

## Articles

### Overcoming dormancy and viability of pecan seeds (*Carya illinoensis*) subjected to different physical methods

Superação de dormência e viabilidade de sementes de noqueira-pecã (*Carya illinoensis*) submetidas a diferentes métodos físicos

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## ABSTRACT

This study evaluated physical treatments for overcoming dormancy in pecan seeds. Treatments combining refrigeration, disinfestation, scarification, and stratification were tested, and significant effects were observed on emergence, emergence speed index, and seedling height. The treatment involving refrigeration followed by scarification and stratification produced the best results; however, overall values remained low when compared with those reported in previous studies. The limited emergence was likely associated with reduced seed viability due to storage conditions. These findings suggest that pecan seed emergence can be improved by combining physical treatments, while also highlighting the need for further studies on storage conditions that preserve seed viability and promote more successful seedling establishment.

**Keywords:** Seed dormancy; Cold stratification; Scarification; Seed viability

## RESUMO

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Este estudo avaliou métodos físicos de superação de dormência em sementes de noqueira-pecã. Foram testados tratamentos combinando refrigeração, desinfestação, escarificação e estratificação, observando diferenças significativas na emergência, índice de velocidade de emergência e altura de plântulas. O tratamento com refrigeração seguida de escarificação e estratificação apresentou os melhores resultados, embora ainda baixos quando comparados a estudos prévios. A reduzida emergência provavelmente está associada à perda de viabilidade causada pelo armazenamento das sementes. Os resultados indicam que a emergência da noqueira-pecã é favorecida pela combinação de estímulos físicos, mas reforçam a necessidade de pesquisas sobre condições adequadas de armazenamento para garantir maior sucesso no estabelecimento de plântulas.

**Palavras-chave:** Dormência de sementes; Estratificação a frio; Escarificação; Viabilidade de sementes

## 1 INTRODUCTION

Pecan (*Carya illinoensis* [Wangenh.] K. Koch), a deciduous tree species belonging to the family Juglandaceae, is native to North America and is cultivated worldwide due to the high commercial value of its nuts, which are used in the food and cosmetic industries (Wang *et al.*, 2025). Commercial propagation is preferably carried out by grafting, which requires vigorous rootstocks capable of improving water and nutrient uptake, tolerance to stress, and adequate support for the canopy (Kaleem *et al.*, 2023). These rootstocks are obtained from seeds, whose quality directly influences seedling production (Casales *et al.*, 2018). However, recalcitrant seeds, such as those of pecan, have high moisture content at maturity and are sensitive to desiccation and low temperatures, which limits long-term storage and compromises viability and vigor (Dambros *et al.*, 2023; Wang *et al.*, 2023).

Proper storage between harvest and sowing is essential to preserve seed viability and vigor, ensuring the production of uniform seedlings. When kept at room temperature, seeds may undergo rapid physiological deterioration, resulting in reduced germination and seedling vigor (Poletto *et al.*, 2015). Although pecan seeds remain metabolically active after maturation and do not exhibit deep dormancy, the hard endocarp may impose physical and mechanical constraints on radicle emergence

(Hong and Ellis, 1996; Liu *et al.*, 2023). Protective coverings may induce physical, chemical, or mechanical dormancy, thereby affecting the speed and uniformity of germination (Lopes *et al.*, 2022; Ren-Fei *et al.*, 2023; Soares *et al.*, 2025). The intensification of these barriers over storage time, especially under uncontrolled conditions, may substantially reduce germination potential and early seedling vigor.

Dormancy breaking is essential to overcome mechanical and physiological barriers that hinder germination (Finch-Savage and Leubner-Metzger, 2006). Physical barriers, high phenolic content, and underdeveloped embryos may be responsible for dormancy in woody seeds, requiring processes beyond simple scarification (Soares *et al.*, 2025). In species with thick seed coats and resistant sutures, such as pecan, germination may be slow and uneven, with limited early seedling development (Wang *et al.*, 2025). Studies have shown that partial mechanical removal of the seed coat in hard-coated species, such as *Cycas revoluta* Thunb., significantly increases germination, highlighting that reducing mechanical resistance can be decisive for improving emergence (Lima and Oliveira, 2024). In pecan, appropriate pre-germinative techniques may compensate for potential physiological losses resulting from storage and enhance the production of high-quality seedlings.

Among the methods most commonly used for seeds with non-deep dormancy are cold or warm stratification and mechanical scarification, which weaken the protective layer and facilitate radicle penetration (Lopes *et al.*, 2022; Ren-Fei *et al.*, 2023; Wang *et al.*, 2025). The choice of technique depends on the type of dormancy and storage conditions, with the aim of balancing germination efficiency and economic feasibility (Xue *et al.*, 2024; Cheng *et al.*, 2022). In pecan, establishing effective protocols for breaking dormancy in seeds stored under natural conditions is essential to ensure uniform and vigorous emergence, thereby supporting rootstock production and, consequently, the successful establishment of new orchards. This study aimed to evaluate the viability of pecan seeds stored under ambient conditions and subjected to different dormancy-breaking methods.

## 2 MATERIALS AND METHODS

Pecan seeds of the Barton cultivar were collected from an orchard at the Federal University of Santa Maria (UFSM), in the state of Rio Grande do Sul, Brazil (29°43'40.99" S; 53°43'27.75" W). After collection, the seeds were dried and stored at room temperature (~25 °C) for four months. To standardize the experiment, only seeds with a mass of approximately 8 g were selected. The trial was conducted in a completely randomized design with four replicates per treatment, each replicate consisting of a tray containing 49 seeds.

Seedling emergence in pecan was evaluated using a dormancy-breaking test with five treatments (Table 1). Disinfestation consisted of immersion in 2.5% sodium hypochlorite for 20 min, followed by rinsing under running water (treatments D, DSS, and CDSS). Scarification was performed with No. 80 sandpaper on the apical region (DSS and CDSS). Cold stratification, a moist treatment for dormancy breaking, was carried out at 4 °C for 150 days in trays filled with autoclaved sand, which had been sterilized at 120 °C for 1 h in three cycles (CS, DSS, and CDSS). In the CDSS treatment, the seeds were first stored dry at 4 °C, packed in plastic bags for 15 days, before the remaining steps were carried out.

Table 1 - Description of the treatments applied to pecan seeds (*C. illinoensis*) for the evaluation of dormancy breaking Treatment description Control, without disinfestation or dormancy breaking

| Treatment description                                                                | Abbreviation |
|--------------------------------------------------------------------------------------|--------------|
| Control, without disinfestation or dormancy breaking                                 | C            |
| Disinfestation only                                                                  | D            |
| Cold stratification (150 days at 4 °C)                                               | CS           |
| Disinfestation + scarification + stratification                                      | DSS          |
| Dry cold storage (15 days at 4 °C) + disinfestation + scarification + stratification | CDSS         |

Source: Authors (2025)

After the treatments, the seeds were sown in trays containing autoclaved sand (three cycles at 120 °C for 30 min each) and maintained in a greenhouse

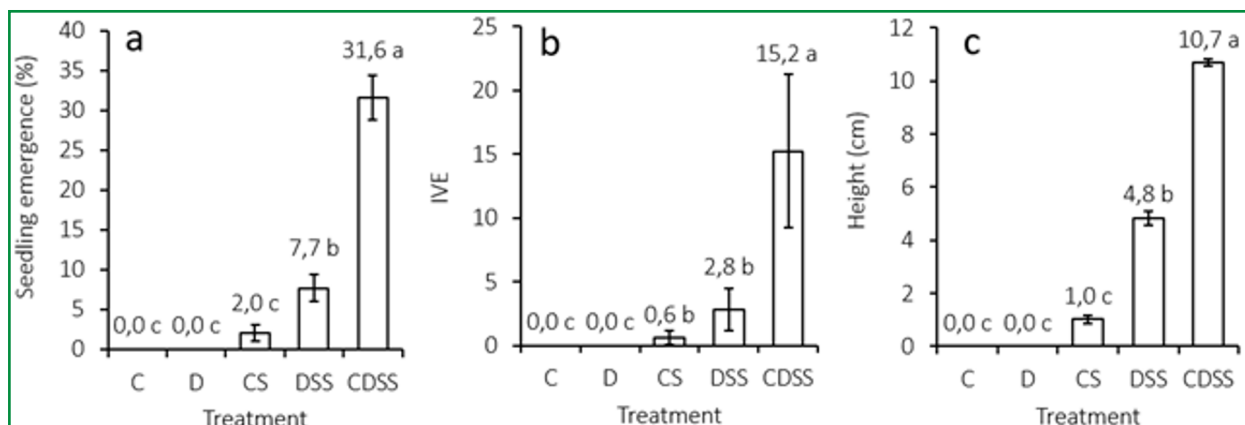
under a mean temperature of 26 °C and mean relative humidity of 80% for 70 days, according to Poletto *et al.* (2016). The following variables were evaluated: (a) seedling emergence (%), (b) emergence speed index, according to Maguire (1962), and (c) seedling height. The data were subjected to analysis of variance (ANOVA), and treatment means were compared using the Scott-Knott test ( $\alpha = 0.05$ ) with RStudio software (RStudio Team, 2020).

### 3 RESULTS AND DISCUSSIONS

The percentage of emergence, emergence speed index, and seedling height differed significantly among treatments (Table 2). Treatments that did not include a physical method for breaking dormancy did not result in seedling emergence (C and D). These results reinforce the need to apply a physical treatment to stimulate germination in pecan. The treatment combining refrigeration, disinfestation, scarification, and stratification resulted in the highest emergence ( $31.6 \pm 2.8\%$ ), ESI ( $15.2 \pm 6.0$ ), and seedling height ( $10.7 \pm 0.1\%$ ), indicating that the combination of physical methods may be an effective strategy to improve the likelihood of successful early seedling establishment. The treatment involving scarification and stratification (DSS), without prior exposure to low temperature (4 °C for 15 days), resulted in 7.7% seedling emergence, whereas stratification alone (CS) resulted in only 2.0% emergence (Figure 1a).

The results suggest that pecan emergence is strongly conditioned by a combination of physical stimuli that act synergistically to overcome dormancy. The 15-day pre-refrigeration period may have promoted initial metabolic changes that, together with subsequent scarification and stratification, facilitated both water uptake and the activation of enzymatic mechanisms involved in dormancy breaking. The importance of combining stimuli for dormancy breaking is not restricted to pecan. In a study with *Spartina alterniflora* Loisel. seeds, a longer stratification period combined with higher temperatures resulted in the highest germination rates (Cheng *et al.*, 2022).

Figure 1 - Emergence percentage (a), emergence speed index (b), and seedling height (c) of *C. illinoensis* subjected to different dormancy-breaking treatments, assessed 70 days after sowing



Source: Authors (2025)

Where: Treatments: C, control without disinfestation or dormancy breaking; D, disinfestation only; CS, cold stratification only; DSS, disinfestation, scarification, and stratification; CDSS, refrigeration, disinfestation, scarification, and stratification. Lowercase letters indicate significant differences according to the Scott-Knott test ( $p < 0.05$ ).

Recent pecan-specific studies have shown that cold stratification induces complex mechanisms associated with dormancy breaking, including the regulation of hormonal balance, with a reduction in abscisic acid (ABA) and an increase in gibberellins (GA), as well as the modulation of reactive oxygen species (ROS), which act as signals to activate germinative metabolism (Xue *et al.*, 2024). This molecular-level activation helps explain the superior emergence and seedling vigor observed in the combined treatment used in this study.

According to Xue *et al.* (2024), cold treatments induce the synthesis and activation of hydrolytic enzymes in the seed, helping to increase seed coat permeability. In addition, the low efficiency observed in the individual treatments (CS and DSS) shows that applying only one or two methods is not sufficient to overcome the physiological barriers imposed by the seed. This pattern is consistent with other studies showing that combining methods, such as mechanical scarification and stratification, is more effective than applying each technique separately (Wang *et al.*, 2025).

The effect of physical and chemical methods on dormancy breaking has also been demonstrated in previous studies. Poletto *et al.* (2015) found that cold stratification at 4 °C for 90 days promoted 93.3% emergence, while mechanical scarification with sandpaper resulted in 76.7%, and the combination of both methods increased emergence to 96.7% (Table 2). Similarly, Adams and Thielges (1978) observed that emergence varied according to the stratification period at approximately 5 °C, reaching 62% after 60 days and 71% after 90 days; by contrast, chemical scarification with sulfuric acid resulted in only 13% emergence, suggesting a damaging effect on the seed coat (Table 2).

Table 2 - Seedling emergence percentage in species of the family Juglandaceae subjected to different dormancy-breaking methods. The values shown correspond to results from different studies

| Species               | Cold stratification                         | Scarification                              | Combination (scarification + stratification) | Reference                    |
|-----------------------|---------------------------------------------|--------------------------------------------|----------------------------------------------|------------------------------|
| <i>C. illinoensis</i> | 2.0% (150 days)                             | -                                          | 31.6% (with prior refrigeration)             | This study                   |
| <i>C. illinoensis</i> | 93.3% (90 days)                             | 76.7% (sandpaper)                          | 96.7% (90 days)                              | Poletto <i>et al.</i> , 2015 |
| <i>C. illinoensis</i> | 62–71% (60 and 90 days)                     | 13% (H <sub>2</sub> O <sub>4</sub> 10 min) | -                                            | Adams and Thielges, 1978     |
| <i>C. illinoensis</i> | 28.7-86.7 (with and without prior freezing) | -                                          | -                                            | Wang <i>et al.</i> , 2025    |
| <i>C. illinoensis</i> | 89–91% (60 e 90 days)                       | -                                          | 98-95% (60 and 90 days)                      | Poletto <i>et al.</i> , 2016 |
| <i>Juglans regia</i>  | -                                           | 66.1% (cracked seeds)                      | 88.3% (45 days)                              | Raoufi <i>et al.</i> , 2020  |

Source: Authors (2025)

Studies with *Tarenaya hassleriana* (Chodat) Iltis, *Campomanesia phaea* (O. Berg.) Landrum, and *Rubus sellowii* Cham. Schltdl. have shown that combining physical treatments, such as scarification and temperature alternation, with physiological treatments, such as stratification and GA<sub>3</sub> application, is effective for breaking seed



dormancy in woody species from temperate and subtropical climates (Ren-Fei *et al.*, 2023; Duarte and Cardoso, 2024; Sinhorini *et al.*, 2025). This pattern is also observed in pecan seeds.

Poletto *et al.* (2016) also highlighted the influence of stratification duration, reporting 89% and 91% emergence after 60 and 90 days, respectively. In the same study, the combination of scarification and stratification increased emergence to 98% after 60 days and 95% after 90 days (Table 2). In another study, Wang *et al.* (2025) reported that stratification without prior freezing promoted 86% emergence, whereas combining it with freezing reduced this value to 28.7%, indicating that freezing is not recommended. Finally, in *Juglans regia* L., another member of the family Juglandaceae, Raoufi *et al.* (2020) showed that the combination of methods resulted in higher emergence (88.3%) compared with scarification alone (66.1%) (Table 2).

Finally, it should be noted that even in the best treatment tested (CDSS), the emergence percentage remained low (31.6%). This performance was likely associated with a loss of viability resulting from the four-month storage of seeds at room temperature (25 °C) before the experiment was established. This is a routine practice in nurseries, where dormancy breaking does not always occur immediately after seed collection. Such a storage period can severely compromise seed vigor, since pecan seeds are recalcitrant and therefore sensitive to desiccation and rapid loss of physiological quality (Dambros *et al.*, 2023; Wang *et al.*, 2023). Pecan seeds show high sensitivity to storage time and conditions, losing viability rapidly when kept under uncontrolled environments (Bonner, 2008). Under these circumstances, deterioration is accelerated by increased respiration and microbial activity, in addition to excessive drying of the seed coat, which may damage the embryo (Dambros *et al.*, 2023; Cai *et al.*, 2025). In this context, future studies should investigate how different storage conditions, including temperature, moisture, and sanitary quality, affect seed viability preservation.

Studies that deepen the understanding of dormancy in recalcitrant seeds, such as pecan, are essential. Such knowledge will support the development of more efficient protocols for rootstock production and the commercial propagation of the species.



## 4 CONCLUSIONS

The results indicate that pecan seedling emergence depends strongly on the combination of physical methods, with pre-refrigeration combined with scarification and stratification being the most effective strategy. Even so, the low emergence obtained highlights the effect of prolonged storage on seed viability loss. Future studies should prioritize appropriate storage conditions and the refinement of dormancy-breaking protocols to improve the success of seedling establishment.

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