seeds is very important regarding selection, considering that seedlings formed from them reflect on the ability to originate superior types (VECHIATO; PARISI, 2013). Rapid and uniform germination are desirable characteristics to form orchards with superior genotypes of *O. discolor*. Thus, studies that identify the genotypic variability of native species, besides adding information involving the characterization and inheritance of germination, should be prioritized for defining selection criteria and parents for crossbreeding aimed to obtain superior genotypes (CRUZ *et al.*, 2014).

The obtaining of genetic and phenotypic parameters in progenies, such as genetic variance, coefficient of genetic variation, heritability, gains with selection, and correlations between characters, allows the selection of the most appropriate methods of genetic improvement for the species, also allowing inferences on the variability of the characteristic studied (CRUZ *et al.*, 2014).

Studies of genetic parameters of the Amazonian tree species *O. excelsa* in seed germination characters were essential for the selection of superior genotypes (TOMAZ *et al.*, 2018). For the *O. discolor* species, this information is still inexistent, being crucial to obtain seedlings with genetic and physiological quality (TOMAZ *et al.*, 2018; VECHIATO; PARISI, 2013), besides establishing guidelines and protocols for the selection of superior matrices to form commercial seed lots and assist in the domestication and genetic conservation of the species.

Therefore, this study estimates the genetic parameters in seed characters of *O. discolor* under different environmental conditions and indicates superior matrices for seedling formation for planting.

# MATERIAL AND METHODS

Matrix selection was conducted from a native population of *O. discolor* located in the municipality of Autazes, in the state of Amazonas (3° 34' 49" S, 59° 7' 53" W). Twenty open-pollinated trees were inventoried, equidistant at least 100 meters apart. For each individual, the sufficient amount of fruit to conduct tests was collected. In the field, pre-processing was conducted to remove the seeds trapped by the funiculus of the fruit, which is dry and dehiscent. The seeds were processed, stored in lots, and analyzed at the Amazonas Native Seed Center (ANSC). Processing consisted of cleaning and selecting the seeds, which were immersed in distilled water for 24 hours, dried naturally, and stored in sealed plastic bags, identified according to the matrix of origin and stored in a cold chamber at 18 °C.

Germination tests were performed in BOD chambers with 20-W fluorescent lamps, temperature variation not exceeding 2 °C for a period of 24 hours (BRASIL, 2013), and photoperiod of 12 hours per day, which corresponds to the light period at the highest temperature. Germination was tested at two constant temperatures of 30 °C (Environment 1) and 35 °C (Environment 2) and at 30 °C after seeds underwent a period of five months submitted to freezing below 0 °C (Environment 3). Each experiment was installed in a completely randomized design formed by 20 half-sib progenies, four replicates, and 25 seeds per plot, totaling 100 seeds.

For having impervious tegument, all seeds were subjected to pre-germination through mechanical scarification, which consisted of making a cut on the opposite side of the hilum in order to obtain uniform

germination. Seed asepsis was also performed with 5% sodium hypochlorite solution (NaClO) and 95% distilled water for five minutes in order to avoid any external contamination before the tests (BRASIL, 2013).

Evaluations were conducted daily for up to 27 days analyzing the two following germination criteria: radicle protusion (2 mm) and normal seedling development, with developed root and shoot. In order determine germination rate (GR), seeds that emitted primary root were considered germinated, with their value corresponding to the percentage of seeds germinated until the end of the experiment, on the 27th day after sowing. The results were expressed as mean percentage based on the number of normal seedlings (BRASIL, 2013). According to Tomaz *et al.* (2018), the characters mean germination time (MGT), synchronization index (SI), and germination speed index (GSI) were determined.

Biometric characters of seeds were also considered. For this, 30 seeds of each matrix were measured with a caliper based on their length (mm), width (mm), and thickness (mm). The wet weight of seeds was weighted on a precision scale (0.001g) and water content (%) was determined by drying seeds in an oven at 105 °C  $\pm$  3 °C for 24 hours (BRASIL, 2013).

The data obtained for the characters evaluated were submitted to the Lilliefors normality test to verify the need for transformation. Only GP values did not show normality and were transformed according to the following equation:

arcsine 
$$\sqrt{\frac{x}{100}}$$

Where x corresponds to the percentage of seeds germinated.

The genetic parameters estimated were heritability, accuracy, coefficient of genetic and environmental variation, and the ratio between them. Phenotypic and genotypic correlations were also estimated according to Cruz *et al.* (2014). The best progenies were selected based on direct selection using the Mulamba and Mock index (1978), with the same economic weight for all characters except for GP, with heavier weight for being the most important character in tests of this nature. The selection pressure used was 33.33%.

For the biometric characters of seeds, the repeatability coefficient was estimated based on principal components and phenotypic variance and covariance matrix, besides the minimum number of measurements required to predict the real value of the genotypes based on a 95% coefficient of determination. All statistical analyzes were obtained using the Genes software (CRUZ, 2013).

#### RESULTS

The mean moisture content of seeds of *O. discolor* was 8.92%. Germination, according to the radicle protrusion criterion, was similar for all environments, beginning on the  $3^{rd}$  to  $4^{th}$  day. Similarly, seedling formation started on the  $18^{th}$  to the  $19^{th}$  day after sowing in the three conditions proposed (Figure 1).

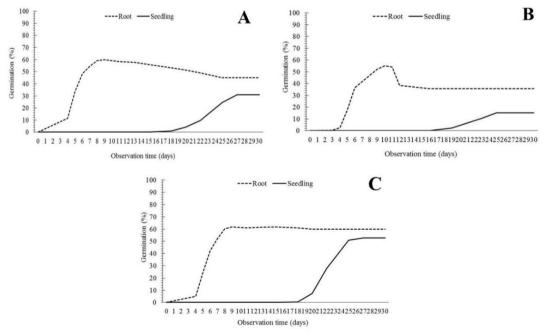


Figure 1. Mean germination rate (%) submitted at ambient temperatures of 30 °C (A), 35 °C (B), and 30 °C with freezing test (C) of 20 *Ormosia discolor* progenies.

Figura 1. Porcentagem média de germinação (%) submetida aos ambientes com temperaturas de 30 °C (A), 35 °C (B) e 30 °C com teste de resistência ao congelamento (C) de 20 progênies de *Ormosia discolor*.

The highest GR was observed in the environment at 30 °C with freezing test with 61%, followed by the environment at 30 °C with 60%, and the environment at 35 °C, with maximum of 55% of seeds germinated on the  $15^{\text{th}}$ ,  $9^{\text{th}}$ , and  $10^{\text{th}}$  day after sowing, respectively.

Significant effect (p <0.01 or p <0.05) of genotypes was observed in the individual analysis of variance for the three ambient temperatures, demonstrating genetic variability between progenies for most of the germination characters evaluated (Table 1).

Regardless of ambient temperatures, the seeds were attacked by fungi (*Aspergillus* spp., *Penicillium* spp., *Trichoderma* sp., *Colletotrichum* spp., *Lasiodiplodia* spp., and *Rizhopus* spp.), which were responsible for the low seed germination and survival rates until the process of seedling formation. The seeds submitted to the temperature of 35 °C obtained the highest mortality.

The lowest GSI was observed at the ambient temperature of 35 °C, with a value of 2.41 and a MGT of 7.15 days. The highest GSI, 2.87, and the lowest TMG, 5.57 days, were observed at the ambient temperature of 30 °C. The lowest SI, representing greater synchrony in germination (RICKLI *et al.*, 2014) was observed at 35 °C (1.56), followed by the ambient temperature of 30 °C (1.68). However, the highest temperature also had the highest coefficient of variation, indicating that this characteristic may not be very accurate.

- Table 1. Mean squares of genotypes (MS) and genetic parameters for germination rate (GR), mean germination time (MGT), synchronization index (ISG), and germination speed index (GSI), of seeds of *Ormosia discolor* submitted to ambient temperatures of 30 °C, 35 °C, and 30 °C (R) with freeze resistance test.
- Tabela 1. Quadrado médio de genótipos (QMG) e parâmetros genéticos para a porcentagem de germinação (PG), tempo médio de germinação (TMG), índice de sincronização de germinação (ISG) e índice de velocidade de germinação (IVG) de sementes de *Ormosia discolor* submetidas aos ambientes com temperaturas de 30 °C, 35 °C e 30 °C (R) com teste de resistência ao congelamento.

Characters	Ambient temperatures	MS	Overall Mean <sup>1</sup>	Mean <sup>1</sup> Genetic parameters <sup>2</sup>		eters <sup>2</sup>		
Characters	Ambient temperatures	MS	Overall Mean	$h_{pm}^{2}(\%)$	Acc	$CV_g(\%)$	$CV_o(\%)$	CVr
	30 °C	0.246**	0.72(45.15)	92.59	96.22	33.03	18.68	1.768
GR	30 °C (R)	0.187**	0.89 (59.90)	92.54	96.20	23.23	13.19	1.761
	35 °C	0.282**	0.61 (35.65)	92.35	96.10	41.88	24.10	1.738
	30 °C	2.509**	5.57	90.19	94.97	13.51	8.91	1.516
MGT	30 °C (R)	2.059**	6.36	83.71	91.49	10.32	9.11	1.133
	35 °C	1.977*	7.15	51.60	71.83	7.06	13.68	0.516
	30 °C	0.448**	1.68	78.76	88.75	17.70	18.39	0.963
SI	30 °C (R)	0.220**	1.98	73.69	85.84	10.18	12.17	0.837
	35 °C	0.249 <sup>ns</sup>	1.56	28.53	53.41	8.56	27.11	0.316
	30 °C	2.508**	2.87	88.07	93.84	25.85	19.03	1.358
GSI	30 °C (R)	2.400**	2.75	91.75	95.78	26.96	16.17	1.667
	35 °C	2.382**	2.41	86.03	92.75	29.70	23.94	1.241

Legend: <sup>ns; \*\*: \*</sup> not significant, p< 0.01, p< 0.05, respectively, by F test. <sup>1</sup>Means of transformed GR data (root arcsine x/100) and original data by the F test;  ${}^{2}\hat{h}_{pm}^{2}$  mean heritability of progenies; *Acc* = accuracy of progeny selection;  $CV_{g}$  = genetic coefficient of variation between progenies;  $CV_{e}$  = experimental coefficient of variation;  $CV_{r} = CV_{g}/CV_{e}$ : Ratio between genetic coefficient of variation and experimental coefficient of variation.

Mean values of the estimate of heritability for the ambient temperature of 30 °C ranged between 78.76 to 92.59%, while at the ambient temperature of 30 °C with freeze resistance test mean values ranged from 73.69 to 92.54% and at the ambient temperature of 35 °C between 28.53 to 92.35% (Table 1). Accuracy, considering all ambient temperatures observed, ranged from 53.41% to 96.22%.

In the joint analysis of variance, it was observed that mean seed germination differed statistically in the three ambient temperatures proposed (Table 2). In addition, significance was observed for the genotype-ambient interaction, showing that the temperatures and ambient conditions tested affected the performance of the characters studied differently. Therefore, inferences were made individually on the genotypes for each ambient.

Table 2. Mean squares of genotype (MSG), ambient (MSA), genotype and ambient interaction (MSAG) and genetic parameters for germination rate (GR), mean germination time (MGT), synchronization index

(SI), and germination speed index (GSI) of *Ormosia discolor* seeds submitted to environments with temperatures of 30 °C, 35 °C, and 30 °C (R) with freeze resistance test.

Tabela 2. Quadrados médios de genótipo (QMG), de ambiente (QMA) e da interação genótipo e ambiente (QMGA), e parâmetros genéticos para a porcentagem de germinação (PG), tempo médio de germinação (TMG), índice de sincronização de germinação (ISG) e índice de velocidade de germinação (IVG) de sementes de *Ormosia discolor*, estimados a partir da análise conjunta dos ambientes com temperaturas de 30 °C, 35 °C e 30 °C (R) com teste de resistência ao congelamento.

Chamastana	MSG	MSA	MS	Overall mean <sup>1</sup>		Genetic parameters			
Characters	MSU	MSA	IVIS	Overall mean	$h_{pm}^2$	Acc	$CV_g(\%)$	$CV_e(\%)$	$CV_r$
GR	0.497**	1.648**	0.109**	0.74 (46.90)	78.12	88.39	24.24	18.03	1.345
TMG	2.982 <sup>ns</sup>	50.092**	1.781**	6.36	40.27	63.46	4.97	11.26	0.442
SI	0.267 <sup>ns</sup>	3.781**	0.325**	1.74	00.00	00.00	0.00	00.00	0.000
GSI	5.664**	4.640**	0.813**	2.68	85.64	92.54	23.73	19.64	1.209

Legend: <sup>ns; \*\*: \*</sup> not significant, p< 0.01, p< 0.05, respectively, by F test. <sup>1</sup>Means of transformed GR data (root arcsine x/100) and original data by the F test;  ${}^{2}\hat{h}_{pm}^{2}$  = mean heritability of progenies; Acc = accuracy of progeny selection;  $CV_{g}$  = genetic coefficient of variation between progenies;  $CV_{e}$  = experimental coefficient of variation;  $CV_{r} = CV_{g}/CV_{e}$ : Ratio between genetic coefficient of variation and experimental coefficient of variation.

The only characters that showed significant genetic variability between genotypes were GR and GSI. In the joint analysis, heritability estimates were mostly lower than those obtained in individual analyzes.

Regarding the association between germination characters (Table 3), at the ambient temperature of 30 °C the greatest genetic correlations were estimated between GR and GSI (r = 0.822) and between the characters TMG and ISG (r = 0.801). At the ambient temperature of 30 °C with freeze resistance test, higher values were observed between GR and GSI (r = 0.869), followed by MGT and SI (r = 0.785), correlated positively to selection. For the ambient temperature of 35 °C, GR and GSI (r = 0.710), MGT and SI (r = -0.959), and MGT and GSI (-0.991) were significantly correlated. Among all significant correlations, only that obtained between MGT and SI (r = -0.959) in the ambient temperature of 35 °C did not show a favorable result for direct selection, inferring that decreasing the MGT would result in a lower SI. Phenotypic and genotypic coefficients of correlation behaved similarly in the three ambient temperatures.

- Table 3. Phenotypic (rF) and genotypic (rG) correlations between germination rate (GR), mean germination time (MGT), germination synchronization index (SI), and germination speed index (GSI) of seeds of *Ormosia discolor* submitted to ambient temperatures of 30 °C, 35 °C, and 30 °C (R) with freeze resistance test.
- Tabela 3. Correlações fenotípicas (rF) e genotípicas (rG) entre a porcentagem de germinação (PG), tempo médio de germinação (TMG), índice de sincronização de germinação (ISG) e índice de velocidade de germinação (IVG) em sementes de 20 progênies de *Ormosia discolor*, avaliadas em ambientes com temperaturas de 30 °C, 35 °C e 30 °C (R) com teste de resistência ao congelamento.

Chanadana		r <u>Ambient temperature</u> r <u>of 30 °C</u> MGT SI GSI			Am	bient temp		Ambient temperature			
Characters	r				of 30 °C (R) MGT ISG IVG			of 35 °C MGT SI GSI			
GR	F	0.224	0.460*	0.801**	0.206	0.543*	0.859**	-0.159	0.070	0.668**	
UK	G	0.223	0.475	0.822*	0.213	0.619	0.869**	-0.273	0.121	0.710*	
MGT	F		0.757**	-0.135		0.679**	-0.300		-0.605**	-0.648**	
MG1	G		0.801**	-0.186		0.785*	-0.318		-0.959**	-0.991**	
SI	F			0.220			0.179			0.386	
51	G			0.187			0.193			0.563	

Legend: \*\*: \* p < 0.01, p < 0.05, respectively, by t test (phenotypic correlations) and by bootstrap method with 5,000 simulation runs (genotypic correlations).

Selection gains (SG) estimated by direct selection were higher at the ambient temperature of 35 °C for the characters GR (47.3%) and GSI (34.2%), and for MGT (14.5%) and SI (18.8%) at the ambient temperature of 30 °C (Table 4).

Table 4. Genetic gain estimates and identification of progenies selected directly and indirectly for germination rate (GR), mean germination time (MGT), germination synchronization index (SI) and germination speed

index (GSI) of seeds of *Ormosia discolor* submitted to ambient temperatures of 30 °C, 35 °C, and 30 °C (R) with freeze resistance test.

Tabela 4. Estimativas de ganho genético e identificação das progênies selecionadas por seleção direta e indireta para a porcentagem de germinação (PG), tempo médio de germinação (TMG), índice de sincronização de germinação (ISG) e índice de velocidade de germinação (IVG) em sementes de *Ormosia discolor*, avaliadas em ambientes com temperaturas de 30 °C, 35 °C e 30 °C (R) com teste de resistência ao congelamento.

Characters	Ambient temperatures	SG (%)	Indire	ct selec	tion ga	in (%)	Salastad progenies
Characters	Ambient temperatures	Direct selection	GR	MGT	SI	GSI	Selected progenies
	30 °C	32.44		-3.32	5.67	22.9	7 16 2 12 11
GR	30 °C (R)	24.96		6.15	4.71	19.53	11 7 2 19 15
	35 °C	47.31		0.44	0.25	17.33	16 7 2 1 11
	30 °C	-14.48	-6.45		-11.9	5.86	2 20 19 9 17
MGT	30 °C (R)	-9.98	1.28		-4.95	12.59	9 16 18 8 2
	35 °C	-5.04	20.32		2.84	23.05	12 13 16 20 11
	30 °C	-18.84	-29.77	-7.25		-21.93	20 15 18 9 14
SI	30 °C (R)	-11.21	-25.53	-3.77		-22.38	8 3 1 10 5
	35 °C	-4.63	0.57	5.74		-19.74	8 7 20 10 1
	30 °C	28.17	26.97	-1.66	-0.82		16 7 6 12 9
GSI	30 °C (R)	31.02	20.4	-6.71	-1.37		2 19 9 16 7
	35 °C	34.20	27.19	-4.66	3.42		16 12 19 13 2

Selection gains estimated by direct selection indicated the five best progenies based on a selection pressure of 33.33% for each seed character and temperature analyzed, in which progenies 2, 7, 9 and 16 stood out for being common to most characters and ambient temperatures, being indicated in at least five of the 12 scenarios evaluated. Another criterion considered to select the progeny with the best performance was obtained from the combined results of all germination characters. For this, the Mulamba and Mock (1978) selection index was used based on the sum of ranks (Table 5). Through this methodology, for optimal germination conditions, progenies 2, 7, 9, 12, and 16 were selected as superior at the temperature of 30 °C. Progenies 2, 7, 9, 16, and 19 were selected at the ambient temperature of 30 °C with freeze resistance test and progenies 2, 7, 11, 13, and 16 were selected at the ambient temperature of 35 °C. It can be noted that progenies 2, 7, and 16 had superior performances in the three ambient temperatures tested, being indicated for planting.

- Table 5. Genetic gain estimates and identification of progenies selected by Mulamba and Mock (1978) index for germination rate (GR), mean germination time (MGT), germination synchronization index (SI), and germination speed index (GSI) in seeds of *Ormosia discolor* submitted to ambient temperatures of 30 °C, 35 °C, and 30 °C (R) with freeze resistance test.
- Tabela 5. Estimativas de ganho genético e identificação das progênies selecionadas pelo índice de Mulamba e Mock (1978) para a porcentagem de germinação (PG), tempo médio de germinação (TMG), índice de sincronização de germinação (ISG) e índice de velocidade de germinação (IVG) em sementes de *Ormosia discolor*, avaliadas em ambientes com temperaturas de 30 °C, 35 °C e 30 °C (R) com teste de resistência ao congelamento.

	-			Coincidence (%)
Ambient temperatures	Characters	SG (%)	Selected progenies	Directly selected and Selection
				index
	GR	30.88		80.00
30 °C	MGT	-5.41	16 2 7 12 9	40.00
50 C	SI	1.55		20.00
	GSI	25.72		80.00
	GR	20.4	2 19 16 9 7	60.00
30 °C	MGT	-6.71		60.00
(R)	SI	-1.37		0.00
	GSI	31.02		100.00
	GR	44.77		80.00
25.00	MGT	-2.48	0 16 7 12 11	60.00
35 °C	SI	1.27	2 16 7 13 11	20.00
	GSI	23.52		60.00

The biometric characters of seeds of *O. discolor* progenies showed significant genetic variability for the characters fresh weight, length, width, and thickness (Table 6).

- Table 6. Mean squares of genotypes (MSG), overall mean, experimental coefficient of variation (CVe), repeatability estimates (r) and coefficient of determination (R<sup>2</sup>) for biometric characters in *Ormosia discolor* seeds.
- Tabela 6. Quadrado médio de genótipos (QMG), média geral, coeficiente de variação experimental (CVe) e estimativas de repetibilidade (r) e coeficiente de determinação (R<sup>2</sup>) para caracteres biométricos em sementes de *Ormosia discolor*.

Biometric	MSG	Overall Mean	$CV_e$ (%)	Repeatability via principal components analysis and covariance matrix			
Characters				r	$\mathbb{R}^2$	No. of evaluations for $R^2 = 0.95$	
Fresh weight	0.041**	0.2	15.66	0.586	0.977	13	
Length	11.955**	7.7	5.41	0.705	0.986	8	
Width	9.730**	7.6	5.36	0.669	0.984	9	
Thickness	3.451**	5.5	6.07	0.517	0.969	18	

Legend: \*\*; \* p < 0.01, p < 0.05, respectively, by F test.

Fresh weight ranged between 0.29 to 0.17 g (mean of 0.2 g), length ranged between 9.3 to 6.3 mm (mean of 7.7 mm), width ranged from 8.4 to 6.6 mm (mean of 7.6 mm), and thickness ranged between 6.1 to 4.9 mm (mean of 5.5 mm), demonstrating that the seeds were not uniformly sized (Table 6). For the strong effects caused by the ambient temperature in biometric data, the experimental coefficients of variation are considered satisfactory for this analysis (TOMAZ *et al.*, 2018; NASCIMENTO JÚNIOR *et al.*, 2016).

Considering the evaluation of biometric characters and progeny selection, which showed superior characters for future generations, the repeatability coefficient was estimated for all biometric characters to quantify and standardize the minimum and reliable number of individual measurements in seeds of *O. discolor* to obtain character accuracy. Thus, through principal component analysis, the coefficient of repeatability was obtained, ranging from 0.517 to 0.705 for the thickness and length of seeds, respectively (Table 6). All coefficients of determination for biometric characters were considered high (>95%), suggesting that the evaluation with 30 seeds per progeny for the four biometric characters was performed with high reliability. However, in order to optimize resources and available time, it is preferable to reduce the number of samples.

The biometric analysis of seeds is performed based on the recommendation of a pre-determined number of seeds not yet standardized for native seeds. In this case, with the objective of a joint evaluation covering all the biometric characters evaluated, selection with accuracy of 95% of superior genotypes of *O. discolor* requires at least 18 seeds.

#### DISCUSSION

*O. discolor* is an impervious-tegumented and orthodox species according to the water content and vigor observed in the seeds during germination tests. These characteristics, commonly found in most of the species that are part of the genus *Ormosia*, are useful for species identification, being known as hard-coated seeds (TOMAZ *et al.*, 2018). Hard-coated seeds are generally orthodox and drying-tolerant, remaining alive with water content of approximately 5 to 7%, withstanding temperatures below zero and remaining viable for many years (GARCIA *et al.*, 2015).

Orthodoxness and tegument dormancy are characters found in most species of Fabaceae. Both are important qualities from an ecological viewpoint, as seeds with these characteristics have a greater chance of survival in nature, remaining viable after dispersion with low water content and being impervious-tegumented when the environment is not favorable to germination, entering the dormancy stage (CARVALHO; NAKAGAWA, 2012).

Although these characteristics are common in *Ormosia* seeds, it is known that species of the same genus can respond in very differently to factors such as water, light, temperature, oxygen, and interaction with microorganisms. Thus, it is necessary to know the optimal conditions for germination in species of interest (CARVALHO; NAKAGAWA, 2012). However, due to the lack of information in the literature on the species *O*. *discolor*, it is difficult to compare the results of this study with other studies.

For the *O. discolor* species, the initial period of germination and seedling formation is within the standards specified for most Fabaceae, between the second and 15th day after planting (CARVALHO; NAKAGAWA, 2012). Similar results were obtained for *Ormosia excelsa* (Fabaceae), in which germination started up to the fourth day after sowing, with seedling formation beginning up to the 19th day after the beginning of the experiment (TOMAZ *et al.*, 2018). For *Stryphnodendron pulcherrimum*, a impervious-tegumented Fabaceae, cutting was used

to break dormancy, similarly to the preparation for *O. discolor*, and germination occurred until the third day after sowing. Fully formed seedlings were observed on the tenth day after sowing, corroborating the results of this study regarding initial germination period in species with tegument dormancy from the break of dormancy (SANTOS *et al.*, 2020).

There is no definition in the literature of the optimal germination temperature for seeds of *O. discolor*, although temperatures from 25 to 30 °C are expected to be ideal for the development of tropical species (LIMA JUNIOR, 2016), as lower temperatures generally provide slower germination. In this study, the lowest germination speed and the longest time were observed for the seeds at the highest temperature (35 °C). Results contrary to this study were obtained by Tomaz *et al.* (2018), as the germination of *O. excelsa* presented lower GSI at ambient temperature of 30 °C and higher at 35 °C. Nascimento Júnior *et al.* (2016) also found that higher temperatures increased the precocity of germination of *Jacaranda copaia*.

The fact that seeds were colonized by fungi is the main justification that helps to explain the behavior of GSI for seeds of *O. discolor* at the highest temperature in this study. Recent studies on germination in seeds of *S. pulcherrimum* also achieved lower germination speeds and rates at controlled ambient temperature of 35 °C, caused mainly by the accelerated deterioration of seeds by endophytic fungi of the genera *Aspergillus* and *Rhizopus*, which indicates that this temperature is one of the ideal factors for rapid leaching of the cellular content of seeds by microorganisms (SANTOS *et al.*, 2020).

Although the two ambient temperatures at 30 °C proposed in this study presented higher germination rate and speed than at 35 °C, germination synchrony, which is a desirable characteristic in tests of this nature, was greater at higher temperatures, although this index has indicated to be imprecise. As for the ambient temperature of 30 °C, other studies with tree species confirmed greater germination synchrony in this temperature range (DUARTE *et al.*, 2015; TOMAZ *et al.*, 2018). High germination rates for *O. discolor* were already expected for the two ambient temperatures at 30 °C, as according to Lima Júnior (2016), constant temperatures between 25 to 30 °C are ideal for approximately 90% of the subtropical forest species evaluated in a study of germination and optimal temperature, among them one species of the genus *Ormosia* (*O. excelsa*).

The heritability of germination characters is an important genetic estimate for selection for ensuring the expression of superior characters from their respective progenitors over generations. This estimate showed wide variability among *O. discolor* progenies. The ambient temperature of 35 °C proved to be less favorable to selection, as heritability values were low for all characters evaluated. The best results were obtained at both ambient temperatures of 30 °C, increasing the possibility of significant gains with production of seedlings of the species. High heritability values (> 80%) for germination characters were also obtained for Amazonian seeds of *S. pulcherrimum, O. excelsa*, and *J. copaia* in similar studies (SANTOS *et al.*, 2020; TOMAZ *et al.*, 2018; NASCIMENTO JÚNIOR *et al.*, 2016). These results are important for selection of seeds with superior characters for the formation of vigorous orchards that meet the needs of reforestation in degraded areas, besides conserving their respective matrices or progenitors. Selective accuracy values obtained for all characters evaluated were also promising, as according to Resende (2007), values close to or above 70% are preferable in the initial and intermediate phases of plant breeding programs.

Joint analysis of variance showed that the genotype-ambient interaction was significant for all characters evaluated. Thus, temperatures had a distinct influence on the expression of germination characters. Heritability was considered moderate and satisfactory in the joint analysis, although contributing with lower values compared to individual analysis. Heritability was null only for the synchronization index, which is explained by the high and approximate values of phenotypic means for the genotypes, resulting in the nullity of the genotypic variance value for this character. This demonstrated that there is a purely environmental influence on the values of this index, which is not considered relevant for selection.

For the presence of high and favorable correlations between germination characters, it is possible to infer that dependence between these characters is a representative result of selection and, for this study, provides adequate information on selection gains, redirecting them to the progenies of *O. discolor* that germinate faster and in less time. It is important to highlight that high and favorable correlations to selection allow satisfactory indirect gains for all other characters, being an efficient procedure for distributing the expected gains adequately (TOMAZ *et al.*, 2018). Other studies with seeds of native species from the Amazon also used genetic correlations to select characters of interest, simplifying the selection process for obtaining the desired gains (SANTOS *et al.*, 2020; NASCIMENTO JÚNIOR *et al.*, 2016).

Breeding programs are designed to achieve high selective gains along with satisfactory genetic variability for the characters of interest. Through direct selection applied to the germination characters, the most expressive results, as expected, were estimated for GR and GSI in the three ambient conditions, as in studies of this nature these two indices are commonly highlighted, demonstrating to be the main variables for selection. In studies by Tomaz *et al.* (2018) and Nascimento Júnior *et al.* (2016), the highest selective gains were also estimated for germination rate, with 17.3% for the species *O. excelsa* and 28.1% for *J. copaia*. These studies are important for

facilitating the selection process by standardizing sets of character and variables with more expressive genetic contributions.

High gains by direct selection must be accompanied by significant genetic variability between the characters evaluated. For this, extensive sampling must be performed during seed collection among populations of interest (TOMAZ *et al.*, 2018). This study recommends five progenies, considering the different ambient conditions and vigor characters evaluated through direct selection and Mulamba and Mock (1978) index, which ensures the efficiency of the phenotypic selection of germination characters for the first generations of genetic improvement of the species. When composing seed lots, these progenies are expected to have speed and uniformity in germination and withstand low temperatures without losing germination vigor, which may be favorable to the conservation and storage of superior seeds of the species.

Significant genetic variability was observed for all biometric characters evaluated in the seeds of *O. discolor.* Studies that discuss biometric characters are important for providing subsidies for morphological differentiation of seeds in species of the same genus, besides being considered fundamental for the study of dispersion and establishment of seedlings and for differentiating species belonging to different ecological groups in tropical forests (BASKIN; BASKIN, 2014). Thus, also knowing that the selection of superior progenies from biometric characters is desirable for the formation of vigorous orchards, the repeatability coefficient was estimated to establish the number of phenotypic observations required for a reliable and efficient biometric evaluation. For each character, the results obtained from 30 seeds showed coefficients of determination greater than 90%, corroborating the studies by Lima Júnior (2016), who observed that 30 seed units are ideal for biometric characterization of seeds.

However, in order to optimize resources and time during evaluations, the number of seeds sampled according to the repeatability estimate can be reduced for this analysis maintaining good accuracy to select superior matrices of the species. Thus, considering the joint evaluation of all biometric characters evaluated, a sample of at least 18 seeds of *O. discolor* with 95% accuracy is necessary. In the evaluations by Santos *et al.* (2020) for the *S. pulcherrimum* species, the study of repeatability from biometric characters was also useful to standardize the ideal sample of seeds for characterizing the seeds of the species, requiring 14 seeds with 80% accuracy. Therefore, knowledge of the repeatability coefficient allows evaluation to be conducted efficiently (RESENDE, 2007). For *O. discolor*, this evaluation is unprecedented and useful for future collection of seeds of the species in the field.

## CONCLUSIONS

Analyzes conducted in this study allow concluding that:

- All characters evaluated in *O. discolor* seeds have significant genetic variability. Ambient temperatures of 30 °C and 30 °C with freeze resistance increased germination rate and decreased mean germination time.
- High heritability and accuracy values suggest that the phenotypic value of germination characters is a reliable parameter for selecting superior genotypes, being more effective when practiced individually in each ambient.
- Progenies 2, 7, and 16 showed good germination characters for all ambient temperatures, being suitable for planting and storage for long periods under low temperature with increased vigor.
- Significant correlations favorable to selection for all ambient were obtained between the characters germination rate and germination speed index.
- Eighteen seeds are sufficient to perform biometric analyzes on *O. discolor*.

## ACKNOWLEDGEMENT

We thank the Research Support Foundation of the State of Amazonas (FAPEAM) and the National Council for Scientific and Technological Development (CNPq) for the financial support to conduct this study.

#### REFERENCES

BASKIN, C. C.; BASKIN, J. M. **Seeds**: ecology, biogeography, and evolution of dormancy and germination. San Diego: Academic Press, 2 ed. 2014, 1586 p.

BASQUEIRA, R. A.; PESSA, H.; LEAL, T. DE. S.; MORAES, C. P. DE. Superação de dormência em *Ormosia arborea* (Fabaceae: Papilionoideae) pela utilização de dois métodos de escarificação mecânica em diferentes pontos do tegumento. **Revista em Agronegócios e Meio Ambiente**, Maringá, v. 4, n. 3, p. 547 - 561, 2011.

BRASIL. Ministério da Agricultura, Pecuária e Abastecimento. **Instruções para análise de sementes de espécies florestais**. Brasília: Secretaria de Defesa Agropecuária, 1 ed. 2013, 98 p.

CARVALHO, N. M.; NAKAGAWA, J. Sementes: ciência, tecnologia e produção. Jaboticabal: FUNEP, 5 ed. 2012, 590 p.

CRUZ, C. D. A software package for analysis in experimental statistics and quantitative genetics. Acta Scientiarum, Maringá, v. 35, n. 3, p. 271 - 276, 2013.

CRUZ, C. D.; CARNEIRO, P. C. S. Modelos biométricos aplicados ao melhoramento genético. Viçosa: UFV, 3 ed. 2014, 668 p.

DUARTE, M. M.; MILANI, J. E. F.; BLUM, C. T.; NOGUEIRA, A. C. Germinação e morfologia de sementes e plântulas de *Albizia edwallii* (Hoehne) Barneby& J. W. Grimes. **Revista Caatinga**, Mossoró, v. 28, n. 3, p. 166 - 173, 2015.

GARCIA, L. C.; SOUSA, S. G. A.; LIMA, R. B. M. Coleta e manejo de sementes florestais da Amazônia. Brasília: Embrapa Amazônia Ocidental, 2 ed. 2015, 33 p.

LIMA JUNIOR, M. J. V. Manejo de Sementes para o cultivo de espécies florestais da Amazônia. São Paulo: Brasil Seikyo Ltda, 1 ed. 2016, 285 p.

MOREIRA, F. M. DE S.; SILVA, M. F. DA; FARIAS, S. M. de. Occurrence of nodulation in legume species in the Amazon region of Brazil. New Phytologist, v. 121, n. 4, p. 563 - 570, 1992.

MULAMBA, N. N.; MOCK, J. J. Improvement of yield potential of the Eto Blanco maize (Zea mays L.) population by breeding for plant traits. **Egyptian Journal of Genetics and Cytology**, Cairo, v. 7, p. 40 - 57, 1978.

NASCIMENTO JÚNIOR, L. G. L.; LOPES, M. T. G.; VALENTE, M. S. F.; MARTINS, C. C.; COLARES, C. R. B.; LIMA JÚNIOR, M. J. V. Estimativa de parâmetros genéticos em sementes de caroba. **Revista de Ciências** Agrárias, Pernambuco, v. 59, n. 4, p. 311 - 319, 2016.

OLIVEIRA, A. K. M.; PEREIRA, K. C. L. Efeito de diferentes temperaturas na germinação e crescimento radicular de sementes de jatobá-mirim (*Guibourtia hymenaefolia* (Moric.) J. Léonard). Ciência Florestal, Santa Maria, v. 24, n. 1, p. 111 - 116, 2014.

OLIVEIRA, A. K. M.; SOUZA, J. S.; CARVALHO, J. M. B.; SOUZA, S. A.; BOCCHESE, R. A. Germinação de sementes e crescimento de *Ormosia arborea* em diferentes temperaturas e substratos. **Gaia Scientia**, Paraíba, v. 10, n. 4, p. 262 - 271, 2016.

REFLORA. **Fabaceae in Flora do Brasil 2020 em construção.** Disponível em: <a href="http://reflora.jbrj.gov.br/reflora/floradobrasil/FB83509">http://reflora.jbrj.gov.br/reflora/floradobrasil/FB83509</a>>. Acesso em: 27 jun. 2018.

RESENDE, M. D. V. Matemática e Estatística na Análise de Experimentos e no melhoramento genético. Colombo: Embrapa Florestas, 1 ed. 2007, 535 p.

RICKLI, H. C.; NOGUEIRA, A. C; KOEHLER H. S.; RIBAS, K. C. Z. Germinação de sementes de *Vochysia bifalcata* em diferentes substratos e temperaturas. **Floresta**, Curitiba, v. 44, n. 4, p. 669 - 676, 2014.

SANTOS, I. N. L.; LOPES, M. T. G.; VALENTE, M. S. F.; LIMA JÚNIOR, M. J. V.; FRAXE, T. J. P. Avaliação genética em sementes de *Stryphnodendron pulcherrimum* sob diferentes níveis de temperatura. **Scientia Forestalis**, Piracicaba, v. 48, n. 125, p. 1-12, 2020.

SILVA, R. G.; CHAVES, M. C. L.; ARNHOLD, E.; CRUZ, C. D. Repetibilidade e correlação fenotípicas de caracteres do fruto de bacuri no estado do Maranhão. Acta Scientiarum Agronomy, v. 31, p. 587 - 591, 2009.

TOMAZ, J. S.; LOPES, M. T. G.; VALENTE, M. S. F.; LIMA JÚNIOR, M. J. V.; MUNIZ, G. I. B.; ROSADO, S. I. P. Genetic evaluation of seed germination and development of seedlings in *Ormosia excelsa* Benth. **Floresta**, Curitiba, v. 48, n. 3, p. 331 - 342, 2018.

VECHIATO, M. H.; PARISI, J. J. D. Importância da qualidade sanitária de sementes florestais na produção de mudas. **O Biológico**, São Paulo, v. 75, n. 1, p. 27 - 32, 2013.