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Evaluation of the effect of pre-germination treatments on Albizia guachapele seeds

Evaluación del efecto de tratamientos pregerminativos en semillas de Albizia guachapele

Avaliação do efeito de tratamentos pré-germinativos em sementes de Albizia guachapele

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ABSTRACT

Albizia guachapele (guachapelí) is a fast-growing tree species native to the tropical dry forests of Ecuador. It is a viable alternative for the reforestation of these ecosystems, which are threatened by urban pressure and deforestation. However, it faces a challenge in seed germination due to physical dormancy. The objective of this research was to evaluate the effect of pre-germination treatments – soaking in water, coconut water, and a gibberellic acid solution – on scarified *A. guachapele* seeds for seedling production in nurseries for reforestation projects. The research was conducted at the Pro-Bosque Foundation's Forest Nursery in Guayaquil, Ecuador. A completely randomized design was used with four treatments: scarification with sandpaper (T1), scarification with sandpaper and soaking in gibberellic acid (T2), soaking in water (T3), and soaking in coconut water (T4). Seed quality and germination percentage were evaluated, as well as seedling height and diameter growth. The results showed that the immersion treatments (T2, T3, T4) significantly increased the germination percentage and reduced the emergence time compared to the control treatment (T1). The gibberellic acid treatment (T2) achieved the highest germination percentage at 88%. No statistically significant differences were found between the immersion treatments. It was concluded that soaking in water after scarification is an economical and effective alternative for the production of guachapelí plants.

Keywords: dormancy, germination, guachapelí, reforestation, viability.

RESUMEN

Albizia guachapele (guachapelí) es una especie forestal de rápido crecimiento nativa de los bosques secos tropicales en Ecuador es una alternativa viable para la reforestación de estos ecosistemas amenazados por la presión urbana y la deforestación, pero enfrenta un desafío en la germinación de sus semillas debido a la dormancia física. El objetivo de la investigación fue evaluar el efecto de tratamientos pregerminativos de inmersión en agua, agua de coco y una solución de ácido giberélico en semillas escarificadas de *A. guachapele*, para la producción de plántulas en viveros para proyectos de reforestación. La investigación se llevó a cabo en el Vivero Forestal de la Fundación Pro-Bosque en



Guayaquil, Ecuador. Se utilizó un diseño completamente al azar con cuatro tratamientos: escarificación con lija (T1), escarificación con lija e inmersión en ácido giberélico (T2), en agua (T3) y en agua de coco (T4). Se evaluó la calidad de las semillas y el porcentaje de germinación, así como el crecimiento en altura y diámetro de las plántulas. Los resultados mostraron que los tratamientos de inmersión (T2, T3, T4) incrementaron significativamente el porcentaje de germinación y redujeron el tiempo de emergencia en comparación con el tratamiento control (T1). El tratamiento con ácido giberélico (T2) obtuvo el mayor porcentaje de germinación con 88%. No se encontraron diferencias estadísticamente significativas entre los tratamientos de inmersión. Se concluyó que el remojo en agua después de la escarificación es una alternativa económica y efectiva para la producción de plantas de guachapelí.

Palabras clave: dormancia, germinación, guachapelí, reforestación, viabilidad.

RESUMO

Albizia guachapele (guachapelí) é uma espécie florestal de rápido crescimento, nativa das florestas tropicais secas do Equador. É uma alternativa viável para o reflorestamento desses ecossistemas, que estão ameaçados pela pressão urbana e pelo desmatamento. No entanto, enfrenta um desafio na germinação das sementes devido à dormência física. O objetivo desta pesquisa foi avaliar o efeito de tratamentos pré-germinativos – imersão em água, água de coco e solução de ácido giberélico – em sementes de *A. guachapele* escarificadas para a produção de mudas em viveiros para projetos de reflorestamento. A pesquisa foi conduzida no Viveiro Florestal da Fundação Pro-Bosque em Guayaquil, Equador. Foi utilizado um delineamento inteiramente casualizado com quatro tratamentos: escarificação com lixa (T1), escarificação com lixa e imersão em ácido giberélico (T2), imersão em água (T3) e imersão em água de coco (T4). Foram avaliadas a qualidade das sementes e a porcentagem de germinação, bem como o crescimento em altura e diâmetro das mudas. Os resultados mostraram que os tratamentos de imersão (T2, T3, T4) aumentaram significativamente a porcentagem de germinação e reduziram o tempo de emergência em comparação com o tratamento controle (T1). O tratamento com ácido giberélico (T2) apresentou a maior porcentagem de germinação, de 88%. Não foram encontradas diferenças estatisticamente significativas entre os tratamentos de



imersão. Concluiu-se que a imersão em água após a escarificação é uma alternativa econômica e eficaz para a produção de mudas de guachapelí.

Palavras-chave: dormência, germinação, guachapelí, reflorestamento, viabilidade.

INTRODUCTION

Modern urban areas consume 67% of the world's energy and generate 71% of CO₂ emissions. According to Castillo-Ruperti *et al.* (2022), this is mainly due to vehicular traffic, the burning of fossil fuels, and the conversion of natural spaces into artificial surfaces. This reality underscores the urgent need to implement strategies to combat the negative effects of climate change, such as reforestation programs.

In this context, biodiversity conservation and the restoration of degraded ecosystems, especially in tropical dry forests, are essential. These forests, a crucial ecosystem for biodiversity, are seriously threatened by deforestation and fires (Hernández Cobos, 2021; Cabrera Verdesoto *et al.*, 2022; Castillo-Ruperti *et al.*, 2022). In Ecuador, dry forests originally covered approximately 35% of the western region of the country (Aguirre, 2012), and according to official figures from the Ministry of Environment, as of 2018, there were an estimated 41,000 hectares of dry forest (Riofrío, 2018).

Within this ecosystem, *Albizia guachapele* (guachapelí), synonym of *Pseudosamanea guachapele* (Plants of the World, 2025), is an emblematic species. Its wood is highly valued for its resistance to seawater, durability, and tolerance to wood-boring insects (Cornejo, 2015). This tree, which can reach between 15 and 25 meters in height, is also ecologically valuable. Its leaves, flowers, and fruits serve as forage, in addition to contributing to atmospheric nitrogen fixation and providing shade in silvopastoral systems (Pérez-Almario *et al.*, 2021; Labre *et al.*, 2022; Castañeda-Garzón *et al.*, 2024).

Given its rapid growth, *A. guachapele* is a viable alternative for reforestation programs. However, its reproduction in nurseries often faces challenges related to seed germination. This difficulty is due to seed dormancy, a phenomenon that prevents germination under favorable conditions, allowing the plant to synchronize with the



optimal environment (Graeber *et al.* 2012). This differs from latency, which is the inability of the seed to germinate due to unfavorable environmental conditions.

In the case of *A. guachapele*, dormancy is physical, a common characteristic in legumes from dry regions (Labre *et al.*, 2022). This is because its seed coat has an impermeable layer that prevents imbibition and gas exchange. To overcome this barrier and ensure successful germination, it is crucial to apply treatments that soften the seed coat without damaging the embryo. The most common pre-germination methods include mechanical scarification (sanding, cutting) or chemical scarification (sulfuric acid), stratification (keeping seeds in moist substrate at low temperatures), and immersion in water (at room temperature or warm) (Labre *et al.*, 2022).

The use of phytohormones such as abscisic acid (ABA), ethylene, and gibberellins is also a strategy to break dormancy and stimulate germination (Miransari and Smith, 2014). Furthermore, coconut water has been used as an effective pre-germination treatment due to its unique composition of sugars, minerals, amino acids, and phytohormones (Yong *et al.*, 2009).

Given the limited specific literature on treatments for guachapelí seeds, the present study evaluated the effect of pre-germination treatments of immersion in water, coconut water and a gibberellic acid solution on scarified seeds of *A. guachapele*, for the production of seedlings in nurseries for reforestation projects.

MATERIALS AND METHODS

Study area

The research was conducted at the Pro-Bosque Foundation's Forest Nursery, located within the Cerro Blanco Protected Forest at kilometer 16 of the Guayaquil-Salinas highway, in the province of Guayas, Ecuador. This forest, covering 6,078 hectares, is one of the last remaining dry forests on the Ecuadorian coast. The area has experienced increasing human pressure due to the population growth of Guayaquil, which rose from



1.7 million in 1989 to 2.7 million according to the 2022 census by the National Institute of Statistics and Censuses (INEC, 2022).

The average weather conditions recorded in the area are: average annual rainfall of 1,650 mm, average annual temperature with a maximum of 31.2 °C and a minimum of 22.8 °C, and an average annual relative humidity of 77% (Figure 1).

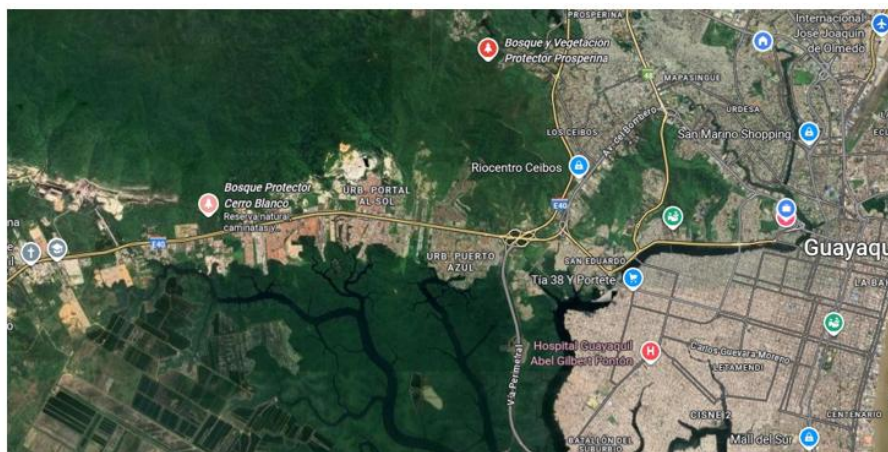


Figure 1 the Pro-Bosque Foundation's forest nursery

Plant production and substrate preparation

The substrate for seedling production was prepared using a mixture of materials available at the nursery, in the following proportions: 25% sand, 25% rice straw (lignocellulosic residue remaining after harvesting rice grain, composed mainly of the plant's chaff), and 50% sifted soil. To prevent fungal diseases such as *damping-off*, the substrate was disinfected with a mixture of systemic and contact fungicides based on carboxamides and phthalimides, respectively. Guachapelí seeds were initially sown in a seedbed protected by a perimeter screen to prevent damage from ants. Once germinated, the seedlings were individually transplanted into 8 cm x 17 cm polyethylene bags, which were labeled according to their corresponding treatment and placed on pallets within the nursery, where all received the same environmental conditions. Weed control was carried out manually, and phytosanitary control was applied when pathogens or pests were detected. Irrigation was done directly with a filter integrated into the nozzle to ensure uniform water distribution.



Seed quality assessment

To determine the quality of the seeds, the following parameters were considered:

Physical purity: Four samples of 10g each were used. The pure seeds were separated from the impurities to calculate the percentage of purity.

Number of seeds per kilogram: Four 10g samples were taken, the number of seeds in each was counted and the average was calculated to estimate the number of seeds per kilogram.

Seed viability: Viability was assessed using the float test. Four replicates were performed, each with 200 seeds. After 24 hours of immersion in water, seeds that sank were considered viable, while those that floated were classified as non-viable.

Experimental design

A completely randomized design (CRD) was implemented to evaluate the effect of four pre-germination treatments on seedling germination and growth in the nursery. The experiment was single-factor, and the following treatments were defined:

Treatment 1 (T1): seed scarification with sandpaper (control treatment).

Treatment 2 (T2): scarification with sandpaper followed by immersion in gibberellic acid for eight hours.

Treatment 3 (T3): scarification with sandpaper followed by immersion in water at room temperature for 24 hours.

Treatment 4 (T4): scarification with sandpaper followed by immersion in coconut water of tender fruits for 24 hours.

For the germination evaluation, 400 seeds were selected, randomly assigning 100 to each treatment. The response variables to the test treatments were the germination percentage (measured 28 days after sowing) and days to emergence (days elapsed until 60% of seedlings germinated).



For growth assessment, 20 seedlings from each treatment, randomly selected from those with the greatest height growth, were transplanted into pots. Height (in cm, from the base of the substrate to the terminal apex) and stem diameter (in mm, measured at the midpoint of the stem) were measured 30 days after transplanting.

Statistical analysis

The statistics of central tendency (average) and dispersion (variance, standard deviation) were calculated for the data obtained from the seed quality and plant growth tests.

To compare the effects of the treatments on germination, the Marascuilo test was used to identify significant differences between pairs of treatments, with a 95% confidence level ($\alpha=0.05$). This analysis was programmed in an Excel spreadsheet.

To evaluate the effect of the treatments on the growth variables (height and diameter), a parametric Analysis of Variance (ANOVA) was performed. Mean comparisons were conducted using Tukey's post-hoc test. The assumptions of normality and homoscedasticity of the residuals were verified using the Shapiro-Wilk and Levene tests, respectively. If the assumptions were not met, the non-parametric Kruskal-Wallis test was used. All tests were applied with a statistical significance level of $\alpha = 0.05$. Statistical calculations and inferential tests were performed using the INFOSTAT statistical package and an Excel spreadsheet. NotebookLM (Google, 2025) and Grok.ia (xAI, 2024) were used for organizing sources and editing text.

RESULTS AND DISCUSSION

Seed quality analysis.

Number of seeds per kilogram

To determine the number of seeds per kilogram, four 10 g samples were analyzed. The average number of seeds was 256.75 (SD = 8.18; CV = 3.19%), which equates to an estimated 25,675 seeds per kilogram, a value within the range of 23,000 to 29,000 seeds per kilogram reported by Noboa (2010). The coefficient of variation (CV) of 3.19% is



considered acceptable, as it is below the 4% limit established by the International Seed Testing Association [ISTA] (2019) standards for this type of analysis, confirming the uniformity of the data.

Physical purity

The physical purity of *Albizia guachapele* seeds was assessed using four 10 g samples. The results show an average weight of pure seeds of 9.205 g (SD = 0.26; CV = 2.83%) and an average weight of impurities of 0.795 g (SD = 0.26; CV = 32.81%). The average purity percentage was 92.05%, indicating a low presence of inert material. This value is slightly lower than the 98% reported by Salazar, Soihet, and Méndez (2000), which could be attributed to differences in the origin, collection site, and seed selection methods.

Seed viability

Seed viability was assessed using a flotation test with four replicates of 200 seeds each. The average number of viable seeds was 182.75 out of 200 seeds (SD = 1.71; CV = 0.93%), for an average viability of 91.4%. This result is consistent with studies by Romero *et al.* (2014), who indicate that the viability of *A. guachapele* seeds can be affected by the presence of coleopteran insects, which impacts their conservation.

Germination percentage

Table 1 presents the germination percentages evaluated 29 days after sowing. Treatment 2 (scarification with sandpaper + immersion in gibberellic acid) achieved the highest percentage at 88%, followed closely by treatment 3 (scarification with sandpaper + immersion in water) at 86% and treatment 4 (scarification with sandpaper + immersion in coconut water) at 85%. In contrast, treatment 1 (scarification with sandpaper) showed the lowest percentage, with only 65% germination.

Table 1- Average germination percentage at 29 days

Treatment	T1	T2	T3	T4
Average %	65	88	86	85



Marascuilo's analysis (Table 2) showed that T1 was significantly inferior to treatments T2, T3, and T4 in its ability to promote germination. However, no statistically significant differences were found among treatments T2, T3, and T4, suggesting that they were all equally effective and superior to the control treatment. This finding is of great practical importance, as soaking in water (T3) is a more economical and accessible method for large-scale nursery production, with results comparable to those of gibberellic acid and coconut water.

Table 2- Comparison of germination proportions by treatment

Comparison	DIFABS	Combined standard error	Critical range	Next**	Chi quad	p-value
p1 vs p2	0.23	0.058	0.161	Yeah	15.88	0.00120
p1 vs p3	0.21	0.059	0.165	Yeah	12.68	0.00539
p1 vs p4	0.20	0.060	0.167	Yeah	11.27	0.01036
p2 vs p3	0.02	0.048	0.133	No	0.18	0.98122
p2 vs p4	0.03	0.048	0.135	No	0.39	0.94310
p3 vs p4	0.01	0.050	0.139	No	0.04	0.99787

Note. Letters p1, p2, p3, and p4 represent the proportions of germinated seeds in each treatment. ABS DIF is the absolute difference between the proportions purchased. Sig. ** indicates statistical significance ($p\text{-value} < .05$).

Days to the emergency

Regarding the time to emergence, treatments T2, T3, and T4 began germination two days after sowing, while T1 began germinating three days later. Treatments T2, T3, and T4 exceeded the 60% germination threshold around 14 days after sowing, a much shorter period than the approximately 28 days it took treatment 1 to reach the same percentage, as shown in Figure 2, where T2 reaches close to 90%, T3 and T4 exceed 80%, and T1 reaches 60% at the end of the germination observation period.



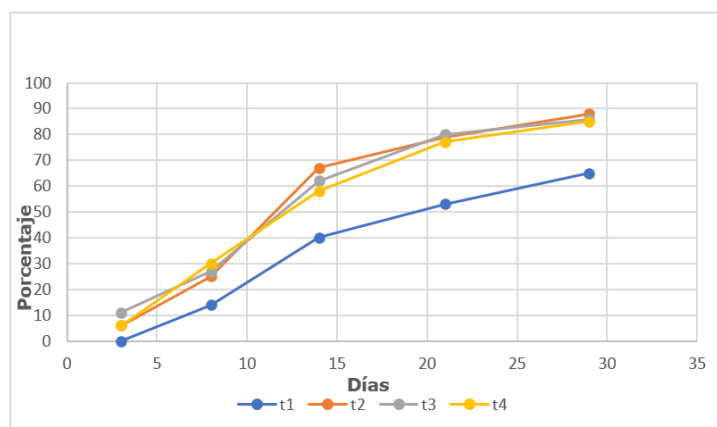


Figure 2- Days of emergency by treatment

These results confirm that scarification combined with immersion in different solutions is more effective at breaking seed dormancy in *A. guachapele*. The germination percentages obtained with treatments T2, T3, and T4 are consistent with previous studies on similar species (Ramírez *et al.*, 2012; Suárez *et al.*, 2014), who also reported high germination rates when applying scarification methods. The effectiveness of gibberellic acid (T2) as a germination promoter is widely documented in the literature (Orantes *et al.*, 2013; González *et al.*, 2008; González *et al.*, 2009), which aligns with the superior results of this treatment in this study.

Effect of pre-germination treatments on plant growth

Descriptive data for growth variables (height and diameter) are presented in Table 3. Treatments T2, T3, and T4 showed higher average heights than treatment T1 (11.28 cm, 10.52 cm, 10.77 cm vs. 9.81 cm, respectively). Differences in diameter were less pronounced, with very similar values among all treatments (0.92 mm to 0.96 mm).

Analysis of variance (ANOVA) for height revealed significant differences between treatments ($F(3, 76) = 18.32, p < .001$). Tukey's post hoc test confirmed that Treatment 1 was significantly different from the others (T2, T3, and T4), while no significant differences were observed among the latter. Although the Shapiro-Wilk normality test indicated that the residuals did not follow a normal distribution ($p < .001$), Levene's test confirmed the homogeneity of variances, a key assumption for the validity of ANOVA. For more robust validation, the non-parametric Kruskal-Wallis analysis of variance was applied to both variables.



Height: The test showed significant differences between treatments ($H(3) = 36.86$, $p < .001$). Treatment 2 (with gibberellic acid) showed the highest average range, confirming it as the most effective in promoting height growth, although it did not differ significantly from the coconut water treatment (T4).

Diameter: The Kruskal-Wallis test found no significant differences ($H(3) = 8.10$, $p = .0409$), suggesting that, at 30 days, the treatments do not have a noticeable differential effect on stem thickness.

These results are consistent with the known function of gibberellins, which, as Taiz and Zeiger (2002) indicate, promote cell elongation and division, resulting in increased height growth. The study's findings are also consistent with those of Jiménez (2014), who reported that pre-germination treatments have a significant and differentiated effect on the height of various forest species.

Table 3. - Descriptive statistics of the growth and significance variables

Variable	Treatment	n	M	OF	ANOVA	Kruskal Wallis
Height (cm)	1	20	9.81	0.49	TO	to
	2	20	11.28	0.88	C	c
	3	20	10.52	0.43	B	b
	4	20	10.77	0.67	b,c	b,c
Diameter (mm)	1	20	0.92	0.05	TO	to
	2	20	0.93	0.07	TO	a,b
	3	20	0.96	0.05	TO	b
	4	20	0.96	0.04	TO	b

NOTE – Means with a common letter are not significantly different, $p > .05$

CONCLUSIONS

The combination of scarification with sandpaper followed by immersion in water, coconut water, or gibberellic acid was highly effective in breaking the physical dormancy of Albizia guachapele seeds, resulting in a significant increase in the



germination percentage and a notable reduction in emergence time compared to scarification alone.

No statistically significant differences were found in the germination and height growth variables between the immersion treatments (gibberellic acid, water, and coconut water).

Immersion treatments promoted greater height growth in seedlings compared to the control treatment (scarification alone), although they did not significantly affect stem diameter at 30 days.

Soaking the seeds in water after scarification is established as an economical and effective alternative for the large-scale production of guachapelí plants in nurseries, since its results were comparable to those obtained with gibberellic acid and coconut water.

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The authors declare no conflicts of interest.

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The authors have participated in the writing of the work and analysis of the documents.



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