

Seed germination and growth of *Irvingia gabonensis* (Aubry-Lecomte ex O'Rorke) Baill. (Irvingiaceae) seedlings in a controlled environment east of Taï National Park (Buyo, South-West of Ivory Coast)

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Abstract

Anthropogenic activities tend to destroy forest resources such as *Irvingia gabonensis*, a species highly prized by the populations living near the Taï National Park (PNT). The enormous pressure on the natural stands of this species leads to its rarefaction, even its disappearance. In order to make the species accessible, the rates of seed germination and seedling growth in a controlled environment were evaluated through five (5) dormancy lifting treatments and control seeds (T0), all in a Fisher random experimental design. The results showed that seeds subjected to T3 treatments (soaking seeds in 33% diluted sulfuric acid for 96 h followed by rinsing with well water), T4 (soaking seeds in well water for 96 h) and T5 (Manual scarification of seeds using a file) have a better germination rate (respectively 83.33%, 80% and 66.66%). The shortest germination times were recorded with T5 (5 days), T2 (heating at 45°C for 10 min followed by soaking the seeds in well water for 24 h) and T3 (8 days) and T1 (soaking the seeds in well water for 24 h followed by heating at 45 °C for 10 min) with 11 days. On the other hand, T1 and T4 gave the shortest germination spans (4 and 6 days respectively). In addition, the height growth of seedlings from T2, T3 and T4 was better. However, serious regressions were observed in the diameter growth of seedlings from T2 and T3 after 21 days. This study showed that although several dormancy breaking techniques are promising, the soaking seeds in sulfuric acid for 96 h (T3) and in water for 96 h also (T4), remain better for the domestication of *Irvingia gabonensis*, a crucial step for its large-scale valorization.

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Keywords : Utility species; anthropogenic activity; rarefaction; domestication; natural reserve

INTRODUCTION

Tropical forests, due to their biological richness, play a vital role in human life. Indeed, for many rural communities, the forest represents not only an economic, but also a social and cultural importance [1]. Thus, many species of forest ecosystems are used in different areas of life. Unfortunately, these natural ecosystems, habitats of these plants, are today particularly threatened by the rise of human activities. As anthropogenic pressures on these ecosystems are increasing, the availability of forest species remains dependent on resource exploitation techniques and the conditions of the natural environment [2 ; 3].

This problem of rapid disappearance of natural plant resources due to human activities is a worrying subject, as highlighted by various studies [4 ; 5 ; 6 ; 7 ; 8].

In Côte d'Ivoire, human pressure is resulting in an alarming reduction in forest areas, mainly due to the excessive collection of useful plants [9].

In this Ivorian context, the Taï National Park (PNT), rich in biodiversity, is facing increasing threats due to human activity, particularly the excessive collection of non-timber forest products (NTFPs) by local populations [10]. The over-

exploitation of species such as *Ricinodendron heudelotii*, *Beilschmiedia mannii* and *Irvingia gabonensis*, essential for food and the local economy, has led to their scarcity in the peripheral areas of the park. This situation is now pushing populations to turn to the park for the collection of products from these species, thus disrupting the biodiversity of the park. Despite several studies on the valorization and conservation of plant species essential for rural populations [11 ; 12 ; 13 ; 14], few efforts have been made in the country for the domestication of these species, a crucial step for their valorization [15].

The objective of this work is to determine better methods of germination of *Irvingia gabonensis* seeds in order to facilitate its integration into cultivation systems on the periphery of the park.

MATERIALS AND METHODS

Plant material

The fruit of *Irvingia gabonensis* is a smooth drupe resembling a small mango, greenish becoming yellow when ripe [16] (Figure 1). It contains a very fibrous yellow juicy pulp adhering to the stone. The latter is hard, subspherical, slightly flattened, 3 to 5 cm long. It contains a white, fleshy, mucilaginous, discoid and very flattened almond. It is this edible almond that is sought after by local populations for food and marketing.

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Figure1: Fruits of *Irvingia gabonensis*

Methods

Choice of study site

The tests took place at the monitoring station of the ADK/V6 sector of the Taï National Park (PNT), in the Nawa region, more precisely in the Buyo sub-prefecture (Figure 2). This monitoring station, bordered by the V villages, offered the best conditions for carrying out the germination and vegetative multiplication tests. Indeed, the monitoring station of this sector is located almost inside the Park. This geographical location offers the experimental site favorable edaphic and climatic conditions. Also, this sector benefited in 2014 from a plant production project (Project financed by GIZ and piloted by OIPR) of three species including *Irvingia gabonensis*. This made it easy to access plant material of different ages.

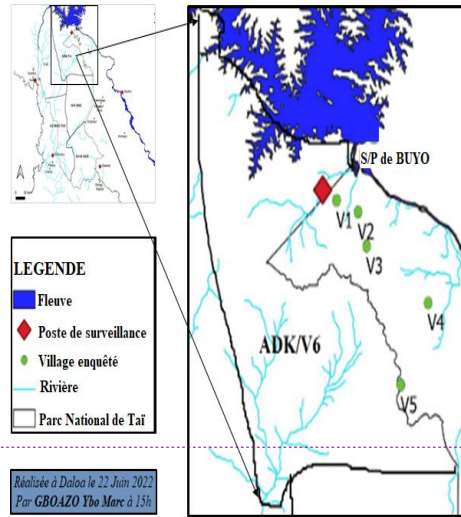


Figure2: Location map of the study area

Data collection

Installation of the shade

A 96 m² (8 mx 12 m) shade house was made with local materials (palm stalks, wooden poles, Chinese bamboo and raffia) and covered with palm leaves to reduce solar intensity on the seedlings (Figure 3).

In addition, 500 15 cm x 5 cm nursery bags were filled with soil from the forest undergrowth, then placed under the shade to receive the seeds.



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Figure3: Shade for germination tests

Acquiring and sorting fruits

The fruiting and harvesting periods of *Irvingia gabonensis* were identified thanks to the work of [10], coupled with information collected from agents of the Ivorian Office of Parks and Reserves (OIPR). Thus, approximately 800 fresh fruits were collected in the Park, at the foot of the mother trees (seed trees) in December 2022. In order to keep only good quality fruits (well formed), careful sorting was carried out (Figure 4).



Figure4:Sorting *Irvingia gabonensis* fruits

Seed extraction and pre-treatment

The fruits have been stripped of their fleshy mesocarp covering the hard endocarp or stone containing the seed. It is this stone which represents the seed of the species (Figure 5).



Figure5:Seeds of *Irvingia gabonensis* stripped of the fleshy part

With reference to the work of [17], five pre-treatments (T1, T2, T3, T4, T5) and a control (T0) were adopted, at a rate of 60 seeds per treatment.

- treatment witness T0: untreated seeds.

- treatment T1: the seeds were soaked in well water for 24 h away from sunlight before being heated in the laboratory in an autoclave at 45°C for 10 minutes.

- treatment T2: the seeds were first heated before being soaked, as in treatment T1. The seeds thus pretreated were kept in a moist earth clod to be transported to the experimental site.

- T3 treatment: the seeds were soaked in sulfuric acid (H₂SO₄) diluted to 33% for 96 h (4 days), then rinsed with well water.

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- T4 treatment: seeds were soaked in well water for 96 h.

- T5 treatment: the seeds were manually scarified using a new file at the level of the part close to the embryo, by sanding until the seed was exposed [18].

Experimental device

In order to standardize the level of the tests, an experimental design in completely randomized blocks with three repetitions per treatment was adopted (Figure 6), in accordance with [19 ; 20 ; 21]. Indeed, this design makes it possible to reduce the effect of variations between blocks and to better evaluate the effect of treatments. Thus, three blocks were set up under the shade to accommodate the seeds (Figure 8). Each block was subdivided into six sub-blocks (from T0 to T5) of 20 sachets each, for a total of 360 sachets.

T0	T1	T4
T1	T5	T3
T3	T4	T2
T2	T0	T5
T5	T2	T1
T4	T3	T0

Figure6: Diagram of the experimental device adopted



Figure7: View of the experimental device under the shade

Sowing and measuring seedling germination and growth

One seed was sown per sachet followed by systematic watering of the entire device, once a day (in the morning) until germination. When the leaves appeared, watering was done twice a day (in the morning and in the evening) except on rainy days to ensure good growth of the seedlings.

In this work, a seed was considered to have germinated when its seedling appeared above the surface of the substrate contained in the sachet [17]. The germination parameters measured were: time to first germination, time to last germination and number of seeds germinated.

Growth parameters measured on seedlings were: seedling height (from collar to terminal bud) using a graduated ruler

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(Figure 8), diameter at the collar using a caliper (Figure 9). Data collection took place weekly for four weeks.



Figure8:Measuring the hauteur of a seedling of *Irvingia gabonensis* using a graduated ruler



Figure9:Measuring the Diameter at the collar of an *Irvingia gabonensis* seedling using a caliper

Data processing

Excel spreadsheet was used for data entry. R software version 4.3.2 was used to perform statistical analyses, including

analysis of variance (ANOVA) to compare treatments.

Calculating the germination rate

For each treatment, the germination rate (Tg) was determined according to the following formula proposed by [22] :

$$Tg = \frac{n_i}{N} \times 100 \text{ with}$$

- n_i : number of seeds germinated on date i
- N : total number of seeds sown.

Seed Latency Time Evaluation

Lag time (LT) is the number of days between sowing and first germination [22]. It was determined using the formula below:

$$T_L = [d_1 - d_0] \text{ with}$$

- d_0 : sowing date
- d_1 : date of first germination

Evaluation of the extent of germination

The germination extent (Eg) is the time between the first and last germination [17]. It was determined using the following formula:

$$Eg = t_1 - t_0 \text{ with}$$

- t_0 : number of days between sowing and first germination
- t_1 : number of days between sowing and last germination

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RESULTS AND DISCUSSION

Results

Effect of treatments on germination

Treatments applied to the kernels significantly influenced seed germination of *Irvingia gabonensis* ($p=0.000013$) (Table I). Indeed, the Newman-Keuls test indicates that the Treatments T3, T4 and T5 induced the best germination rates, respectively 83.33%; 80% and 66.66%. On the other hand, the lowest germination rates were observed in the control T0 (26.66%) and in the pretreatments T1 (38.33%) and T2 (21.66%).

Table I: Effect of treatments on germination rate from *Irvingia gabonensis*

Treatment	Germination rate (%)
T0 (witness)	26.66 ± 7.63a
T1 (H ₂ O soaking + heating)	38.33 ± 7.63a
T2 (heating + H ₂ O soaking)	21, 66 ± 7.63a
T3 (H ₂ SO ₄ soaking)	83.33 ± 16.07b
T4 (H ₂ O soak)	80 ± 10.40b
T5 (scarification)	66.66 ± 7.63b
F	21,6139
p	0.000013

Effect of treatments on germination kinetics

The applied pretreatments significantly influenced the latency time and extent of germination. Indeed, the Treatments T2, T3 and T5 made it possible to shorten the germination time (Table II), while control

seeds (T0) and those from treatment T4 took much longer to germinate (respectively 32 and 37 days after sowing). However, the shortest germination times were observed in treatments T0, T1 and T4.

Table II: Effects of treatments on latency and extent of germination

Treatment	Latency time (days)	Extent of germination (days)
T0 (witness)	32 ± 4.04c	8 ± 1.15ad
T1 (H ₂ O soaking + heating)	11 ± 2.00b	4 ± 1.00c
T2 (heating + H ₂ O soaking)	8 ± 1.15ab	11 ± 1.15ab
T3 (H ₂ SO ₄ soaking)	8 ± 1.00ab	12 ± 2.51b
T4 (H ₂ O soak)	37 ± 2.51d	6 ± 0.57cd
T5 (scarification)	5 ± 0.57a	11 ± 2.08ab
F	114,1295	13,3864
p	0.000000	0.000147

Effect of treatments on seedling growth

Treatments applied to *Irvingia gabonensis* cores had no significant effect on seedling height ($p=0.617486$) aged 28 days (Table III). However, the growth dynamics during the first four weeks favors better height growth of seedlings from treatments T1, T2 and T3 compared to the others (Figure 10). On the other hand, the diameter was significantly influenced by the treatments ($p=0.004037$) (Table III). Thus, the best diametric performances were observed

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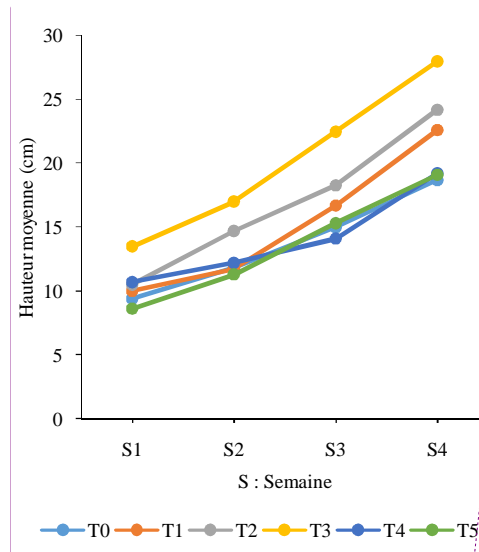
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with treatments T4 and T5 (Figure 11). This influence was also marked by a regression of the diameter after 21 days in seedlings from T1 and T2.

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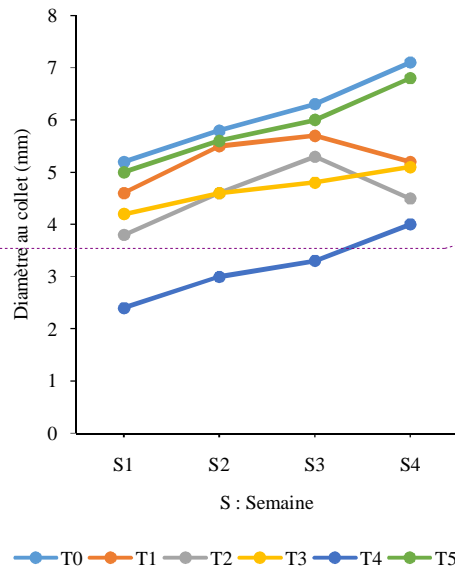
Table III: Effects of treatments on the diameter and height of the seedlings from *Irvingia gabonensis*

Treatment	Average height (cm)	Average diameter (mm)
T0 (witness)	15 ± 2.85a	6.07 ± 1.30c
T1 (H ₂ O soaking + heating)	16.33 ± 1.25a	5.66 ± 0.90bc
T2 (heating + H ₂ O soaking)	16.50 ± 2.65a	5 ± 0.81abc
T3 (H ₂ SO ₄ soaking)	16.53 ± 2.70a	4.13 ± 0.35ab
T4 (H ₂ O soak)	15.50 ± 1.01a	3.40 ± 0.20a
T5 (scarification)	14 ± 0.62a	5.20 ± 0.62abc
F	0.7253	6,4051
p	0.617486	0.004037



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Figure10: Height growth dynamics of seedlings of *Irvingia gabonensis* over four weeks



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Figure11: Diameter growth dynamics of seedlings of *Irvingia gabonensis* over four weeks

Discussion

The results obtained provide a positive insight into the effectiveness of pretreatments applied to *Irvingia gabonensis* seeds. This suggests a certain robustness of these treatments to promote the germination of these seeds. Thus, seeds soaked in sulfuric acid for 96 h followed by rinsing with water (T3), those soaked in well water for 96 h (T4) and those scarified (T5) led to the best germination performances with more than 65% of germinated seeds. The duration of soaking of the seeds (T3 and T4) would have been sufficient to stimulate germination. Indeed, the hard shell of the seeds would have been softened by the long soaking through the activation of enzymes, which increased their permeability essential for water absorption. Sulfuric acid, with its corrosive power due to its acidic nature, would have caused a degradation of the shell, thus facilitating germination. The control seeds (T0), those soaked in water for 24 h followed by heating (T1) and those heated then soaked in water for 24 h (T2) did not germinate sufficiently (less than 40% germination rate). The germination rate of the control seeds is close to those obtained by [23], [10] and [24] with untreated seeds of the same species, i.e. less than 50%. These results could be explained by the variability of the agro-ecological conditions of experimentation. In addition,

the control seeds (T0) and those of T4 required a long germination time (respectively 36 and 39 days), confirming that untreated seeds of *Irvingia gabonensis* have difficult germination in natural conditions. On the other hand, the shortest germination time (5 days) of scarified seeds (T5) corroborates that of [23] who also reported a short germination time of stripped seeds (9 days) unlike those not stripped (about 1 month). Furthermore, seeds from a treatment taking longer to germinate tended to have a shorter germination time, while those that germinated more quickly tended to have a more extended germination over time. This relationship is illustrated by treatments T0, T1 and T4 whose seeds, having taken longer to germinate, had a shorter germination time, while treatments T2 and T3 had the opposite effect. These observations could be explained by the influence of internal or external factors exerted on both the time and duration of germination. Internally, seed viability, water content, and nutrient content are key determinants of seed germination. Seeds must have sufficient reserves and be adequately hydrated to successfully initiate the germination process. Externally, environmental conditions play a major role. Temperature, moisture, light, and nutrient availability in the soil must be conducive to germination. Inhibitors in the

environment or interactions with soil microorganisms can also delay or promote germination.

Seedlings from treatments T1, T2 and T3 produced better height growth compared to the other treatments, although this difference was not statistically significant. On the other hand, regarding diameter growth, seedlings from treatments T4 and T5 showed the best performance. These results suggest that the different treatments have distinct effects on seedling growth, depending on the parameters measured. Height growth seems to be more influenced by treatments T1, T2 and T3, while diameter growth is favored by treatments T4 and T5. However, an interesting point to note is the observed effect of treatments T1 and T2 on diameter growth after 21 days. Despite an initial faster height growth, these treatments showed a regression in diameter growth. This observation suggests that there were specific limiting factors or constraints that hampered the diameter growth of seedlings in these treatments. Indeed, problems related to the development of the seedling root system such as soil structure (compact soil), lack of nutrients, or soil pH, would have contributed to this regression. Also, the seedlings of these treatments could have been confronted with a specific nutritional deficit, which would have

hindered their ability to maintain normal growth.

CONCLUSION

This study highlighted the significant impact of different pretreatments on the germination of *Irvingia gabonensis* seeds.

The analysis revealed little difference in seedling growth across pretreatments. Some seedlings, particularly those in treatments T1, T2, and T3, showed faster height growth. In contrast, those in treatments T4 and T5 showed greater diameter growth. These variations indicate that different pretreatments may promote specific aspects of seedling growth, although overall differences were modest.

Treatments T3 and T4 showed the best overall performance. Treatment T3, with a 96-h sulfuric acid soak followed by a water rinse, and treatment T4, with a 96-h water soak, not only resulted in high germination rates but also significant height and diameter growth. T3 seedlings showed faster height growth, while T4 seedlings showed greater diameter growth, demonstrating a balanced and significant improvement in overall performance.

On the other hand, treatments T1, T2, and T5 showed less uniform results. T1 and T2 seedlings had notable height growth but lower germination performance, while T5 promoted diameter growth without

reaching the high germination rates of T3 and T4.

Ultimately, these results offer promising prospects for improving propagation practices for this valuable species. Indeed, T3 and T4 treatments were particularly effective in optimizing both germination and seedling growth. A better understanding of the mechanisms underlying these performances and an assessment of the long-term impact of different pretreatments on seedling health could have positive ecological and socio-economic implications.

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