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Research Article

Dormancy Breaking in *Ormosia arborea* Seeds

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Ormosia arborea is a tree species planted in urban areas and used to restore degraded areas. Its seeds are dormant and propagation is difficult. This study compares different dormancy breaking methods and physiological seed quality and seedling production. The seeds were germinated in sand in the laboratory of the Universidade Estadual de Santa Cruz, Ilhéus, Bahia, Brazil. The following dormancy breaking treatments were applied: control (intact seeds), 100°C water immersion; boiling water immersion followed by 24 hours of soaking; scarification with number 100 and number 50 sandpaper opposite from root emergence; sulfuric acid immersion for 1 hour, 50, 45, and 30 minutes. Seed immersion in 100°C and boiling water did not break the dormancy. The study species showed a greater vigor of seedling when its seeds were submitted to treatments associated with tegument rupturing by sandpaper or sulfuric acid. On the other hand, seed scarification with sulfuric acid for 1 hour, 50, 45, and 30 minutes or sandpaper favored seed germination and vigor.

1. Introduction

Ormosia arborea (Vell.) Harms belongs to the Leguminosae family. It is a 15–20 m tall tree, with a 50–70 cm diameter trunk. It is used in urban tree planting and the restoration of degraded areas because of its leafy canopy [1]. The seeds are dormant due to a hard seed coat that impedes water absorption. Although this is an efficient mechanism that guarantees the survival and perpetuation of the species, it is also a factor that limits propagation.

It is difficult to find information on techniques to accelerate seed germination for the majority of tropical forest species. This information is needed for the cultivation of these species by nurseries and farmers, to restore degraded areas and maintain sufficient genetic diversity.

Dormancy is a relatively important characteristic in the preservation of cultivated species seeds and especially important to maintain seed viability [2]. It is also one of the greatest obstacles for the germplasm conservation of forest species, which frequently produce dormant seeds.

The impermeable properties of the legume seed coat to water or gases and the mechanical restraint of the embryo are achieved by a combination of structural and/or chemical properties, which have been elucidated by anatomical and ultrastructural studies. While the seed coat can be a hindrance to uniform and rapid germination, it nonetheless performs the critical functions of regulating water uptake, providing a barrier to fungal invasion, and reducing leakage from the embryo during imbibition. Reference [3] identified that 85% of the 260 Leguminosae seeds studied had a tegument totally or partially impermeable to water. This type of dormancy can be overcome by scarification, which is any treatment that results in the rupturing or weakening of the tegument, permitting the passage of water and the initiation of germination [4]. However, the nature of the high soil temperature may be responsible for the permeability of the water to coat the seeds of Fabaceae [5]. In nature, the lens may open in response to heating of the soil.

The impermeability of the tegument is usually associated with the presence of one or more impermeable layers of

palisade cells arranged in thick lignified secondary wall, being the most common are macrosclereids cells [6]. The author in [7] recommends that the seeds are scarified *Ormosia arborea* to increase the germination percentage.

Scarification can occur through heating of the soil or by alternating temperatures. This permits the entrance of water to the interior of the seed in natural conditions. This process can also occur through the action of acids during seed digestion by dispersing animals and the action of soil microorganisms [8].

Primary seed dormancy requires high germination temperatures in stratified seeds. The induction of secondary dormancy is caused in early summer by increasing temperatures. This restricts the "germination window" to a short period after snowmelt. Cautious seed regeneration increases the chance of a seedling becoming established at the expense of the number of germinated seeds. This strategy is based on a light requirement for germination and enables both types of species to build up a large seed reservoir in the soil [9].

Various effective and practical treatments have been developed to break this dormancy. Nicking, hot water soaking, and physical or acid scarification have all been used to good effect with the seeds of many tropical and subtropical legume species. With immersion for 20 minutes, germination was reduced to 89% and seedlings were abnormal with reduced growth rates [10]. Immersion in 80°C water and soaking for 24 hours was effective in *Acacia nilotica* and *Tamarindus indica* seeds [11]. In addition, [7] showed that the use of concentrated sulphuric acid for 60 minutes and mechanical scarification increased germination to 34.0% and 31.5% in *Colubrina glandulosa*. Finally, [12] showed that concentrated sulphuric acid for 40 to 60 minutes increased *Sesbania spp.* seed germination. Hard seeds were completely eliminated, and vigor and viability were not affected. On the other hand, those treated with hot water showed a high number of damaged or dead seeds.

Seed dormancy is an innate seed property that defines the environmental conditions in which the seed is able to germinate. It is determined by genetics with a substantial environmental influence which is partially mediated by the plant hormones abscisic acid and gibberellins [13].

Each of these treatments should be studied because of their differing advantages and disadvantages, including cost and practicality [14]. Furthermore, seeds can have differing dormancy levels. The method employed must break the dormancy, without damaging seeds with low dormancy levels. Therefore, the search for forest seeds analysis methodologies plays a fundamental role in scientific research and other diverse interests.

This study compared different dormancy breaking methods in *Ormosia arborea* seeds to facilitate germination and seedling production.

2. Methodology

The seeds were collected for 10 maternal trees in the city of Arataca, Bahia, Brazil. They were stored in the refrigerator at a temperature of 4°C for three months until the initiation of the tests.

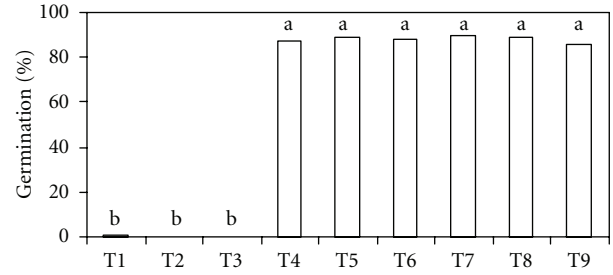

Figure 1: Germination of *Ormosia arborea* seeds submitted to different dormancy breaking treatments. T_1: control; T_2: immersion in 100°C water; T_3: immersion in boiling water followed by 24 h of soaking; T_4 and T_5: scarification with number 100 and number 50 sandpaper, respectively; T_6, T_7, T_8, and T_9: immersion in sulfuric acid for 1 hour, 50 min, 45 min, and 30 min, respectively.

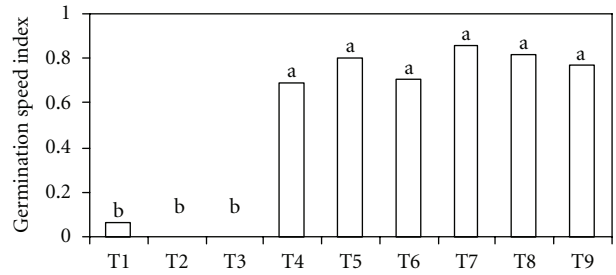

Figure 2: Germination velocity index of *Ormosia arborea* seeds submitted to different dormancy breaking treatments. T_1: control; T_2: immersion in 100°C water; T_3: immersion in boiling water followed by 24 h of soaking; T_4 and T_5: scarification with number 100 and number 50 sandpaper, respectively; T_6, T_7, T_8, and T_9: immersion in sulfuric acid for 1 hour, 50 min, 45 min, and 30 min, respectively.

The experiment was conducted in the Plant Physiology Laboratory of the Universidade Estadual de Santa Cruz (UESC), Ilhéus, Bahia. The following dormancy breaking treatments were applied: control (T_1), seed immersion in 100°C water (T_2) and boiling water followed by 24 hours of soaking (T_3); scarification with number 100 sandpaper (T_4) and number 50 sandpaper (T_5) in the region opposite the root emission; seed immersion in sulfuric acid for 1 hour (T_6), 50 minutes (T_7), 45 min (T_8), and 30 min (T_9).

2.1. Germination Test. The germination test was performed using 100 seeds with five repetitions of 20 seeds for each treatment. These were germinated in aluminum trays (26 cm × 20 cm) with sterilized sand in a 30°C germinator. Counting was initiated on the 16th day after sowing. Seeds were considered as germinated when the cotyledons were present. The average number of normal seedlings was calculated for every repetition and treatment. The test lasted for 45 days.

2.2. Germination Velocity Index. The germination velocity index test was performed together with the germination test.

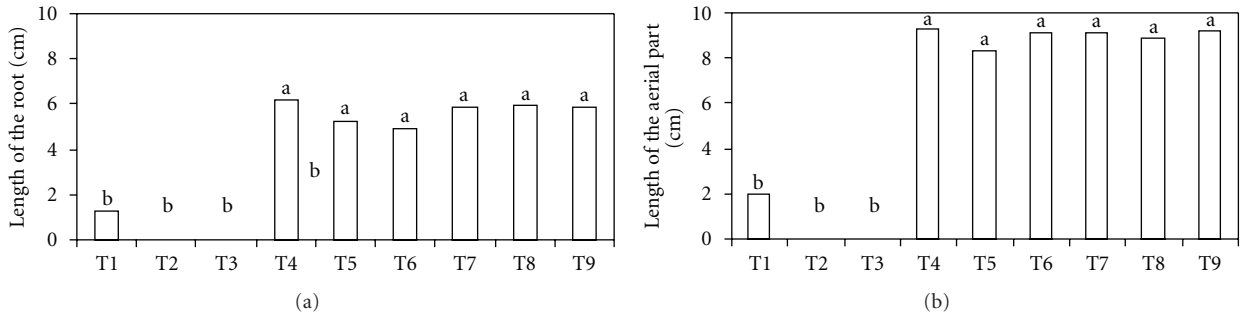

FIGURE 3: Root (a) and aerial part (b) lengths of *Ormosia arborea* seedlings submitted to different dormancy breaking treatments. T_1: Control; T_2: immersion in 100°C water; T_3: immersion in boiling water followed by 24 h of soaking; T_4 and T_5: scarification with number 100 and number 50 sandpaper, respectively; T_6, T_7, T_8, and T_9: immersion in sulfuric acid for 1 hour, 50 min, 45 min, and 30 min, respectively.

The index was calculated according to [], using the formula: $IVG = G_1/N_1 + G_2/N_2 + G_n/N_n$, where G_1, G_2, and G_N are the number of normal seedlings at the first, second, and last counting; N_1, N_2, and N_n are the number of days since the planting to the first, second and last countings. Normal seedling evaluations were realized daily starting from the first germination counting until the last day.

2.3. Seedling Length. At the end of the germination test, the hypocotyl and root of the seedlings were measured. The results were expressed in centimeters per seedling.

2.4. Aerial Part and Root Dry Mass. After the conclusion of the germination test, the cotyledons were removed and normal seedlings were put into a 75°C forced air oven until a constant weight was reached. The seedling parts were then removed, put on polymer water absorption crystals, and weighed using an analytic scale with a precision to 0,0001 g. The data were expressed in g seedling^{-1}.

2.5. Experimental Design. The experiments were conducted using a completely randomized design with nine treatments and five repetitions of 20 seeds. The statistical analysis was done using the ESTAT_UNESP/SP program. The averages were compared with the Tukey test at 1 and 5% probability.

3. Results

The germination test results show the significant effect of the treatments used to overcome seed dormancy (Figure 1).

The treatments using number 100 (T4) and number 50 (T5) sandpaper and immersion in sulfuric acid for 1 hour (T6), 50 min (T7), 45 min (T8), and 30 min (T9) promoted seed germination. There was not a significant difference between these treatments; both sulfuric acid and sandpaper scarification produced satisfactory results.

Seed immersion in 100°C (T2) and boiling water followed by 24 hours of soaking (T3) did not result in germination significantly different from the control. These treatments did not overcome the dormancy of this species.

The study species showed a greater vigor when its seeds were submitted to treatments associated with tegument rupturing by sandpaper or sulfuric acid (Figure 2). There was no significant difference between these treatments.

The greatest root length was observed in the seeds scarified chemically with acid or mechanically with number 50 or 100 sandpaper (Figure 3(a)). The use of organic solvents like alcohol and acetone removes the waxy layer of the seeds of many species.

The same behavior observed in the other tests of this study was shown in the seedling aerial part length. Seeds scarified mechanically with sandpaper and chemically with sulfuric acid resulted in well-developed seedlings (Figure 3(b)). These differed statistically from those that were submitted to heat and soaking.

The root and aerial part dry mass data is presented in Figure 4. Seeds scarified with sandpaper or sulfuric acid had the greater root and aerial part dry mass (Figures 4(a) and 4(b)). On the other hand, the seeds submitted to 100°C or boiling water immersion followed by 24 h of soaking had reduced root and aerial part dry mass that did not differ from the control.

4. Discussion

The species is dormant due to tegument impermeability, which can be overcome by chemical or physical scarification by scarification. The acid and sandpaper treatments used promoted the seed tegument rupturing, facilitating the entrance of water and consequently favoring germination. The rate of absorption of water varies with the species, with the number of pores distributed on the surface of the seed coat, water availability, temperature, contact area of seed/water, chemical and seed quality. Therefore, the imbibition is essentially a physical process related to the characteristics of seed coat permeability and properties of colloids of the seeds, whose hydration is one of its first consequences.

The treatments for dormancy breaking tegumentary seed were efficient because they promoted the rupture of the impermeable layer in the case of physical scarification or

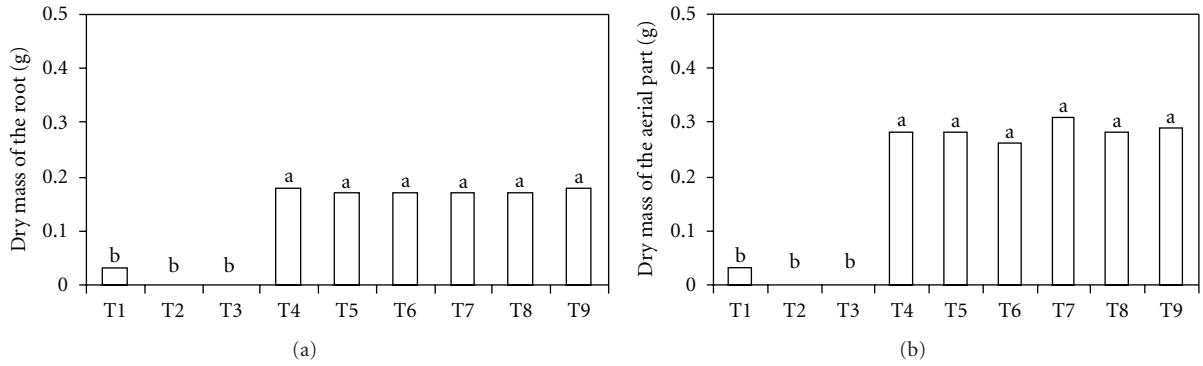

Figure 4: Dry root (a) and aerial part (b) mass of *Ormosia arborea* seedlings submitted to different dormancy breaking treatments. T_1: control; T_2: immersion in 100°C water; T_3: immersion in boiling water followed by 24 h of soaking; T_4 and T_5: scarification with number 100 and number 50 sandpaper, respectively; T_6, T_7, T_8, and T_9: immersion in sulfuric acid for 1 hour, 50 min, 45 min, and 30 min, respectively.

distributing pores in the integument when the sulfuric acid used, thus, enhancing the water absorption by seed and triggering the germination process.

Several studies have been made with the mechanical scarification being shown as an efficient dormancy breaking method to provoke germination in *Astragalus siliquosus* seeds [14]. The same has been observed in *Sesbania spp.* The authors in [12] found that immersion in hot water caused a high quantity of damaged or dead seeds. Seeds of *Bowdichia virgilioides* Kunth scarified for five minutes in sulphuric acid germinated 90% as compared to 21% of the control [15]. The author in [16] observed germination higher than 74% in *Parkia nitida* Miq. seeds scarified for 10, 20, 40, and 80 minutes while the control treatment presented only a germination of 1.5%.

Seedling height at planting is important for survival and initial development. It also contributes to the fresh mass produced. Adequate root development is also necessary and this is related to seed quality, in addition to other factors [17]. The use of hot water, as a treatment for breaking dormancy of this species, is not proved effective.

The seeds have an impermeable seed coat which delays germination forming of seeds on the bank soil.

5. Conclusion

Chemical seed scarification with sulfuric acid for 1 hour, 50, 45, or 30 minutes or mechanical scarification with number 50 or 100 sandpaper was shown as efficient for dormancy breaking in seeds of *Ormosia arborea*.

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