ISSN: 2230-9926

Available online at http://www.journalijdr.com

IJDR

International Journal of Development Research
Vol. 11, Issue, 08, pp. 49810-49815, August, 2021
https://doi.org/10.37118/ijdr.22763.08.2021

RESEARCH ARTICLE OPEN ACCESS

TO WHAT EXTENT DOES THE SEED MASS AFFECT THE QUALITY OF LIBIDIBIA FERREA (MART. EX TUL.) L. P. QUEIROZSEEDLINGS?

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ARTICLE INFO

Article History:
Received 06th May, 2021
Received in revised form
19th June, 2021
Accepted 03rd July, 2021
Published online 29th August, 2021

Key Words:
Forest seeds, jucá, physiological potential, seed size, seedlings vigor.

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ABSTRACT

Seed mass is a physical parameter that, for many species, is directly related to the physiological potential of the seeds and, consequently, to the quality of the resulting seedling. Thus, our research aimed to investigate to what extent the seed mass can influence the physiological potential of the *L. ferrea* seedlings. The seeds were classified, according to their individual mass, into small (≤ 0.14 g), medium (0.15 ≤ x ≤ 0.19 g), and large (≥ 0.20 g). Initially, the water content and weight of a thousand seeds were determined. The vigor of the seeds was evaluated using the emergence percentage, emergence speed index, collar diameter, height, number of leaves, and dry mass of plants. The collar diameter, height, and number of leaves evaluations were performed monthly. The seed mass presented a direct relation with the physiological potential of the seeds, influencing the growth of the seedlings until the 60 DAS, with superiority of the large seeds (≥ 0.20 g). Thus, the seeds of *L. ferrea* with mass ≥ 0.20 g should be prioritized by nurseries, foresters and local communities to produce seedlings with high physiological potential.

Citation: ***Priscila Cordeiro Souto, João Correia de Araújo Neto, Karolyne Priscila Oliveira dos Santos et al.*** "To what extent does the seed mass affect the quality of Libidibiaferrea (Mart. Ex Tul.) L. P. Queirozseedlings?*", International Journal of Development Research*, 11, (08), 49810-49815.

INTRODUCTION

Libidibiaferrea (Mart. Ex Tul.) L. P. Queiroz var. *ferrea*, previously classified as *Caesalpiniaferrea* Mart. ex Tul., commonly known in Brazil as "jucá or pau-ferro", is a legume with generalized dispersion and low population density (BACCHI *et al*., 1995; CARVALHO *et al*., 1996). It is native to Brazil and distributed mainly in the Northern and Northeastern parts of the country, where it is considered endemic (FREITAS *et al*., 2012). Because it is an endemic species, which is not found naturally in other environments, its exploration should be conducted in a suitable way so that it does not become, in the future, target of extinction (CAVALCANTE *et al*., 2017). That is why it is necessary to preserve this species, which also has economic, environmental, landscape and medicinal functionalities (SANTANA *et al*., 2011).

The physiological potential of the seeds, indicated by germinative capacity and vigor, can be influenced by extrinsic or environmental attributes (light, temperature, water availability, chemical factors, gases, and biotic factors) and factors intrinsic to the seed (morphology and viability) (KERBAUY, 2019). For some species, among them *L. ferrea*, some physical quality parameters, such as seed mass, may cause responses in terms of physiological potential (GARDARIN *et al*., 2016). Many authors state that the mass of the seed usually does not affect germination but only the vigor of the seedling. Therefore, it is believed that the larger seeds, due to their well-formed embryos and the greater accumulation of reserve substances, have better ability to form and establish more vigorous plants (FLORES*et al*., 2014; SILVA, 2015). Research demonstrating the direct relationship between the seed mass and physiological potential has been conducted on seeds of *Brosimumgaudichaudii* Trécul (FARIA, COELHO and FIGUEIREDO, 2013), *Sideroxylonobtusifolium* (Roem. & Schult.) (SILVA, 2015), *Afzeliaquanzensis*Welw

(Mtambalika *et al.,*2014), *Anadenanthera colubrina* (Vell.) Brenan. (BISPO*et al*., 2017), *Amburana cearensis* (Allemão) A. C. Sm. (ALMEIDA *et al*., 2017), *Pochotafendleri* (Seem.) W.S Alverson& M.C. Duarte (SMIDERLE*et al*.,2018). However, studies with *L. ferrea* are scarce, limited only to the effect of seed massongermination, with no informationin the seedlings phase. Thus, this research aimed investigate to what extent the seed mass can influence the physiological potential of the *L. ferrea* seedlings.

MATERIAL AND METHODS

***Experimental Conditions and plant material*:** The research was conducted in the seedling nursery at Engineering and Agrarian Science Campus (CECA) of the Federal Universit by of Alagoas, in Rio Largo, AL, Brazil, from May to November 2018. The *L. ferrea* seeds were obtained from mature brown fruits, harvested from individuals located in forest fragments in the rural area of the municipality of Paranatama in the State of Pernambuco, Brazil, with geographical coordinates 8° 52' 59'' S and 36° 34' 36''W, in December 2017. The seeds were manually extraction through the lateral opening in the pods with the aid of a hammer. After this procedure, intact seeds were visually sorted into three size classes (small, medium, and large). Subsequently, 500 seeds of each predefined class were individually weighed on an analytical balance, with 0.0001 g precision, to obtain the mass intervals of each class (ALVES *et al*., 2005; PAGLIARINI *et al*., 2014). Small seeds were ≤ 0.14 g, medium between 0.15 ≤ x ≤ 0.19 g, and large ≥ 0.20 g (Figures 1a, 1b, and 1c). Then the seeds were packed in *Kraft* paper bags and stored in a dry chamber (18 °C and 50–65 % RH) until the beginning of the experiments.

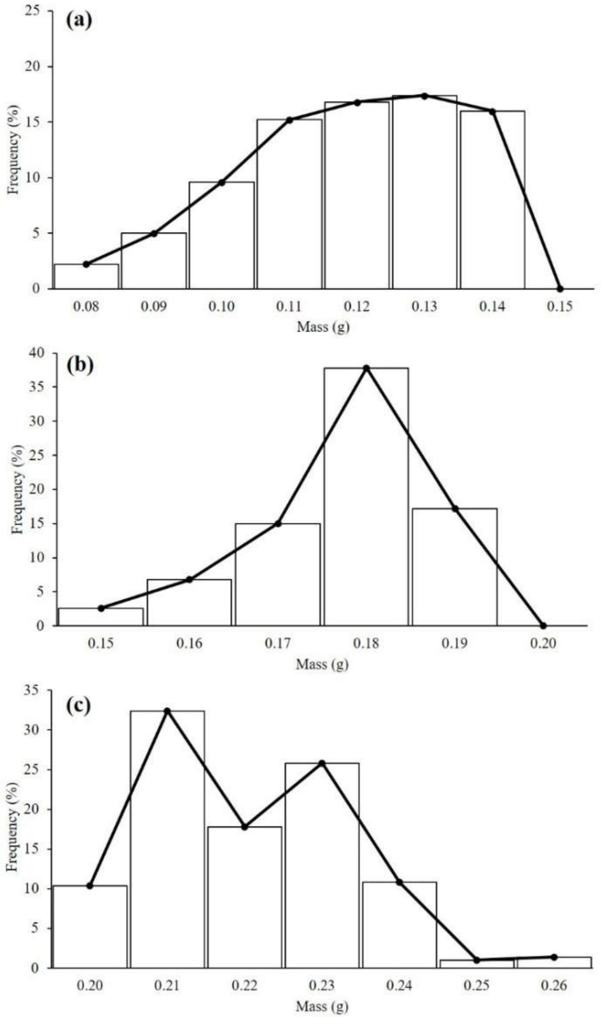

Figure 1. Histogram and frequency polygon of the thickness of small (a), medium (b) and large (c) seeds of *Libidibiaferrea*

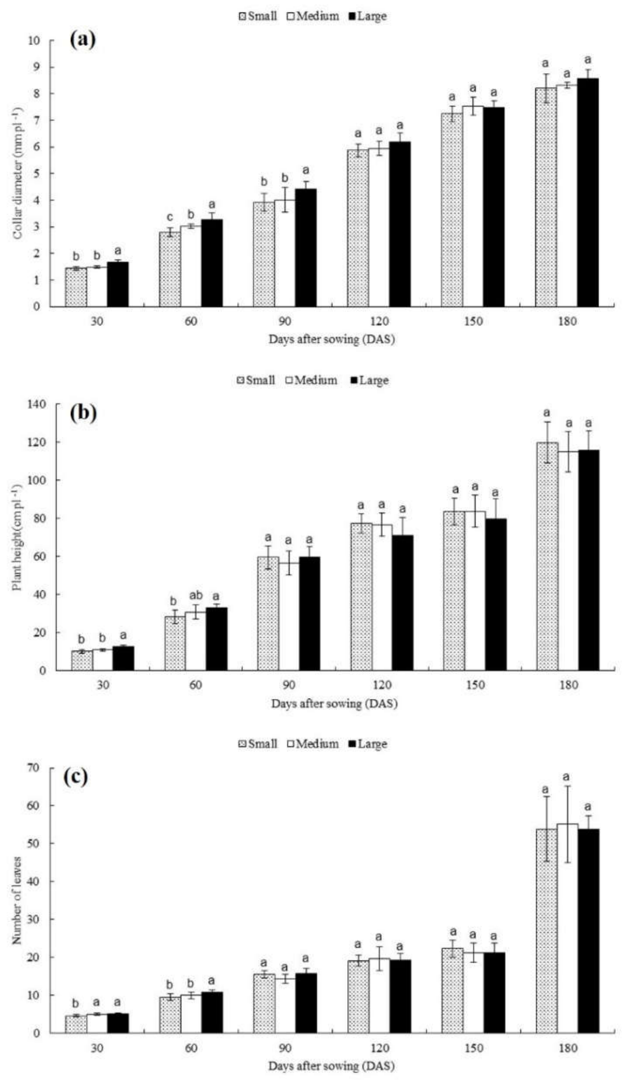

Figure 2. Collar diameter (CD) (a), height (PHT) (b) and number of leaves (NL) (c) of *Libidibiaferrea* plants from seeds with different masses at 30, 60, 90, 120, 150, and 180 DAS. Vertical bars represent standard errors. Different letters indicate significant differences between treatments by the Tukey test (p < 0.05)

***Weight of one thousand seeds (WTS)*:** was carried out with eight sub-samples of 100 seeds of each seed mass (BRASIL, 2009).

***Water content (WC) of seeds*:** was determined by the oven method at 105 ± 3 °C, with four replicates of 10 seeds. The results were expressed as percentage (wet base) for each seed mass (BRASIL, 2009). Dormancy was overcome by subjecting the seeds of each mass to chemical scarification with concentrated sulfuric acid (96% H_2SO_4) for 15 min (ARAÚJO, SILVA and FERRAZ, 2017).

Experimental design: The experiment had a randomized block design with three treatments (small, medium, and large seeds) and eight replications of 10 plants per treatment.

Conduction of the experiment: The seeds of each mass were sown in polyethylene bags (20 × 30 cm) at 1.5 cm deep. Three seeds were sown in each container, totaling 30 seeds per experimental unit and 240 seeds per treatment. The substrate used was obtained from the Engineering and Agrarian Science Campus (CECA) and consisted of a mixture of soil, collected from the topsoil (0–20 cm), and matured cattle manure, both sieved, in the proportion of 2:1 (soil: manure). Thinning was performed 30 days after sowing, leaving only the most vigorous plant per bag. The stem and leaves of the chopped plants were packed separately in *Kraft* paper bags and subjected to drying in a forced air circulation oven at 65 °C for 48 h to obtain the dry mass. The results were expressed in grams/leaves and stem for each mass of seeds. The seedlings were kept in the nursery for 180 days, and during this period, the substrate humidity was maintained with daily watering. Chemical analysis of the substrate (soil and cattle manure) was carried out at the Soil, Water, and Plant Laboratory of the same institution (Table 1). The evaluations were performed at 30, 60, 90, 120, 150, and 180 days after sowing (DAS), considering the following morphological parameters:

Emergence (E %)**:** The emergence of the hypocotyl on the substrate was the emergence criterion for seedlings by 11 days after sowing. The results were expressed in average percentage of seedlings for each mass of seeds.

Emergence speed index (ESI)**:** The number of seedlings emerged from the fifth to the eleventh day after sowing were counted always at the same time of day. The index was calculated according to the formula proposed by Maguire (1962).

Collar diameter (CD)**:** the root collar of the plants was measured with the aid of a digital caliper with 0.01 mm precision, with the values expressed in mm pl^{-1} for each mass of seeds.

Plant height (PHT)**:** a ruler graduated in millimeters was used to measure from the substrate surface to the insertion of the last expanded leaf, and the values were expressed in cm pl^{-1} for each mass of seeds.

Number of leaves (NL)**:** the fully expanded leaves of each plant were counted for each mass of seeds.

Dry mass of leaves (DML), stem (DMS), and roots (DMR)**:** the same methodology was used to obtain the dry mass of leaves and stems as previously explained for 30 DAS. The results were expressed in grams/leaves, stem, and roots for each mass of seeds.

Total dry mass (TDM)**:** the mass of leaves (DML), stem (DMS), and roots (DMR) were added together and the results expressed in g pl^{-1}.

Ratio of dry mass of aerial part and plant root (RDM)**:** The dry mass of the aerial part was determined by adding the dry mass of the stem and leaves together (DML + DMS =DMAP) then this was divided by the dry mass of the roots (DMAP/DMR=RDM) the results expressed in grams.

Ratio of plant height and dry mass of the aerial part (PHT/DMAP)**:** the plant height was divided by the dry mass of the aerial part, and the results expressed in cm g^{-1}.

Ratio of the plant height and root length (PHT/RL)**:** the plant height (PHT) was divided by the root length (RL), with the results expressed in cm.

Ratio of the plant height and collar diameter (PHT/CD)**:** the plant height (PHT) was divided by the collar diameter (CD).

Root percentage (%R)**:** the root dry mass (DMR) was divided by the total dry mass (TDM), and the result multiplied by 100, according to the following formula: (R% = DMR /TDM × 100).

Dickson's quality index (DQI)**:** the formula DQI = TDM/((PHT/CD) + (RDM/DMR)) was used to calculate this formula (DICKSON, LEAF, and HOSNER, 1960).

Statistical analysis**:** Data were analyzed separately for each evaluation period, was subjected to analysis of variance, and the means compared by the Tukey test, at 5% probability.

RESULTS AND DISCUSSION

The weight of a thousand seeds was higher in large seeds, followed by medium and small seeds, with a variability of 89.28g (Table 2). This confirms the biomorphological variation between the seeds of *L. ferrea*. This characteristic, according to Almeida *et al.* (2017), may be a critical factor in competitive environments for the survival and perpetuation of some species. The initial water content of the seeds of each mass was uniform, with approximately 10% moisture (Table 2). The seed mass did not influence the percentage and the emergence speed index of *L. ferrea* seedlings. However, in the evaluation in 30 DAS, superiority was verified regarding the accumulation of dry mass of the leaves and stem in the seedlings from the large seeds, indicating greater vigor of these seeds, intermediate vigor of the medium seeds and low vigor of the small seeds (Table 2). The superiority the largest seeds (≥ 0.20 g) was also confirmed through the collar diameter at 30, 60, and 90 days after sowing, with more evident results at 60 DAS (Figure 2a), confirming the data of dry mass of leaves and stem (Table 2). According to Krishnan *et al.*(2014) and Marcos-Filho (2015) the low physiological potential of small seeds may be due to malformed seeds or late physiological maturity, which result from flowers that were fertilized later, thus requiring them to accelerate the deposition of reserves to reach physiological maturity in the same period as large seeds. These results indicate that the seed mass can affect the physiological potential of *L. ferrea* seedlings. The same finding was also observed in studies by Ristau*et al.* (2018), who analyzed the effects of seed mass on the physiological potential and quality of *Hymenaeacourbaril* L. seedlings. They found that the seed mass affected several variables used to evaluate the physiological potential of the seedlings, such as in collar diameter and dry matter of roots and leaves.

The seed mass affected height (PHT) and number of leaves plants (NL) (Figures 2b and 2c), evaluated at 30 and 60 DAS, always identifying the seeds with mass ≥ 0.20 g as higher. At 30 days, the number of leaves on the seedlings from the medium seeds (0.15 ≤ x ≤ 0.19 g) did not differ statistically from the large seeds. However, by 60 days, seedlings from small and medium seeds had very similar number of leaves. Germination initially consists of soaking, followed by the breaking and mobilization of stored reserves for the formation of new cellular structures, nutrition, and resumption of the growth of the embryonic axis (REGO *et al*., 2011; BEWLEY *et al*., 2013). Hence, initial growth and development of the plants are mainly due to the amount of reserves stored in the seed (TURLEY and CHAPMAN, 2010). Larger and consequently more vigorous seeds, as found in this study (≥ 0.20 g), promote the formation of taller seedlings, with greater collar diameter and more leaves at 30 and 60 DAS. In these periods of seedling formation, the plants were probably still under the effect of the physiological potential of the seeds that they originated from and with low competitive potential due to the resources of the environment.

Some authors have tried to verify a direct relationship between seed mass and seedling growth, for example, Silva *et al*. (2017), evaluating the effect of seed mass on the quality of seedlings of *Euterpeoleracea* Mart., found that plants from heavy seeds (14.2 to 14.6 g) resulted in taller seedlings compared to those from medium (10.85 to 10.98 g) and light seeds (7.6 to 8.0 g). Likewise, Mtambalika *et al*. (2014) working with *Afzeliaquanzensis* Welw, observed that large seeds (mass > 6.0 g and length > 2.5 cm) provided the formation of larger seedlings, whereas medium seedlings (mass between 3.1–5.9 g and length of 1.5 to 2.5 cm) and small (mass of < 3.0 g and length < 1.5 cm), resulted in a slower growth, becoming more susceptible to field conditions (nursery). In seeds of *Artocarpusheterophyllus* Lam., Silva*et al*. (2010) observed a direct relationship between the number of leaves and seed mass. The authors also reported that the average number of leaves was higher in plants from large seeds. However, for *Brosimumgaudichaudii* Trécul seeds, Faria, Coelho, and Figueiredo (2013) did not observe interference of the seed mass on the number of leaves on the plants.

The use of seeds with high physiological potential, associated with edaphoclimatic conditions (temperature, relative humidity, and water) and seed sanity, determine the initial establishment of seedlings, to ensure a uniform stand and with lower production costs (KRZYZANOWSKI, FRANÇA-NETO,and HENNING, 2018).

Table 1. Chemical analysis of the substract

pH	P	K	Na	Ca^{+2}	Mg^{+2}	Al^{+3}	$H^{+} Al^{+3}$	SB	T	T	V	MO
	--mg dm^{-3} --			---------------------- $cmol_c dm^{-3}$ -------------							%	dag kg^{-1}
7.4	75.0	125.0	10.0	8.67	1.70	0.14	1.06	10.73	10.87	11.79	91	2.86

pH in water (1: 2.5); K, P, and Na: $Mehlich^{-1}$ extractor; Ca^{2+}, Mg^{2+}, and Al^{3+}:KCl extractor 1 mol L^{-1}; H + Al: calcium acetate extractor at pH 7.0; SB (Na + K + Ca + Mg): sum of exchangeable bases; T: effective cation-exchange capacity (CEC); at pH 7.0; V: base saturation; MO: Organic matter - Welkley-Black method.

Table 2. Weight of one thousand seeds (WTS), water content (WC), emergence (E), emergence speed index (ESI), dry mass of the stem (DMS), and leaves (DML) at 30 DAS of *Libidibiaferrea* plants from seeds with different masses

Seed Mass							
	Small		Medium		Large		CV %
WTS (g)	127.82	c	173.51	b	217.10	a	1.0
WC (%)	10.31		10.64		10.51		-
E (%)	100.00	a	100.00	a	100.00	a	0.00
ESI	3.99	a	4.03	a	4.13	a	4.90
DMS(g pl^{-1})	0.03	c	0.04	b	0.05	a	7.00
DML (g pl^{-1})	0.07	c	0.10	b	0.11	a	5.10

*Comparison of means within each line (Tukey's test, $p < 0.05$).

Table 3. Dry mass of leaves (DML), stem (DMS), root (DMR), and total (TDM); Ratios of dry mass of the aerial part to dry root(DMAP/DMR), height to dry mass of aerial part (PHT/DMAP), height to collar diameter (PHT/CD),and plant height to root length (PHT/RL); percentage of roots (%R); and Dickson's quality index (DQI) of *Libidibiaferrea* plants from seeds with different masses, at 180 DAS

	Seed Mass						
	Small		Medium		Large		CV %
DML (g pl^{-1})	5.6	a	5.5	a	5.5	a	15.0
DMS (g pl^{-1})	10.2	a	10.2	a	10.8	a	12.3
DMR (g pl^{-1})	4.2	b	4.6	ab	4.9	a	10.9
TDM (g pl^{-1})	20.0	a	20.4	a	21.1	a	9.6
DMAP/DMR (g g^{-1})	3.8	a	3.5	a	3.3	a	9.6
PHT/DMAP (cm g^{-1})	7.8	a	7.3	a	7.2	a	9.0
PHT/DC (cm mm^{-1})	15.0	b	14.0	a	13.6	a	4.9
PHT/RL (cm cm^{-1})	4.7	a	4.4	a	4.2	a	10.3
%R	21.2	a	22.7	a	23.2	a	7.2
DQI	3.8	a	3.5	a	3.3	a	11.6

*Comparison of means within each line (Tukey's test, $p < 0.05$).

Thus, the mass of the seed must be taken into account in the selection of seeds, because mass is a decisive factor in the development and establishment of plants (SULEWSKA *et al.*, 2014), as found in the present study, where large *L. ferrea* seeds (≥ 0.20 g) resulted in higher morphological parameters (collar diameter, height, and number of leaves) in the first months after sowing (Figures 2a, 2b, and 2c).
However, at 120, 150, and 180 DAS (Figure 2a), the seed mass was not reflected in the collar diameter. For the height and number of leaves, this absence was observed earlier, at 90 DAS. With the appearance of additional leaves, the plants begin to photosynthesize and become autotrophic, starting to depend not only on the reserves stored in the endosperm or cotyledons, but on the photosynthate available for their growth (MWASE and MVULA, 2011; MTAMBALIKA *et al.*, 2014). These observations concur with the results of the present study, in which the *L. ferrea* plants from the small seeds (≤ 0.14 g) initially had slow growth, but by 90 DAS, regardless of the initial quantity of the seed reserves, were equalled plants originating from large seeds (≥ 0.20 g) as to height and number of leaves, as well as the collar diameter. Thus, the difference in the plant stand, resulting from seeds with different masses, may decrease in the more advanced stages of the plant growth cycle, which is influence more by the genotype and the environment (EBOFIN *et al.*, 2003; BERNARDI, RAMOS and SILVA, 2019). Mtambalika *et al.*(2014) also stated that seedlings from small seeds, under conditions of high competitiveness for the environmental resources, can match their growth with seedlings from heavier seeds, as long as the edaphoclimatic factors are ideal for their development. This same pattern was reported by Souza and Fagundes (2014) in *Copaiferalangsdorffii* Desf., who observed that seeds with greater mass guaranteed the fast establishment of the plants up to 60 DAS. From there, the authors noticed a reduced effect of seed mass on the variables used in the seedling quality analysis. At 180 days of sowing only dry root mass (DMR) maintained its superiority in seedlings from large seeds. However, the variables: the dry mass of leaves (DML), stems (DMS), and total plant (TDM); as well as the ratios between: dry mass of the aerial part to dry root mass (DMAP/DMR), plant height to dry mass of the aerial part (PHT/DMAP), and plant height to root length (PHT/RL) were similar for different seed masses (Table 3). These results can be attributed to the evaluation time, because in this same period, there were also no differences in the diameter, height, and number of leaves (Figures 2a, 2b, and 2c). Due to the results obtained from the dry mass of the plants, as well as their parts, no differences were found in the percentage of roots and Dickson's quality index, since its calculation is partially dependent on these variables (Table 3).

The diameter of the collar and the height of the aerial part of plants, mentioned above, were considered fundamental morphological parameters to estimate the growth of seedlings. In view of this, the resulting value of the ratio between height and their respective collar diameter defines the growth balance of the plants. Therefore, the joint evaluation of these two parameters generates a robustness quotient (CARNEIRO, 1995). Thus, seedlings from medium (0.15 ≤ x ≤ 0.19 g) and large (≥ 0.20 g) *L. ferrea* seeds had the lowest indexes (PHT/CD) (Table 3), demonstrating that these seedlings have increased physiological potential, because indices with low values indicate that more reserve substances were produced, and therefore, the plant is more resistant, which increases its chances of survival when once permanently transplanted into the field (BIRCHLER *et al.*, 1998). The present study provided relevant information the effect of the individual mass of the *L. ferrea* seeds on the physiological potential of the seedlings. Thus, seedlings from heavier seeds (≥ 0.20 g) exhibited superior quality up to 60 days after sowing, a period that can be decisive in the survival of these seedlings in the field.

CONCLUSIONS

Seed mass is directly related to the physiological potential of *L. ferrea* seedlings. The classification of *L. ferrea* seeds by individual mass is a recommended technique to obtain more vigorous and uniform seedlings. Large seeds (≥ 0.20 g) of *L. ferrea* provide superior quality seedlings up to 60 DAS. Seeds of *L. ferrea* with mass ≥ 0.20 g should be prioritized by nurseries, foresters and local communities to produce seedlings with high physiological potential.

ACKNOWLEDGEMENTS

We thank to the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) and Fundação de Amparo à Pesquisa do Estado de Alagoas (FAPEAL) for the doctorate sponsorship granted to the first author.

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