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Study of *Moringa Oleifera* growth under different pre-germination treatments

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ABSTRACT

Aim of study: The objective of this study was to optimize the germination traits (germination rate, Germination kinetics and Radicle length kinetics) of *Moringa Oleifera* seeds after different pre-germination treatments such soaking seeds for 24, 48, 72 and 96 hours and physico-chemical treatments of soaking the seeds in oxygenated water for 10min seed coat removal.

Area of study: The study was conducted in the laboratory on *M. Oleifera* Lam. cultivar originally from Tebelbella (400 km south of Bechar, Algeria).

Material and methods: Laboratory experiment was designed to investigate the influence of different pre-germination treatments such soaking and physico-chemical treatments on *Moringa Oleifera* germination traits.

Main results: Seed coat removal treatments gave the highest values on germination rate after 13 days of germination. Prolonged soaking treatment has a negative effect on seed germination, leading to a significant reduction in germination rates. Germination kinetics enhanced by both of chemical (H₂O₂) and of physical (seed coat removal) treatments. Seed coat removal exhibited the highest values for root elongation among all the treatments.

Keywords: *Moringa Oleifera*, Seed's Germination, soaking, physico-chemical treatments, Radicle length kinetics.

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INTRODUCTION

Moringa Oleifera, belongs to the family Moringaceae; originated from the sub-Himalayan region of northwest India and Pakistan, where it is now endemic, and it has been brought to various tropical countries (Gandji et al., 2018). The *Moringa Oleifera* Lam. tree is a species that is able to thrive in a multitude of tropical and subtropical locations (Emongor, 2019).

Moringa Oleifera commonly known as horseradish tree Lam. The name "horseradish" used to describe the taste of the ground root preparations. Additionally, it is known as the 'drumstick tree' due to its immature seedpod appearance and the 'ben oil tree' due to its seed-derived oils. In certain regions, immature seedpods are a common food choice, while fresh leaves commonly used as a primary food due to their high nutritional content (Kutawa et al., 2016).

M. Oleifera is a medium-sized tree that reaches a height of 3-15 m. It is a fast-growing tree and considered drought and heat tolerant. The plant grows best at an average daily range of 25-35°C, but can tolerate summer temperatures of up to 48°C for a limited period and winter frost (Ntsangani, et al., 2018).

Temperature, light, hormones, and enzymes are among the pre-treatment methods and processes that break seed dormancy. While there are various conventional methods that can break seed dormancy and improve seedling germination, they are often limited. Uneven seed hydration can lead to non-uniform germination due to hydropriming, even though it is widely used (Chandel et al., 2024).

The germination process of *Moringa Oleifera* seeds involves several factors that can influence its success. These factors include soaking and physico-chemical treatments, which play a significant role in preparing the seeds for germination. Understanding the effects of these treatments is vital for optimizing the germination rate and overall success of *Moringa Oleifera* cultivation (Gomaa et al., 2011).

Soaking the seeds before planting, have been found to enhance the germination process by initiating the metabolic activities within the seed. This process can help break dormancy and prepare the seed for germination under suitable conditions (Ashraf & Foolad, 2005, Tesfay et al., 2016). In fact, when components like substrate quantity, water availability, thermal properties and absence of physical obstacles for the emergence of some species are controlled, the seeds have better conditions for germination and emergence, and the seedlings better conditions for primer development (Yerima et al., 2016).

Physico-chemical treatments, on the other hand, involve the application of various physical and chemical methods to improve seed germination. These treatments include scarification, which

involves mechanically damaging the seed coat to promote water absorption, and seed priming, which involves exposing the seeds to specific solutions to improve water and nutrient uptake (Ardiarini et al., 2021). Therefore, this paper aims to assess the germination rate, Germination kinetics and Radicle length kinetics of *moringa Oleifera* seeds of different soaking and physico-chemical treatments.

MATERIAL AND METHODS

M. Oleifera Lam. cultivar originally from Tebelbella (400 km south of Bechar, Algeria) (Fig.01). The seeds were generously donated by a commercial farmer. The seeds used in this experiment collected in June 2022. Seeds collected directly from plants at the mature seed stage; they were stored in paper bags, labelled with the name of the species, the date and the place of collection, until they used.

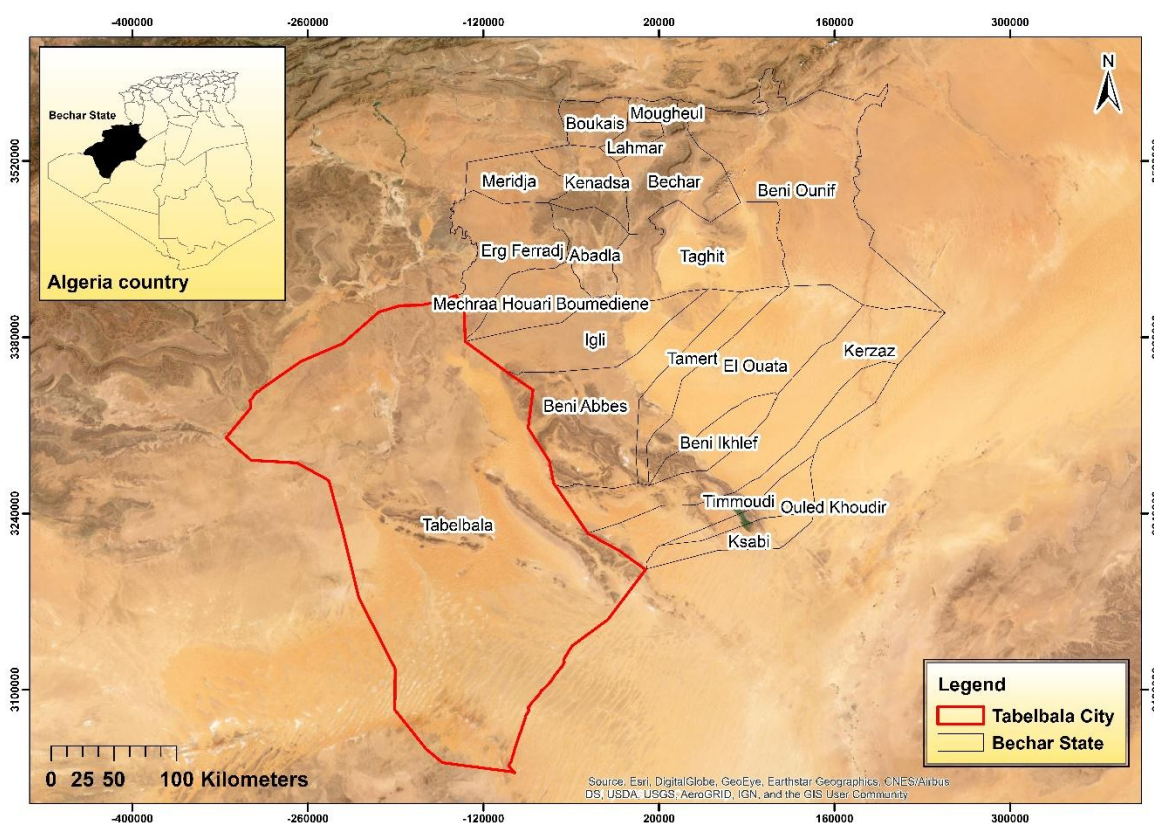


Figure 01: Localization of *moringa Oleifera* seeds sampling site of Tebelbella (400 km south of Bechar, Algeria)

Growing conditions and experimental set-up

Preparation of seeds for germination tests

The seeds used for germination tests were disinfected with 1% sodium hypochlorite for 3 minutes and then thoroughly rinsed with distilled water before starting the germination tests.

The tests were carried out with three replicates of five seeds per box for each treatment. A double layer of moistened filter paper was placed in each box.

Conduct of experiment

The experimental set-up adopted is a complete randomized block with seven treatments:

1. 24h treatment, soaking the seeds in distilled water for 24 hours.
2. 48h treatment, soaking the seeds in distilled water for 48 hours.
3. 72h treatment, soaking the seeds in distilled water for 72 hours.
4. 96h treatment, soaking the seeds in distilled water for 96 hours.
5. H₂O₂ treatment, soaking the seeds in oxygenated water for 10min.
6. Seed Coat Removal: the external coat of seed removed and seeds placed directly in the boxes.
7. Control treatment, seeds are placed directly in the boxes.

Seeds germinated in a growth chamber for 15 days at a temperature of 28°C, and a 16h /8h photoperiod (day/night respectively). As soon as the tip of the radicle appeared through the husks, we counted the germinated seeds regularly until the end of our experiments.

Measurements

Germination rate (%)

Based on the total number of seeds used (N_t), the percentage of germinating seeds (N_i) was calculated according to the equation: $GR\% = N_i \times 100 / N_t$

Germination kinetics

Germination kinetics is a germination curve that describes the germination process of the seed lot in question under very precise conditions. It most often represents the evolution of cumulative germination percentages as a function of time. kinetics are established from the cumulative rates of germinated seeds, i.e., the variation in germination rates as a function of time expressed in days under all the treatment conditions tested.

Germination curves give a complete idea of the development of germination in a batch of seeds placed under specific conditions.

Radicle length kinetics

The length of the radicle (embryonic root) measured using a digital caliper a digital caliper (BERENT BT4171 with 0.01 mm precision) at the tip of the radicle and the measurement displayed directly on the screen.

STATISTICAL PROCESSING

The results obtained were statistically processed using STATISTICA 8.0 software, with a safety threshold of 5%.

Results

Germination rate

Application of different soaking and physico-chemical treatments causes significant variations in germination rate. Thus, during the 1st day, the highest germination rates recorded after the application of seed coat removal treatments (53.3%). There was a gradual increase to 93.3% after 13 days of germination. Same evolution observed after 24h followed by 48h of soaking treatment. An increase around 65% in germination rate observed over a 72h soaking period, from day 7 to day 13. When soaking was carried out for 96 hours, a 45% reduction was observed, representing the minimum average decrease in germination rates on days 4 and 10 (Fig.02). This indicates that the prolonged soaking treatment has a negative effect on seed germination, leading to a significant reduction in germination rates.

After soaking seeds in H₂O₂ for 10 min, the germination rate starts to increase from day 4, gradually until it reaches a rate corresponding to 90% in 13 days.

When the seed coats were removed, there was an immediate increase in the germination rate from day one, reaching 100%. This rate is considerably higher than that of the control group, indicating that removing the seed coats has a significant positive effect on seed germination (Fig.03).

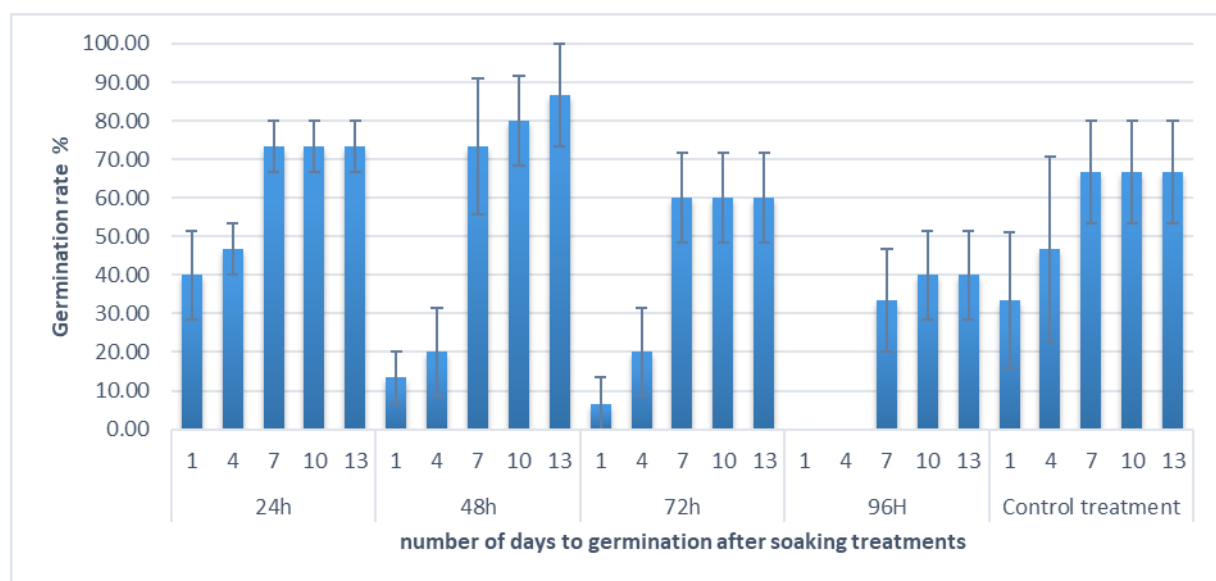


Figure 02: Germination rate after soaking treatment for *Moringa Oleifera* seeds.

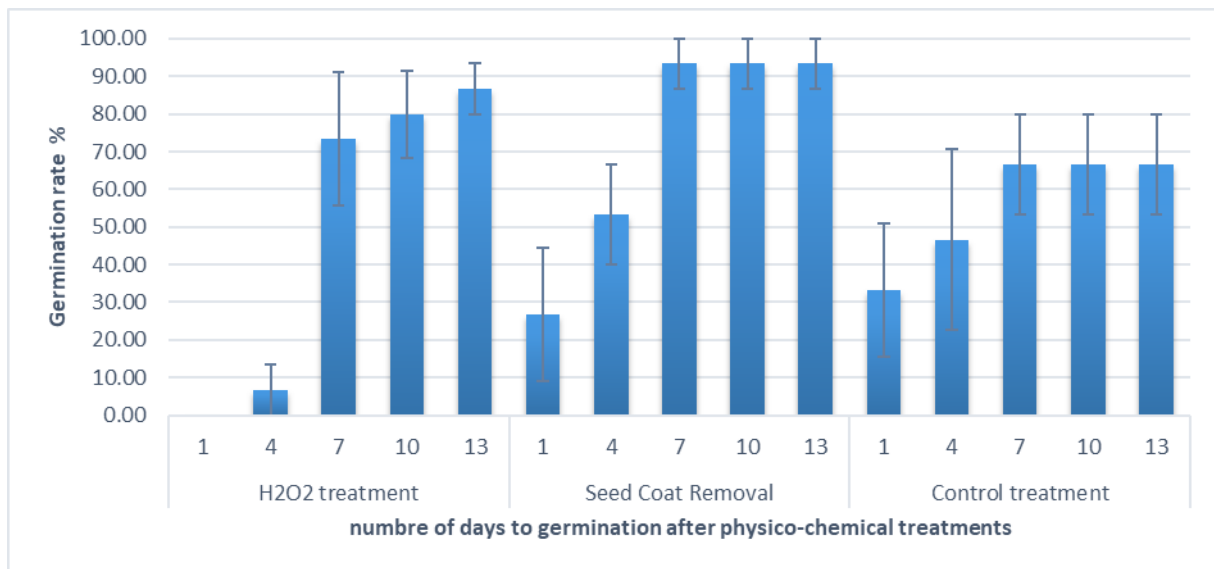


Figure 03: Germination rate after physico-chemical treatment for *Moringa Oleifera* seeds.

Germination kinetics:

Germination kinetics presented by the cumulative rates of *Moringa Oleifera* seeds under the effect of the soaking and physico-chemical treatments. Analysis of the results (Fig.04) shows that variations in seed germination rates strongly influenced by the different treatments applied. For germinated seeds, the germination kinetics curve showed the 02 classic germination phases: exponential acceleration followed by a phase corresponding to a complete stop of germination after reaching maximum germination capacity. Results indicate that both of chemical (H_2O_2) and of physical (seed coat removal) treatments enhance germination kinetic beyond control treatment. However, seed coat removal proves to be the most effective method, resulting in substantially higher germination rates across all time intervals.

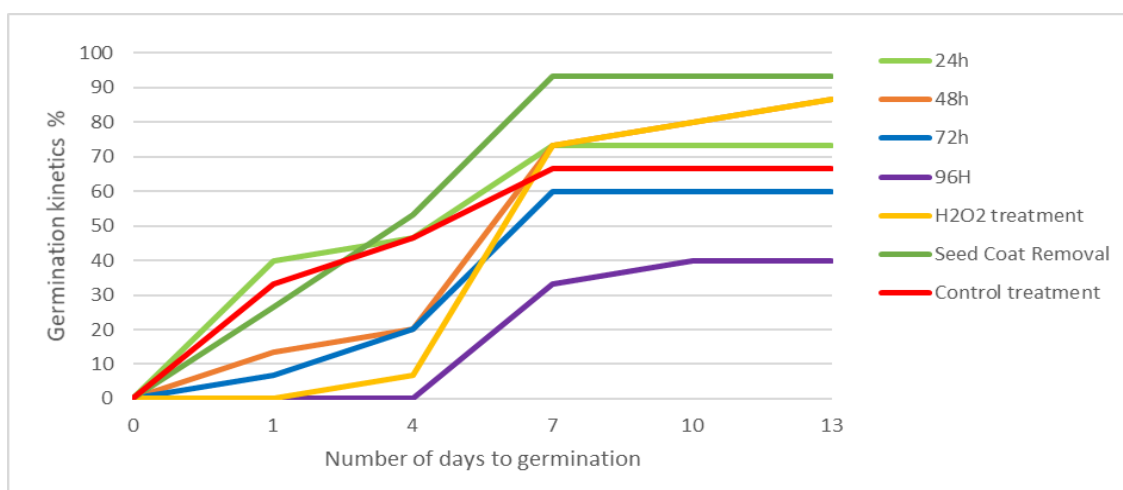


Figure 04: Germination kinetics after soaking treatments and physico-chemical treatments of *Moringa Oleifera* seeds.

Radicle length kinetics (mm)

Radicle length kinetics (mm) was studied under various soaking treatments for durations of 24, 48, 72 and 96 hours, as well as under physico-chemical treatments such as oxygenated water and seed coat removal

A slight increase in root elongation after soaking for 24h compared to the control. The values increased gradually from 0.06 cm to 1.19 cm, with a significant increase from day 11 onwards. After soaking for 48h, values fluctuate between 0.03 cm and 0.73 cm, indicating a consistent increase over time.

Application of chemical (H_2O_2) treatments resulted in a progressive increase in root elongation, with values ranging from 0.33 cm to 2.16 cm. Conversely, the treatment involving seed coat removal exhibited the highest values for root elongation among all the treatments. The values range from 0.82 cm to 8.33 cm, indicating a notable increase in root elongation with the removal of teguments (Fig.05).

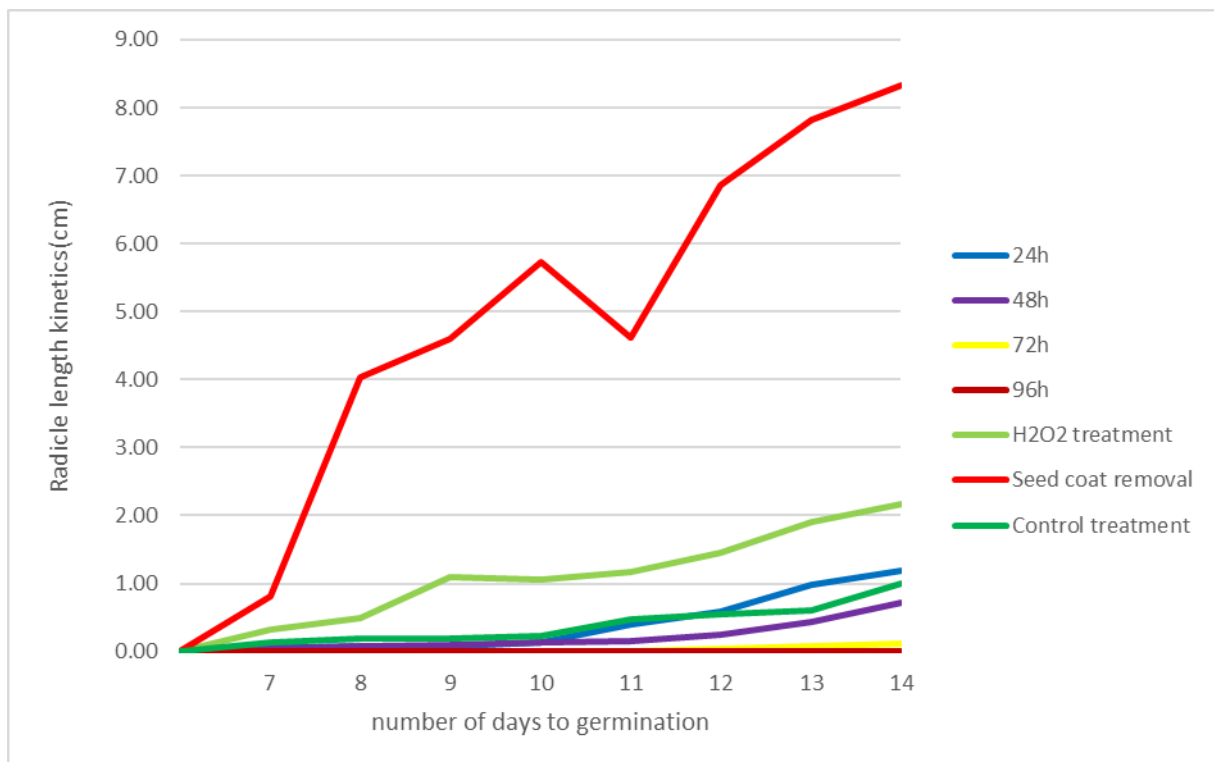


Figure 05: radicle length kinetics (cm) after soaking treatments and physico-chemical treatments of *Moringa Oleifera* seeds.

Discussion

The present study investigates the impact of soaking treatments and physico-chemical treatments on the germination of *Moringa Oleifera*. In Algeria, this species is little known and there is much to be done to popularize it among foresters and professionals in the sector.

As a reforestation species, it is important to know at least a minimum amount about the seed of the species and the treatments that are likely to improve its germination capacity. Moringa seeds have seed coats that cause inhibition problems, making germination in nurseries and direct sowing uncertain. Certain treatments can overcome this barrier (Bendifallah et al., 2018; Mansouri, 2014).

Whether a viable seed germinates depends on many factors, including what is present in the seed's environment. To begin with, the chemical environment must be suitable. Water must be present, oxygen must be present and the seed must be able to breathe. The indoor environment must also be present (Bewley, 1994; Finch-Savage et al., 2006).

Several variables influence the germination of seeds, including their age. In particular, there have been reports indicating that the viability decreases when the planting height reaches 1,200 meters above sea level.

Pre-germinative treatments facilitate the breakdown of the latent phase in moringa seeds, consequently promoting fast emergence and consistent growth of seedlings. Soaking has been one of the few technologies that have been examined for exposure. The results suggest that soaking helps with germination and even enhances plant resilience to stress (Núñez-Gastélum et al., 2023).

The results demonstrated the significance of the time elapsed since the commencement of the experiment and the impact of the various pre-treatments on germination traits. The physicochemical treatments, in particular the removal of seed coats and the application of hydrogen peroxide-based treatments, were found to be conducive to germination by creating an environment conducive to the development of seed embryos (Gbenou et al., 2021; Nwangburuka et al., 2012).

The analysis of germination kinetics revealed significant variations in germination rates as a function of treatment duration and experimental conditions.

A considerable number of studies have been conducted with the objective of identifying the optimal conditions for the germination of this plant. This is exemplified by the work of Ndong-On-Mebi (2014). This study examines the effects of temperature, light and pre-germination treatments on the germination of *Moringa Oleifera* seeds. Short-term treatments, such as the 24-hour treatment and the H₂O₂ treatment, initially exhibited lower germination rates, but these gradually increased over time. In contrast, the seed coat removal treatment demonstrated high germination rates from the outset, thereby demonstrating the positive effect of this practice on germination.

Extended durations of treatment, such as 48 hours, 72 hours, and 96 hours, initially resulted in decreased germination rates, which subsequently improved over time. Nevertheless, these rates consistently remained lower than those observed in other treatments, potentially exerting an inhibitory influence on germination.

In terms of root elongation, the 24-hour and 48-hour soaking treatments, as well as the physicochemical treatments, found to promote root elongation. However, there was no noticeable impact found for the 72-hour and 96-hour treatments, underscoring the significance of treatment duration in promoting root growth.

CONCLUSION

In conclusion, this study highlights the importance of treatment time, physicochemical treatments and the environment on germination, root elongation and the physiological responses of *Moringa Oleifera* to water stress. These results contribute to a better understanding of the germination and growth mechanisms of this plant, which could be useful for its use in agronomic and environmental applications.

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