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Thermal-biological aspects of germination of seeds in tropical forest tree species

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Abstract

The present study was carried out with the objective of evaluating the ecological and applied aspects of temperature in the germination of *Colubrina glandulosa* (Rhamnaceae), *Chloroleucon dumosum* (Fabaceae), *Enterolobium contortisiliquum* (Fabaceae), *Mimosa bimucronata* (Fabaceae) and *Sapindus saponaria* (Sapindaceae). Then we assessed germination, average germination time, germination uniformity and germination activation energy as a function of temperatures. The experiment was conducted at the Plant Propagation Laboratory, on the Engineering and Agricultural Sciences Campus, at the Federal University of Alagoas, Rio Largo, AL, Brazil. The experimental design was completely randomized with four replications of 25 seeds per treatment. The data were subjected to analysis of variance and the means were compared using the Tukey test at 5% probability. The isothermal incubation was performed in Biochemical Oxigen Demand (B.O.D.) germination chamber, at constant temperatures of 5, 10, 15, 20, 25, 30, 35 and 40 ºC and alternating at 20-30 ºC. The seeds of *C. glandulosa*, *C. dumosum*, *E. contortisiliquum* and *M. bimucronata* germinated in the range of 10 ºC ≤ T ≤ 35 ºC, and *S. saponaria* germinated in the range of 20 ºC ≤ T ≤ 35 ºC. We found that seeds in the optimal temperature range has unimodal distribution of relative frequency, concentrating germination in the shortest time. The activation energy was positive in the range of 10 ºC ≤ T ≤ 30 ºC, with an inversion of the signal at a temperature of 35 ºC. The studied species had a wide range of temperature tolerance and the speed was curvilinearly dependent on them. The germination process is predominantly endergonic.

Keywords: Caatinga, Activation energy, Atlantic Forest, Northeast.
Abbreviations: BOD_Biochemical Oxygen Demand; SISVAR_System for Analysis of Variance; CECA_Engineering and Agricultural Sciences Campus; UFAL_ Federal University of Alagoas.

Introduction

The Atlantic Forest is a biome representing one of the most complex species diversity on the planet and the Caatinga is the only biome exclusively Brazilian, which means that a large part of its biological heritage cannot be found elsewhere. However, these assets are threatened. Currently, 7.3% of its original cover remains in the Atlantic Forest and about 70% of the Caatinga is already altered by human. This is a consequence of predatory exploitation. Particularly in the coastal and semi-arid region of the Northeast, changes in natural landscapes present risks for several species. These regions are home to distribution centers for several families, with some species widely used in reforestation, such as *Colubrina glandulosa* (Rhamnaceae), *Chloroleucon dumosum* (Fabaceae), *Enterolobium contortisiliquum* (Fabaceae), *Mimosa bimucronata* (Fabaceae) and *Sapindus saponaria* (Sapindaceae), known as colubrina, arapiraca, monkey's ear, maricá and soap dish, respectively (Lorenzi, 2000; Melo Junior et al., 2018; Melo et al., 2018; Neves et al., 2018).

The seeds are the vehicles of the genetic characteristics of a species. Therefore, for the implantation of any culture, this input must be the first concern. The production technology must start with the use of good quality seeds. In assessing the physiological quality of seeds, the germination test is routinely used. However, in the Rules for Seed Analysis (Brazil, 2009) and in the Instructions for Analysis of Seed of Forest Species (Brazil, 2013), which is still being updated, there is a lack of specific information on the germination ecophysiology of the species in question, which still does not have the criteria established for the standardization of the methods of analysis, in view of the production of seedlings.

According to Oliveira et al. (2016) temperature is one of the main environmental factors that governs seed germination, as it strongly influences both the speed of water imbibition by the seed and the biochemical reactions that determine the entire process. In this way, temperature variations affect the speed, percentage and uniformity of germination (Carvalho and Nakagawa, 2012). Each species has a temperature range where germination will occur: temperature or temperature range, considered optimal, where the efficiency of the process is total, and extreme limits of maximum and minimum tolerated by the seeds, above or below, respectively, germinability is not measurable (Bastos et al., 2017). Therefore, it is expected that species with different geographical and ecological distributions produce seeds with variations in terms of thermal requirements for germination. Based on this, the present study was carried out with the objective of evaluating the ecological and applied aspects of temperature in the germination of seeds of *C. glandulosa, C. dumosum, E. contortisiliquum, M. bimucronata* and *S. saponaria*, verifying the germination, the average germination time, germination uniformity and germination activation energy as a function of temperatures.

Results and discussion

Germination rate and germination time
Seeds of *Colubrina glandulosa, Chloroleucon dumosum, Enterolobium contortisiliquum* and *Mimosa bimucronata* germinated in the range of 10 ºC ≤ T ≤ 35 ºC, and Sapindus saponaria seeds germinated in the range of 20 ºC ≤ T ≤ 35 ºC (Table 1). These results probably occurred due to the physiological adaptation of these seeds to the places of occurrence in dry tropical forest areas.

The optimum temperature for seeds of *Colubrina glandulosa, Chloroleucon dumosum, Enterolobium contortisiliquum* and *Sapindus saponaria* was found in the range of 25 ºC <T <30 ºC, and for seeds of *Mimosa bimucronata* in the range of 20 ºC <T <30 ºC , which enabled greater germination (G) in less germination time (TG) (Tables 1 and 2). However, adaptive plasticity dissipation has occurred, since these species also can establish themselves naturally in the Atlantic Forest, Seasonal Semideciduous Forest and Seasonal Deciduous Forest.

Similar results regarding thermal requirements were found for other tree species in the Northeast region of Brazil: *Amburana cearensis* (Almeida et al., 2017), *Mimosa tenuiflora* (Benedito et al., 2017), *Senegalia bahiensis* (Lima et al., 2017) and *Mimosa ophthalmocentra* (Nogueira et al., 2017).

It was found that seeds submitted to temperatures, in the optimum range, presented supposed unimodal distribution of the relative frequency, concentrating the G in the lowest TG (Tables 1 and 2).

Germination uniformity and germination activation energy
For seeds of *Colubrina glandulosa, Chloroleucon dumosum, Enterolobium contortisiliquum* and *Sapindus saponaria*, germination uniformity (U) was lower in the range of 20 ºC ≥ T ≥ 35 ºC, and for *Mimosa bimucronata* seeds the U was lower in the range of 15 ºC ≥ T ≥ 35 ºC (Table 3). In turn, in these intervals, there was possibly a polymodal distribution of the relative frequency and a greater degree of spreading of the G along the TG.

This strategy provided efficiency in the establishment of seedlings, because the seeds can produce seedlings that they may find, ideal conditions for their development (Oliveira et al., 2017). The ecological implications for tropical and subtropical areas, especially in summer, can be an advantageous alternative for the establishment of *Colubrina glandulosa, Chloroleucon dumosum, Enterolobium contortisiliquum* and *Mimosa bimucronata*.

For this reason, higher environmental temperatures, such as those expected as a result of global warming, may increase germination reproduction (Sanhueza et al., 2017). In addition, it was observed that the increases in the populations of existing *Sapindus saponaria* would be caused by an improvement in their reproductive performance as a result of regional warming.

The germination activation energy (ΔH) for seeds of *Colubrina glandulosa, Chloroleucon dumosum* and *Enterolobium contortisiliquum*, in the range of 25 ºC ≤ T ≤ 30 ºC, and for *Mimosa bimucronata* seeds, in the range of 20 ºC ≤ T ≤ 30 ºC, was less than 12 Kcal (Table 4). Therefore, the lower the ΔH consumed in the TG, caused more efficiency in seedling development.

ΔH was positive in the range of 10 ºC ≤ T ≤ 30 ºC, with an inversion of the signal at a temperature of 35 ºC (Table 4). Therefore, ΔH in the range of 10 ºC ≤ T ≤ 30 ºC was directed towards the resumption of growth by the embryonic axis (Silva et al., 2017). On the other hand, at a temperature of 35 ºC, the mobilized ΔH was also diverted to any other process not linked to the process of resuming embryo growth (Medeiros et al., 2017).

Material and Methods

Location
The experiment was conducted at the Plant Propagation Laboratory, on the Engineering and Agricultural Sciences Campus, at the Federal University of Alagoas, Rio Largo, AL, Brazil.

Plant materials
To obtain the seeds, ripe fruits were harvested from January to December 2019, from trees belonging to forest fragments located in the state of Alagoas, Brazil located at 09° 10′ 11′′S, 36° 40′ 47′′W and 654 meters above sea level. According to the Köppen climate classification, the climate is BSh type, hot semi-arid (Emperaire, 1984).

The fruits were harvested at the end of the maturation period with aerial scissors with an extension cord and then kept in the shade (shelter protected from the sun and rain) for a few days, to complete the drying process and facilitate the dehiscence of the fruits (Melo Junior et al., 2018).

The isothermal incubation was carried out in a germination chamber of the BOD type, at constant temperatures of 5, 10, 15, 20, 25, 30, 35 and 40 ºC and alternating at 20-30 ºC, with a photoperiod of eight hours, simulated for four daylight-type 20 W fluorescent lamps. The accuracy of the temperature control was within the range of ± 0.5 ºC. To install the germination test, first, the seeds were sterilised by immersion in 70% alcohol for 1 minute, followed by washing in running water (Rios et al., 2016). Subsequently, topping with scissors was used in the region opposite the hilum, with the seeds placed to germinate on two sheets of paper towels moistened with a volume of distilled water equivalent to 2.5 times the weight of dry paper (Brazil, 2009), contained in transparent plastic boxes (11.0 x 11.0 x 3.5 cm).

The germinated seeds were counted daily for 19 days (*C. glandulosa*), 15 days (*C. dumosum*), 15 days (*E. contortisiliquum*), 15 days (*M. bimucronata*) and 45 days (*S. saponaria*), from the test installation. The subsequent addition of waterwas carried out when necessary to ensure that the substrate remained sufficiently moist throughout the entire experiment. At constant temperatures of 5, 10 and 15 ºC, the test was extended for another seven days, transferring the seeds to the ideal temperature.

Variables analyzed
Germinability: gi = (Σki = 1ni / N) x 100 (Carvalho; Santana; Ranal, 2005), ni being the number of seeds germinated in time i and N the total number of seeds placed to germinate.

Table 1. Germination (%) of seeds of tropical tropical tree species, under different temperatures.

Temperature (ºC)	*Colubrina glandulosa*	*Chloroleucon dumosum*	*Enterolobium contortisiliquum*	*Mimosa bimucronata*	*Sapindus saponaria*
10	13 e	8 e	9 f	5 e	1 d
15	31 d	17 d	32 e	33 d	1 d
20	60 c	30 c	77 c	93 b	2 d
25	72 b	73 b	83 b	94 b	54 b
30	98 a	87 a	99 a	99 a	67 a
35	27 d	14 d	48 d	69 c	24 c
20-30	59 c	29 c	76 c	92 b	58 b
Value“F”	492.71**	243.32**	453.25**	409.70**	240.32**
CV (%)	7.85	9.85	8.43	10.01	14.12

Averages followed by the same lower case letter in the column do not differ at 5% probability by the Tukey test.
** Significant at the 1% probability level.

Table 2. Germination time (days) of seeds of tree species from tropical forests, under different temperatures.

Temperature (ºC)	*Colubrina glandulosa*	*Chloroleucon dumosum*	*Enterolobium contortisiliquum*	*Mimosa bimucronata*	*Sapindus saponaria*
10	13 f	14 e	13 e	10 e	30 d
15	11 e	12 d	10 d	8 d	30 d
20	7 d	9 c	8 c	6 b	30 d
25	5 b	7 b	7 b	6 b	15 b
30	4 a	6 a	5 a	4 a	14 a
35	6 c	12 d	8 c	7 c	19 c
20-30	5 b	9 c	7 b	6 b	15 b
Value “F”	1158.58**	724.38**	946.46**	337.18**	359.12**
CV (%)	6.61	8.42	7.33	9.05	13.43

Averages followed by the same lower case letter in the column do not differ at 5% probability by the Tukey test. ** Significant at the 1% probability level.

Table 3. Germination uniformity (bit) of seeds of tree species from tropical forests, under different temperatures.

Temperature (ºC)	*Colubrina glandulosa*	*Chloroleucon dumosum*	*Enterolobium contortisiliquum*	*Mimosa bimucronata*	*Sapindus saponaria*
10	2.5105 f	2.7317 e	2.6216 e	2.5216 e	2.9821 d
15	2.4078 f	2.6208 d	2.4168 d	2.3258 d	2.9822 d
20	2.2147 e	2.1183 c	1.9325 c	1.4502 b	2.9733 d
25	1.4577 b	1.5599 b	1.5588 b	1.3588 b	2.5833 b
30	0.9072 a	0.9978 a	0.9475 a	0.9777 a	1.4155 a
35	2.0733 d	2.6319 d	1.9834 c	2.0834 c	2.7501 c
20-30	1.6325 c	2.1436 c	1.5546 b	1.4766 b	2.5944 b
Value “F”	1352.75**	191.11**	734.53**	27.31**	174.09**
CV (%)	5.47	12.74	9.37	14.16	14.01

Averages followed by the same lower case letter in the column do not differ at 5% probability by the Tukey test.
** Significant at the 1% probability level.

Table 4. Germination activation energy (Kcal) of tree species from tropical forests, under different temperatures.

Temperature (ºC)	*Colubrina glandulosa*	*Chloroleucon dumosum*	*Enterolobium contortisiliquum*	*Mimosa bimucronata*	*Sapindus saponaria*
10	31.18	34.75	32.26	33.31	37.64
15	32.29	34.42	26.13	20.07	37.42
20	17.38	32.30	14.16	9.97	35.53
25	10.07	9.99	9.85	9.64	18.49
30	9.42	9.78	9.47	9.53	15.77
35	-37.24	-37.57	-31.94	-26.65	-38.35

Average germination time: t = Σki = 1 (niti) / Σki = 1ni (Czabator, 1962), Where: ti: time from the beginning of the experiment to the i nth observation (days or hours); ni: number of seeds germinated in time i (number corresponding to i nth observation); k: last day of germination.
Germination uniformity: U = –Σki = 1Filog2Fi ≍ Fi = ni / Σki = 1ni (Labouriau, 1983), with Fi: relative germination frequency; ni: number of seeds germinated in time i (number corresponding to i nth observation); k: last day of germination. Synchrony index: Z = ΣCn1,2 / N ≍ Cn1,2 = ni (ni – 1) / 2; N = Σni (Σni – 1) / 2 (Primack, 1980), where Cn1,2 is the combination of seeds germinated in the ith time and ni the number of seeds germinated in time
From the Arrhenius equation – (–RlnV) / ∂ (1 / T) = ΔH ≠ + RT, the net variation of energy (enthalpy) for germination activation was calculated both in the infra (V1) and in the above range optimal (V2), using the minimum (Tm) and maximum (TM) germination temperatures as parameters (Labouriau and Labouriau, 1997). Thus, in band V1, ΔH ≠ 1 = R.Tm. [T / (T – Tm)], and in band V2, ΔH ≠ 2 = –R.TM. [T / (TM

– T)], the net variation of enthalpy (ΔH ≠) as a function of temperature was represented by the expression (Labouriau, 1978): ΔH ≠ = [RT (θ - T). (Tm + TM)] / [(T - Tm (TM - T)], where θ (harmonic mean of minimum and maximum temperatures) = [(2Tm.TM) / (Tm + TM)], and T is the experimental temperature, following the physiological interpretation of the opposite signs of ΔH ≠ in the infra and supra-optimal germination ranges.

Statistical analysis

The experimental design was completely randomized, with four replications of 25 seeds per treatment. The data were subjected to analysis of variance and the means were compared using the Tukey test at 5% probability. The statistical program used was Sisvar version 5.6 (Ferreira, 2011).

Conclusions

The seeds of the studied species showed a wide range of temperature tolerance, with minimum and maximum limits of 10 and 35 ºC, respectively. The optimum temperature for germination of the analyzed species is 30 ºC, the speed of which depends on the temperature. The germination process is of the species is predominantly endergonic.

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