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Impact of seed size on germination parameters in *Flacourtia indica* (Burm.f.) Merr.

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Abstract

Seed size is a plastic trait in plants that directly influences seed germination and seedling recruitment. This study aimed to investigate the relationship between seed size, seed germinability, and seedling performance in *Flacourtia indica* (Burm.f.) Merr., a species with significant ecological and medicinal importance. Seeds were categorized into five size classes: very large, large, medium, small, and very small, and analyzed using seed image analysis. Morphometric parameters, including area, length, width, aspect ratio, average diameter, perimeter, convex perimeter, and roundness, were measured for each size class. The analysis revealed significant variations in these parameters across the different size classes. Results indicated that smaller seeds had a higher germination percentage and a proportionally greater amount of resources to their roots compared to those from larger seeds. This suggests that smaller seeds, with their higher germination rates and faster germination times, are better suited for colonizing transient habitats. Conversely, larger seeds, which produce more vigorous seedlings, are more advantageous for establishment in stable habitats. Thus, we argue that the high variability in seed size of *Flacourtia indica* (Burm.f.) Merr. supports its widespread geographic distribution. This study underscores the importance of seed size as a determinant of successful germination and early seedling development in *Flacourtia indica*. The findings have implications for seed selection strategies in reforestation and conservation programs, aiming to enhance the propagation success and establishment of this valuable species.

Keywords: Seed size, image analysis, morphometry, viability, vigour, germination

Introduction

Flacourtia indica (Burm.f.) Merr., commonly known as the Governor's Plum, is a species of considerable ecological, medicinal, and economic importance. Native to tropical and subtropical regions, *Flacourtia indica* is valued for its edible fruits, traditional medicinal uses, and potential in agroforestry systems. It can tolerate a wide range of habitats, from coastal and riverine areas to lowland dry and humid environments (Dunlop *et al.*, 2005) ^[7]. It is found at altitudes ranging from 0 to 2400 meters (PIER, 2014) ^[19]. Commonly found in tropical dry deciduous and thorn forests, *Flacourtia indica* is more abundant in the former. It also thrives in seasonally dry forests, woodlands, bushlands, thickets, wooded grasslands, and often in riparian vegetation (Orwa *et al.*, 2009) ^[18]. However, the primary propagation method through seeds is challenged by poor germination rates, often less than 20% (Prins & Maghembe, 1994) ^[21]. Successful propagation and establishment of this species remain challenging, particularly due to variability in seed quality and germination performance.

Seed size is a critical determinant of plant reproductive success, influencing key processes such as seed germination and seedling establishment. In many plant species, seed size exhibits considerable plasticity, with significant implications for ecological adaptation and species distribution. Seed size directly influences the germination time (Murali, 1997) ^[16], germination percentage (Mälken *et al.*, 2005) ^[15], and seedling vigor (Yanlong *et al.*, 2007) ^[27], which can indirectly determine plant distribution and abundance across different habitats (Silveira *et al.*, 2012) ^[24]. However, seed size produced by plants varies between and within plant species, sometimes by several orders of magnitude (Leishman *et al.*, 1995; Silvertown & Bullock, 2003; Moles & Westoby, 2006) ^[12, 25, 14].

Several ecological and evolutionary factors can affect the process of seed germination and further plant establishment (Marques & Oliveira, 2005) ^[13]. Seed size has been considered an important evolutionary trait that affects the reproductive outcome of many plant species (Cordazzo, 2002) ^[5]. A number of studies on the effect of seed size on seedling success have been made but in depth knowledge of this relationship in tropical tree species remains incomplete. Understanding the relationship between seed size and germination characteristics in *Flacourtia indica* is essential for developing effective propagation protocols. This study aims to analyse the morphometric parameters of *Flacourtia indica* seeds across different size classes and evaluate their impact on germination performance and seedling vigour. By identifying optimal seed size classes for propagation, this research seeks to inform seed selection strategies that enhance the establishment and growth of *Flacourtia indica* in reforestation and conservation initiatives.

Materials and Methods

Seed Collection and Categorization

Seeds of *Flacourtia indica* were collected from mature fruits in a natural population located in Mandla Forest division of Madhya Pradesh. The seeds were cleaned, air-dried, and then categorized into five size classes by optical observation: very large, large, medium, small, and very small. This categorization was based on visual inspection and grouped using seed image analysis software.

Seed Image analysis

An automated image analysis system (Biovis Seed Image Analyzer PSM 3000) was used to characterize the morphometric properties of *Flacourtia indica* seeds. This system employs a digital camera or flat-bed scanner to capture high-resolution images. During the analysis, seed samples were evenly distributed on a specialized measuring plane, ensuring consistent image capture under optimal lighting conditions to avoid shadows and reflections. The images were processed using dedicated software, which automatically measured various morphometric parameters such as seed area, length, width, aspect ratio, average diameter, perimeter, convex perimeter, and roundness. These parameters were then statistically analysed to identify significant differences between different seed size classes.

Germination Trials

The germination test was performed by placing three replicates of 50 seeds each on top of paper (TP) in petri plates at 30 ± 2 °C with a 16/8 light/dark photoperiod. Germination was counted daily for 28 days. Seeds were considered germinated when the radicles reached at least 1 cm. The percentage of germination was then calculated.

Seed Germination Data measurements: Data on seed germination were recorded starting from seed germination till culmination and computed germination-related parameters.

Germination-related parameters were determined as follows:

1. Time Spread of Germination (TSG) (Kader, 1998) ^[11]

$$\text{TSG} = \text{LDG} - \text{FDG}$$

Where

First Day of Germination (FDG) (Kader, 1998) ^[11]

$$\text{FDG} = \text{Dg} - \text{Ds}$$

where Dg = first germination day and Ds = seed sowing day.
Last Day of Germination (LDG) (Kader, 1998) ^[11]

$$(\text{LDG}) = \text{Df} - \text{Ds}$$

where Df = final germination day and Ds = seed sowing day.
2. Final Germination Percentage (FGP) (Scott *et al.*, 1984) ^[23]

$$\text{FGP} = G / T \times 100$$

where G = no. of germinated seeds and T = no. of seeds sown.
3. Mean germination time (MGT) (Orchard, 1977) ^[17]

$$\text{MGT} = G_t \times D_t / G$$

where G_t = no. of germinated seeds at day-t, D_t = no. of days at 't' from the day of sowing and G = total no. of germinated seeds.

4. Mean daily germination (MDG) (Czabator, F. J., 1962) ^[6].

$$\text{MDG} = \text{GP} / \text{Gd}$$

where GP = germination percentage and Gd = no. of days to complete germination.

5. Vigour index I (Abdul-Baki, and Anderson, 1973) ^[1]

$$\text{VI I} = \text{FGP} \times \text{Seedling length (cm)}$$

Statistical Analysis

Data were analysed using ANOVA to test for significant differences in seed size, weight, moisture content, viability, and germination parameters between different developmental stages and pre-treatment methods.

Results

Morphometric Seed Image Analysis

Significant variations were observed in the morphometric parameters across the different seed size classes (Figure1). Very large seeds had the highest values for area, length, width, average diameter, and perimeter, while very small seeds had the lowest. Medium-sized seeds exhibited intermediate values for most parameters.

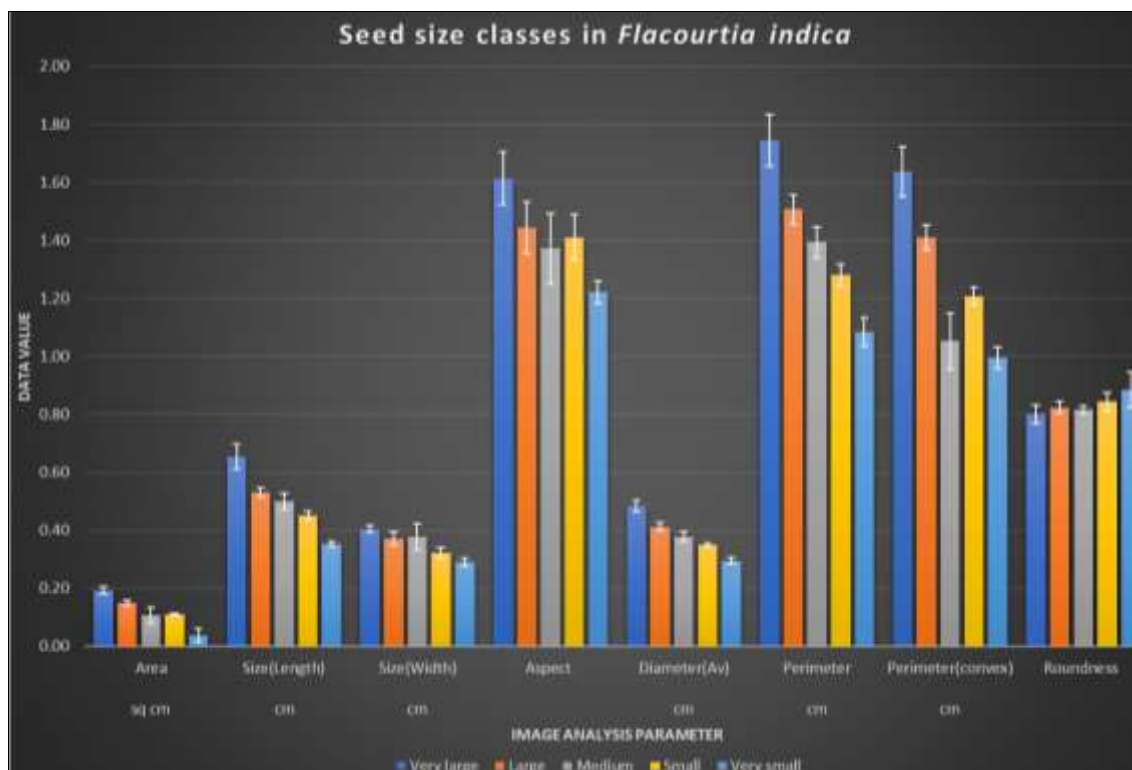


Fig 1: Seed Size Classes Based on Image Analysis (Standard Errors Depicted in the Graph)

Germination Dynamics

On the 4th day, initial germination was highest in small seeds with a mean germination of 3.50% (SE 0.29), followed by very small seeds at 2.83% (SE 0.44). Medium seeds had a mean germination of 1.50% (SE 0.76), large seeds 0.67% (SE 0.67), while very large seeds showed no germination. This indicates a quicker start in the germination process for smaller seeds, with very large seeds showing a delay. By the 7th day, large seeds exhibited the highest germination at 18.76% (SE 0.80), followed by very large seeds at 12.67% (SE 0.93), small seeds at 12.84% (SE 1.45), medium seeds at 12.67% (SE 0.87), and very small seeds at 8.28% (SE 0.40). This suggests rapid progression in

larger seeds, while smaller seeds maintained strong rates, though the initial advantage diminished. On the 10th day, very large seeds led with 16.27% (SE 1.18), followed by small seeds at 10.70% (SE 1.46), medium seeds at 9.91% (SE 0.79), large seeds at 7.73% (SE 0.79), and very small seeds at 6.00% (SE 1.26). This continued strong rate in very large seeds may indicate delayed but sustained germination potential. By the 14th day, germination rates declined for all sizes, with very large seeds at 4.33% (SE 0.60), medium seeds at 5.25% (SE 0.66), and large seeds at 4.00% (SE 0.87), while small and very small seeds showed no additional germination, suggesting they had completed their germination earlier.



Fig 2: Germination dynamics for different seed size classes

Time Spread of Germination

Very large seeds had a mean time spread of 7.00 days. Large seeds had a mean time spread of 8.00 days (SE 1.00). Medium seeds had a mean time spread of 9.00 days (SE 1.00). Small and very small seeds both had a mean time spread of 6.00 days.

Medium seeds had the longest time spread, indicating a more extended and possibly more resilient germination period. Small and very small seeds had the shortest spread, suggesting a more condensed germination period.

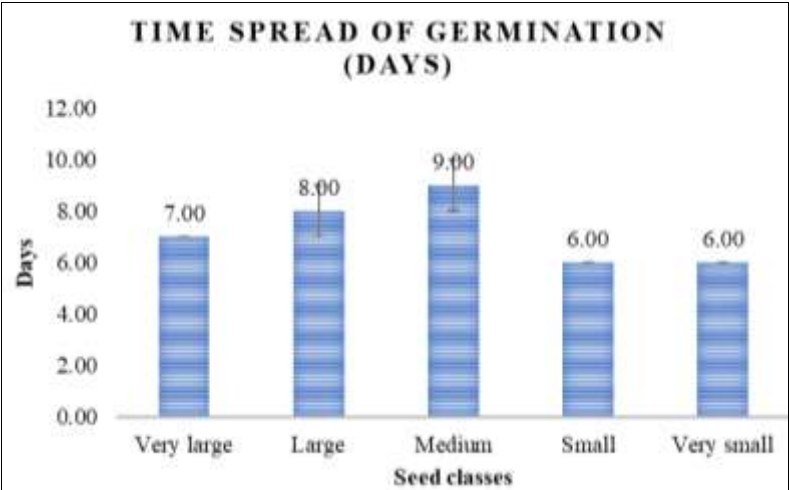


Fig 3: Time Spread of Germination (days) for different seed size classes

Final Germination Percentage (FGP)

Very large seeds had a mean FGP of 33.27% (SE 1.95). Large seeds had a mean FGP of 31.16% (SE 0.65). Medium seeds showed a mean FGP of 29.33% (SE 1.76). Small seeds exhibited a mean FGP of 27.04% (SE 3.09). Very small seeds had a mean

FGP of 17.12% (SE 1.59). Very large seeds achieved the highest final germination percentage, indicating a robust overall germination potential. Very small seeds had the lowest FGP, reflecting less successful germination compared to larger seeds.

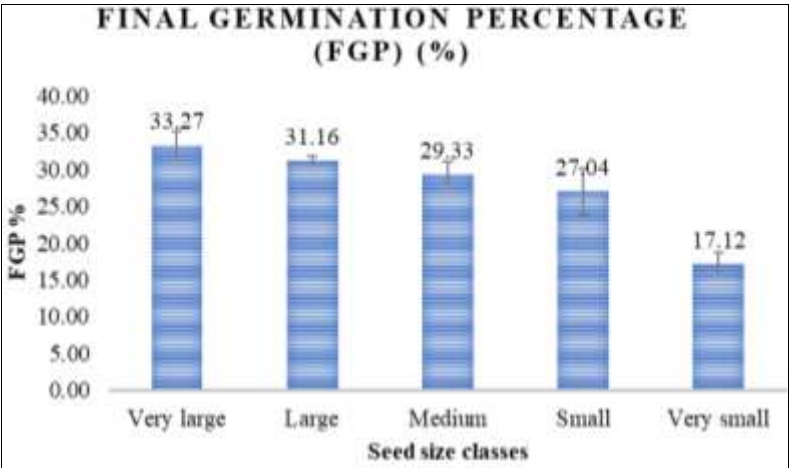


Fig 4: Final Germination Percentage (FGP) (%) for different seed size classes

Mean Germination Time (MGT): Very large seeds had a mean MGT of 9.37 days (SE 0.12). Large seeds had a mean MGT of 8.58 days (SE 0.17). Medium seeds showed a mean MGT of 9.15 days (SE 0.28). Small seeds exhibited a mean MGT of 7.79

days (SE 0.06). Very small seeds had a mean MGT of 7.54 days (SE 0.19). Medium and very large seeds took slightly longer to complete germination compared to smaller seeds, which showed a quicker germination cycle.

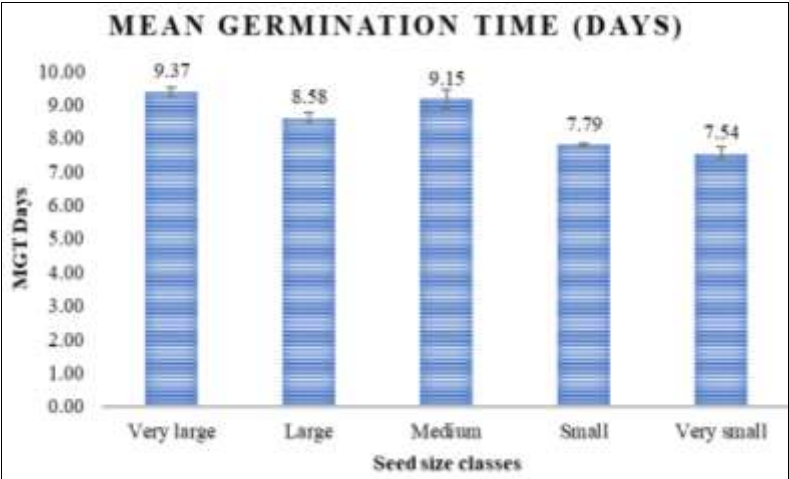


Fig 5: Mean Germination Time (Days) for different seed size classes

Mean Daily Germination: Very large seeds had a mean daily germination of 8.47% (SE 0.23). Large seeds had a mean daily germination of 7.90% (SE 0.32). Medium seeds showed a mean daily germination of 8.17% (SE 0.18). Small seeds exhibited a mean daily germination of 6.21% (SE 0.18). Very small seeds

had a mean daily germination of 5.10% (SE 0.17). Very large seeds had the highest mean daily germination, indicating consistent germination activity. Very small seeds had the lowest mean daily germination, aligning with their lower overall germination performance.

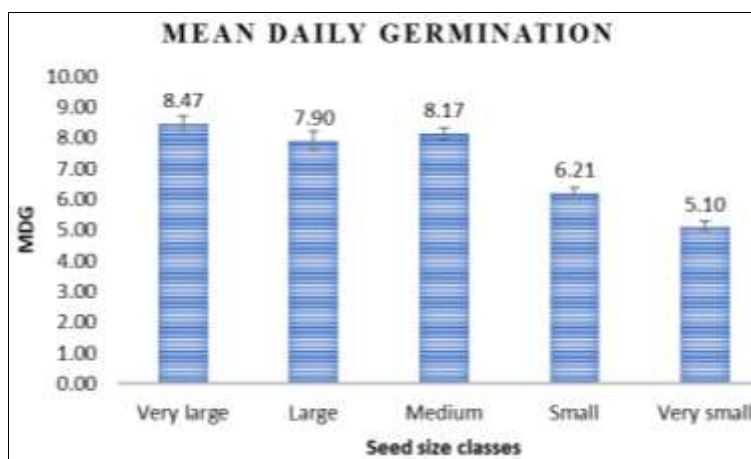


Fig 6: Mean Daily Germination for different seed size classes

Vigour Index I

Very large seeds had a mean vigour index of 280.93 (SE 11.61). Large seeds had a mean vigour index of 246.38 (SE 13.90). Medium seeds showed a mean vigour index of 240.08 (SE 19.23). Small seeds exhibited a mean vigour index of 168.28 (SE 20.55). Very small seeds had a mean vigour index of 87.61 (SE 10.39). Very large seeds demonstrated the highest vigour index, indicating robust seedling development. Very small seeds

had the lowest vigour index, reflecting weaker seedling growth and vitality. The data show that while smaller seeds may start germinating sooner, larger seeds tend to have higher overall germination rates, longer germination periods, and produce more vigorous seedlings. This information is crucial for seed selection strategies in propagation and conservation programs, as it highlights the trade-offs between seed size, germination speed, and seedling vigor in *Flacourtia indica*.

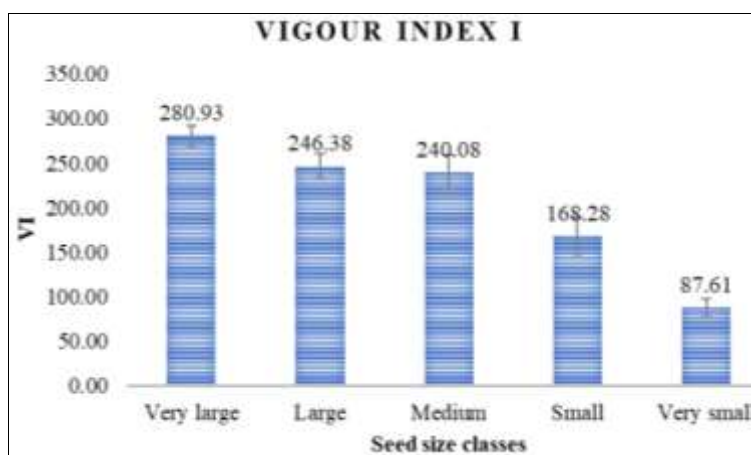


Fig 7: Vigour index I for different seed size classes

Discussion

This study provides a detailed analysis of the influence of seed size on the germination dynamics and seedling vigour of *Flacourtia indica*. The data reveal several key trends that enhance our understanding of seed biology and have implications for seed selection in conservation and propagation efforts. The morphometric data confirm that very large seeds have the highest values for area, length, width, average diameter, and perimeter, whereas very small seeds exhibit the lowest values. This finding aligns with established botanical knowledge, where seed size is generally correlated with seed reserve quantity and seedling growth potential (Armstrong & Westoby, 1993; Baskin & Baskin, 1998) [2, 3]. The larger dimensions of very large seeds likely contribute to their enhanced initial growth capabilities due to increased nutrient

reserves. The initial germination data indicate that smaller seeds (Small and very small) exhibit a faster start compared to larger seeds. This rapid initiation can be attributed to smaller seeds' lower energy requirements and more efficient metabolic processes during the early stages of germination (Beninger *et al.*, 1998) [4]. However, the trend shifts by the 7th day, where larger seeds, particularly large and very large, surpass smaller seeds in germination rates. This shift suggests that while smaller seeds initiate germination more quickly, larger seeds, with their greater nutrient reserves, support prolonged and more substantial germination processes. The continued strong germination in very large seeds by the 10th day, despite their initial delay, underscores their robust germination potential. This delayed but sustained germination could be advantageous in fluctuating natural environments, allowing these seeds to capitalize on

extended favorable conditions (Fowler & Bianchetti, 2000) [8]. By the 14th day, the overall germination rates declined across all seed sizes, which may reflect the completion of the germination phase for seeds with high initial rates, while larger seeds continued to germinate but eventually plateaued. The time spread of germination was longest for medium-sized seeds, indicating a more extended germination period that might reflect greater resilience to environmental fluctuations. Conversely, small and very small seeds had shorter time spreads, suggesting a more condensed germination period. This characteristic may benefit rapid colonization but could imply less adaptability to varying conditions (Yang & Midmore, 2005) [29]. The final germination percentage was highest for very large seeds and lowest for very small seeds. This result emphasizes the advantage of larger seeds in achieving higher overall germination success. The higher FGP in very large seeds likely results from their substantial nutrient reserves, which support successful germination (Primack, 1987) [20]. Larger seeds took slightly longer to complete germination compared to smaller seeds, which may reflect the more complex metabolic processes associated with larger seeds. This extended germination time is consistent with the notion that larger seeds require more time to fully utilize their nutrient reserves (Geritz, 1995) [9]. Very large seeds exhibited the highest mean daily germination, suggesting consistent germination activity. This result aligns with their overall higher germination rates, indicating that larger seeds maintain a steady germination rate over time. In contrast, very small seeds had the lowest mean daily germination, which corresponds with their lower overall germination performance. The vigour index highlighted that very large seeds produced the most vigorous seedlings, while very small seeds produced the weakest seedlings. This observation underscores the relationship between seed size and seedling vigor, with larger seeds contributing to stronger and more resilient seedlings (Yanlong *et al.*, 2007) [27]. The results are consistent with findings in other species (Beninger *et al.*, 1998; Harper, 1977; Cordazzo, 2002; Walters, 1998) [4, 10, 5, 26]. This comparison suggests that while smaller seeds may offer quick germination, larger seeds often provide advantages in terms of overall performance and resilience, aligning with the observed patterns in *Flacourtia indica*.

The study's findings have practical implications for seed selection in conservation and propagation programs. Medium-sized seeds emerged as the most effective for sowing, offering a balance between germination rate and seedling growth. This insight is valuable for optimizing seed selection strategies, particularly in reforestation and conservation efforts (Rees & Westoby, 1997) [22].

Conclusion

In conclusion, this research demonstrates that seed size significantly influences germination dynamics and seedling vigor in *Flacourtia indica*. While smaller seeds may germinate more quickly, larger seeds tend to have higher overall germination rates and produce more vigorous seedlings. These insights can guide seed selection in conservation and propagation programs, contributing to the successful establishment and management of *Flacourtia indica* populations.

Conflict of interest: The authors declare no conflicts of interest.

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