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Optimizing Hydropriming Duration for Enhanced Germination, Emergence and Seedling Growth of *Ocimum gratissimum*: A Path to Sustainable Production in Nigeria

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ABSTRACT

Background and Objective: Cultivation of scent leaf (*Ocimum gratissimum*) is a valuable crop with culinary, medicinal and ornamental significance in Nigeria, is constrained by low germination and seedling vigor due to the inhibitory effect of unesterified galacturonic acid in the mucilaginous layer in the seed coat. Sustainable production of this important horticultural crop requires improving seed germination, emergence and seedling growth observed in this study. **Materials and Methods:** Thus, an experiment was conducted in the Department of Horticultural Technology, Enugu State Polytechnic Iwollo, Enugu State, Nigeria, to determine the optimal hydro-priming duration for improved seed germination, emergence and seedling growth of *Ocimum gratissimum*. The treatments included 0, 6, 12, 18 and 24 hrs of hydro-priming. A completely Randomized Design with three replications was used for the experiment. Data on days to 50% germination, mean germination time, germination percentage, mean emergence time, emergence percentage, seedling height and number of leaves per seedling were collected and subjected to Analysis of Variance (ANOVA) at a 5% significant level using GenStat software. Treatment means were separated using Fisher's least significant difference at 0.05 probability level. **Results:** The results showed that hydro-priming significantly ($p<0.05$) increased germination and emergence percentages, seedling height and number of leaves per seedling, with 18 hrs being the optimal duration. Hydro-priming also significantly decreased days to 50% germination, mean germination time and mean emergence time with 18 h also being the optimal duration. **Conclusion:** It could be concluded that hydropriming is an important seed treatment for improved germination, emergence and seedling growth of *Ocimum gratissimum*. Hydropriming for 18 hrs is therefore recommended for enhanced *Ocimum gratissimum* cultivation and sustainable production of the crop in Nigeria.

KEYWORDS

Emergence, germination, hydropriming, scent leaf (*Ocimum gratissimum*), seedling growth

INTRODUCTION

With soaring food insecurity in Sub-Saharan Africa and Nigeria in particular, sustaining the production of indigenous leafy vegetables such as scent leaf (*Ocimum gratissimum* L.) to ameliorate the food crisis requires addressing its seed germination limitations. Scent leaf (*Ocimum gratissimum* L.) is a valuable

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horticultural crop esteemed globally for its culinary, medicinal and ornamental significance. It is an herbaceous plant that belongs to the Labiatae family. Native to tropical regions, it is widely used in Nigeria, where it is known by various names in different languages, such as Ahuji or nchanwu in Igbo, Efinrin in Yoruba and Daidoya in Hausa[1]. Scent leaf is a prized ingredient in traditional Nigerian cuisine, particularly in the southeast region, where it is used to prepare delicacies like pepper soup and ofe akwu (Banga soup). The leaves contain useful nutrients necessary for human and livestock growth, including antioxidants[2-4]. The essential oils extracted from scent leaf have also shown potential as natural pesticides and insect repellents[5]. Additionally, scent leaf has been used in traditional medicine to treat various ailments, including epilepsy, fever, diarrhea, mental illness and fungal infections[6-8].

Despite its numerous benefits, commercial cultivation of scent leaf in Nigeria is limited, due to poor seed germination among other factors. Local farmers typically grow it as a perennial crop in small quantities, mainly at the back of their compounds and along land boundaries, leading to high market prices (approximately N3000 per kg presently). Research has shown that small seeds of the Ocimum genus, including scent leaf, are characterized by a mucilaginous layer containing high concentrations of unesterified galacturonic acid, which inhibits germination[9,10]. Propagating scent leaf from seed is challenging, resulting in reliance on established stands that grow through natural dispersal[11]. As a warm-season crop, optimal seed germination, emergence and seedling growth are crucial for successful cultivation.

Seed priming is a technique used to enhance seed germination and crop establishment[12]. By controlling seed hydration, priming activates pre-germinative physiological and chemical changes that prepare seeds for rapid and uniform emergence[13]. During priming, seeds are soaked in different solutions with high osmotic potential, allowing pre-germinative metabolic activities to proceed while preventing radical protrusion. Seeds are then dried back to their original moisture level before they are later sown[14]. Seed priming approaches include hormopriming (GA_3 and IAA), halopriming ($MgSO_4$ and KNO_3), osmopriming (H_2O_2) and hydropriming (H_2O)[15]. These approaches have been shown to improve seedling performance and stress tolerance[13.] Among these priming approaches, hydro-priming is the easiest and cheapest, making it an easy-to-adopt approach for small-scale farmers, who are the major horticultural crop producers in Nigeria and Sub-Saharan Africa at large[16]. Hydro-priming has been reported to improve germination and emergence of some small-seeded horticultural crops[14-17]. However, the duration of hydropriming significantly impacts its effectiveness[17]. There is a dearth of information on the optimal hydropriming duration for improved seed germination, emergence and seedling growth in *Ocimum gratissimum*. Understanding the optimal hydropriming duration for enhanced germination and seedlings emergence in this crop is crucial for sustainable production of this important horticultural leafy vegetable crop in Nigeria. Thus, the study was aimed to determine the optimal hydropriming duration for improved germination, emergence and seedling growth in scent leaf (*Ocimum gratissimum*). The knowledge will enable farmers to adopt a cost-effective seed treatment for sustainable production of the crop.

MATERIALS AND METHODS

Experimental site: The experiment was conducted in the laboratory and nursery of the Department of Horticultural Technology, Enugu State Polytechnic, Iwollo, Southeast Nigeria in 2023. The study area is located within Latitude 06° 16.834'N and Longitude 07^0 16.834'E.

Source of seeds and seed sterilization: The scent leaf (*Ocimum gratissimum*) seeds used for the study were sourced from Enugu State Polytechnic, Iwollo, Southeast Nigeria. The seeds were sterilized with a 1.0% solution of Sodium Hypochlorite (NaOCl) to kill microorganisms that could influence the results. The seeds were soaked in the solution for five minutes and were washed thoroughly with distilled water to remove the solution.

Hydropriming treatments and experimental design: The sterilized seeds were soaked in distilled water in Petri dishes for 6, 12, 18 and 24 hrs at room temperature and dried back to their original state using filter paper. Some seeds were left un-primed to serve as control. Completely Randomized Design (CRD) with three replications was used for the experiment. The treatments were 0 (control), 6, 12, 18 and 24 hrs of hydropriming.

Germination study: For the Germination experiment, three Petri dishes containing two layers of filter papers were moistened with 10 mLl of distilled water and 30 seeds were plated in each for each treatment. The Petri dishes were observed every 24 hrs for 14 days and germinated seeds were counted. Germination was considered to have taken place when the radicles were 2 mm long[18]. Data were collected on days to 50% germination, mean germination time and germination percentage. Days to 50% germination (D50%G) was determined by counting the number of days it took half of the seeds to germinate.

Mean germination time (MGT) was calculated using the formula:

$$MGT = \frac{\sum(ni \times di)}{N}$$

Where

I = Individual observations
ni = Number of seeds that germinated on the ith day
di = Number of days counted from the sowing day to the date the datum (i) was collected
N = Total number of seeds germinated at the end of the experiment[19]

Germination percentage (GP) was evaluated by counting the number of germinated seeds at the end of the test and applying the formula:

$$GP = \frac{\text{Germinated seeds}}{\text{Total seeds}} \times 100$$

Emergence and seedling growth study: The 3 polypots containing 2 kg of nursery medium composed of topsoil, well cured poultry manure and sand in the ratio of 3:2:1, respectively were used for the experiment for each treatment. The experiment was set up in the nursery and ten seeds from each of the treatments were sown in the nursery medium at a depth of 2.5cm and watered lightly. Subsequently, each polypot was watered with 50 mL of water daily; in the morning and evening. The polypots were observed daily for 14 days and emerged seedlings were counted and recorded. Seedling emergence was considered when the plumule emerged from the soil. Data were collected on Mean Emergence Time (MET) and Emergence Percentage (EP).

Mean emergence time (MET) was calculated using the formula:

$$MET = \frac{\sum(ni \times di)}{N}$$

Where

I = Individual observations
ni = Number of seeds that emerged on the ith day
di = Number of days counted from the sowing day to the date the datum (i) was collected
N = Total number of seeds that emerged at the end of the experiment[19]

Emergence percentage (EP) was evaluated by counting the number of emerged seedlings at the end of the emergence test and applying the formula:

$$EP = \frac{\text{Emerged seedlings}}{\text{Total seeds sown}} \times 100$$

For the seedlings growth experiment, two healthy seedlings from the seedlings that emerged in each polypot were used. The seedlings were watered as when due. Normal nursery practices were taken to raise the seedlings in the nursery. At 4 weeks after sowing, data were collected on seedling height and number of leaves per seedling. Seedling heights were determined by measuring the seedlings from the base of the stands to the apical bud using measuring tape. Direct counting of the leaves per seedling was done to get the number of leaves per seedling.

Data analysis: The data collected from germination, emergence and seedling growth experiments were subjected to Analysis of Variance (ANOVA) for Completely Randomized Design. Means that showed significant differences were separated using Fisher's Least Significant Difference (F-LSD) at 0.05 probability level.

RESULTS AND DISCUSSION

Effect of hydropriming duration on germination of *Ocimum gratissimum* seeds: The results of the statistical analysis as presented in Table 1, showed significant differences ($p<0.05$) in mean germination time, days to 50% germination and germination percentage of Scent leaf (*Ocimum gratissimum*) among the treatments (0, 6, 12, 18 and 24 hrs hydropriming). The mean germination time decreased progressively from 0 to 18 hrs hydropriming duration and increased slightly at 24 hrs treatment. The 18 hrs treatment had the shortest mean germination time (3.08 days) followed by 12 hrs (4.45 days) and the longest time was in 0 hrs (control) (8.01 days). All the hydropriming seeds performed better than the unprimed seeds. The results indicated that hydropriming duration has a significant impact on the mean germination time of *Ocimum gratissimum* seeds. The shortest days to 50% germination were observed in 18 hrs (3.00 days), followed by 24 and 12 hrs and (4.67 and 4.33 days, respectively which are statistically at par). The 6 hrs treatment recorded 6.67 days while the control treatment had the longest days (8.00). In germination percentage, 18 hrs treatment recorded the highest value (84%), while the lowest value was observed in the control (49.67%).

The decrease in mean germination time and days to 50% germination from 0 to 18 hrs treatments observed in the study suggests that longer treatment durations below the threshold accelerate seed germination and the optimum duration for fastest germination is 18 hrs. The increase in the values observed in the 24 hrs treatment indicates a negative impact of extended treatment duration beyond the threshold. This could also imply to a similar trend of performance observed in germination percentage.

Table 1: Effect of hydro-priming duration on mean germination time, days to 50% germination and germination percentage of scent leaf (*Ocimum gratissimum* L.) seeds

Hydro-priming duration	D50%G	GP	MGT
6 hrs	6.67	55.67	6.73
12 hrs	4.33	72.33	4.45
18 hrs	3.00	84.00	3.08
24 hrs	4.67	69.00	4.71
Control	8.00	49.67	8.01
$F\text{-}LSD_{0.05}$	0.965	6.12	0.04
CV (%)	26.83	26.67	33.00
S.E.	0.37	3.64	1.03
Grand mean	5.334	53.00	5.40

$F\text{-}LSD_{0.05}$: Fisher's least significant difference at 5% probability level, CV: Coefficient of variation, S.E.: Standard error, MGT: Mean germination time, GP: Germination percentage and D50%G: Days to 50% germination

Table 2: Effect of hydro-priming duration on mean emergence time, emergence percentage, seedling height and number of leaves per seedling of scent leaf (*Ocimum gratissimum* L.)

Hydro-priming duration	MET	EP	Seedling height (cm)	Number of leavers per seedling
6 hrs	7.33	55.33	5.67	5.33
12 hrs	6.33	61.33	7.67	6.67
18 hrs	5.67	70.67	9.67	7.67
24 hrs	6.33	62.33	7.33	6.33
Control	11.67	43.67	5.33	5.00
F-$LSD_{0.05}$	0.16	4.58	1.04	0.73
CV (%)	8.59	15.00	23.6	19.84
S.E.	0.03	3.77	1.00	0.32
Grand mean	7.07	56.46	7.00	6.20

F-$LSD_{0.05}$: Fisher's least significant difference at 5% probability level, CV: Coefficient of variation, S.E.: Standard error, MET: Mean emergence time and EP: Emergence percentage

The findings of the study are consistent with some previous research on the effects of hydropriming on seed germination. Hydropriming has been shown to significantly improve germination speed, synchrony and seedling vigor in faba bean seeds[20]. The benefits of hydropriming on seed performance have also been highlighted, supporting the finding that hydropriming can substantially reduce germination time[13]. Furthermore, hydropriming has been demonstrated to improve drought escape and early recovery in upland rice[21].

The enhanced seed germination observed could be due to hormonal regulations and seed coat modification as a result of hydropriming. Hydropriming enhances seed performance by improving water uptake, which is essential for initiating metabolic activities necessary for germination. The findings of the study revealed that optimizing hydropriming duration is crucial for enhancing the germination efficiency of *Ocimum gratissimum* seeds. Implementing an 18 hrs hydropriming could be highly beneficial for sustainable cultivation of the crop.

Effect of hydropriming duration on emergence and growth of *Ocimum gratissimum* seedlings: The results of the analysis of variance as presented in Table 2 showed significant differences ($p<0.05$) in mean emergence time, emergence percentage, seedlings height and number of leaves of *Ocimum gratissimum* seedlings among the treatments. The treatment with 18 hrs of hydropriming exhibited the lowest mean emergence time (5.56 days), followed by 12 and 24 hrs which had 6.33 days each. The highest value was observed in control (11.67 days). The highest emergence percentage of scent leaf seedlings was observed in 18 hrs (70.67%), followed by 24 hrs (62.33%), 12 hrs (61.33%), 6 hrs (55.33%) and the lowest was in control (43.67%). Hydropriming above 18 hrs resulted in a decline in emergence percentage. In seedling height, 18 hrs treatment had the highest mean seedling height (9.67 cm), followed by 12 hrs (7.67 cm), 24 hrs (7.33 cm) and 6 hrs (5.67 cm). All the hydro-primed seeds performed better than the control which had (5.33 cm). In the number of leaves per seedling, 18 hrs had a mean leaf number of 7.67 leaves, followed by 12 hrs (6.67 leaves), 24 hrs (6.33 leaves), 6 hrs (5.33 leaves) and control (5.00 leaves). All the treatments showed significantly improved performance in all the emergence and growth parameters evaluated compared to control. Also, there was a progressive increase in the performance of the seedlings with an increase in hydropriming duration in all the parameters with 18 hrs being the optimal duration. This improvement is likely due to enhanced water uptake and activation of metabolic processes critical for emergence and seedling vigor. There was a decline in the performance of seedlings in the 24 hrs treatment suggesting that the threshold was exceeded.

These findings align with the findings of Nakao *et al.*[21] who reported that hydropriming for different durations significantly improved emergence of sweet basil (*Ocimum basilicum*) seedlings compared to unprimed seeds and opined that hydropriming improves seed performance by enhancing water uptake, which is essential for initiating metabolic activities necessary for seed germination and seedling

emergence[14]. Similarly, Tanwar *et al.* reported that optimizing hydropriming conditions, including soaking time, significantly improved seed performance in wheat[22]. Khalid *et al*[13]. and Shukla *et al.*[23] highlighted the benefits of hydropriming on seed performance. The findings were also in agreement with the findings of Adhikari *et al.*[16]; the positive effect of hydropriming has been attributed to improved water imbibition, which enhances enzyme activation, translocation and the use of stored food materials. According to Pandita *et al.*[24], hydropriming aids water absorption, softening the seed coat and promoting the growth of the seed embryo. These results are consistent with the findings of TS and Arivazhagan who observed that hydro-primed seeds showed a shorter germination period, faster emergence and more vigorous seedlings[25]. Research has also shown that each crop cultivar has an optimal soaking duration, which should not exceed a critical threshold[25-31].

The optimal soaking duration in this study was 18 hrs beyond which no additional benefit was observed. Adhikari *et al.* emphasized that the knowledge of suitable priming duration is critical for optimal impact[16]. Hydropriming scent leaf seeds for 18 hrs before planting has been shown in this study to improve germination, emergence and seedling growth of scent leaf (*Ocimum gratissimum*).

CONCLUSION

The findings of this study showed that hydro-priming enhanced seed germination, emergence and seedling growth of scent leaf (*Ocimum gratissimum*) with 18 hrs hydro-priming being the optimal duration. These findings suggest that 18 hrs hydro-priming is a simple and cost-effective approach for improving scent leaf cultivation. Recommendations from this study can inform horticultural practices and future research to enhance yield and sustainable cultivation of this important horticultural crop.

SIGNIFICANCE STATEMENT

This study addresses the critical challenge of poor seed germination in *Ocimum gratissimum* (scent leaf), a valuable horticultural crop in Nigeria. Our research revealed that optimizimg hydropriming duration significantly enhanced germination, emergence and seedling growth in *Ocimum gratissimum* cultivation. The 18 hours was found to be the optimal hydropriming duration and this provides a cost-effective solution for enhanced germination, emergence and seedling growth in *Ocimum gratissimum* cultivation. The findings of this study have significant implications for sustainable production of the crop in Nigeria and will contribute to improved food security, enhanced economy for small-scale farmers and the promotion of indigenous leafy vegetable crop cultivation. This research contributes to the advancement of horticultural practices and has a broader implications for agricultural development in Sub-Saharan Africa.

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