

Influence of priming treatments on seed germination and growth characteristics of *Moringa oleifera* seedlings under laboratory and greenhouse conditions

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Abstract

Seed priming is the treatment of seeds with various priming substances to improve germination and seedling quality, and plant growth. The procedure entails advancing the seed to an equivalent stage of the germination process to achieve rapid and uniform emergence when planted, which is required in the case of *Moringa oleifera* to reduce the damage of ageing seeds, stimulate their performance, and overcome uneven germination rate due to seedcoat hardening, that leads to wide variation in terms of commercial qualities in the planting field. Therefore, a series of seed priming experiments were conducted to determine the optimal methods of priming treatments that maximise *Moringa oleifera* total germination, germination rate, and seedling vigour. *Moringa oleifera* seeds were treated with hydropriming, osmopriming (KNO₃), and nutripriming (*SeedActivator*) for 4, 8, and 12 hrs. Germinability was improved by priming treatments. Results indicated that the highest germination percentage was recorded on seeds subjected to hydropriming (63.25%), followed by nutripriming (61.75%) and osmopriming (60%) with the exact duration of 4 hrs soaking time. All pre-germination treatments have significantly early mean germination days (MGT) compared to the unprimed seeds. In the seed pouch study, seeds treated with hydropriming for 4 hrs had shown the best results to improve roots growth in total root length (cm), root volume (cm³), the number of forks, and the number of crossings with 240.52 cm vs. 163.64 cm, 0.47 cm³ vs. 0.29 cm³, 517.80 vs. 271.47 and 59.20 vs. 28.53 respectively when compared to unprimed seeds. The positive effect of priming can also be seen in the seedling growth of *M. oleifera* grown in a pot with two different growing media.

1. Introduction

Moringa oleifera, also known as horseradish, drumstick tree, ben oil tree, kelor, or merunggai, is the most neglected multipurpose tropical crop. For the most part, this neglected crop does not require large inputs. It is a fast-growing perennial soft-wooded tree that tolerates drought and grows well in poor soil (Palada and Chang, 2003). The tree is well-known for its suitability as a source of nutrients for both people and livestock. It is a good source of macronutrients and micronutrients necessary for proper diet and nutrition daily (Anwar *et al.*, 2007). Apart from being used as food for humans and animals, by-products of *M. oleifera* are used for food fortification, food supplements (vitamins), and cosmetics (soap, shampoo, and skin lotion). Recent interest in this perennial leafy vegetable has increased due to its low cost and high nutritional value. Therefore, more

exploration is needed to study the most favourable conditions for cultivating *M. oleifera*. The tree commonly reproduces through seed and stem cuttings. However cutting propagation is unreliable since the stem cuttings must be longer and thicker (Fuglie and Sreeja, 2001), thus resulting in a high mortality rate for the mother plant (Ramachandran *et al.*, 1980). As a result, seeds continue to be the most favourable choice for the *M. oleifera* multiplication method since they are simple, inexpensive and produce many plants (Fuglie, 2001; Palada and Chang, 2003; Radovich, 2011). Germination of *M. oleifera* occurs within 5 - 30 days, depending on the age of the seed, soil or media type, and pre-treatment method used, which might include: cracking the shells, soaking seeds with shells, dehulling seeds, and soaking seeds (Quintin, 2009). Germination of *M. oleifera* has been low due to unfavourable environmental conditions

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such as changes in soil chemistry and drought stress, among other things (Urva *et al.*, 2017). Furthermore, moringa seeds deteriorate more rapidly during storage, which reduces the quality of the seed. Studies have shown that greater dehydration and storage reduce seed viability (Bonner, 1990; Kioko *et al.*, 2006). Ageing is manifested as a reduction in germination percentage and those seeds that germinate weak seedlings (Veselova and Veselovsky, 2003). The factors that determine the rate of this 'ageing' are the temperature and moisture content at which seeds are stored, affecting seed quality. Seeds deteriorate and lose their germinability during periods of prolonged ageing (Ansari and Sharif Zadeh, 2013; Seiadat *et al.*, 2012). Many seed priming treatments have been used to reduce the damage of ageing and stimulate their performance in many crops (Basra *et al.*, 2003; Farooq *et al.*, 2009; Abdolahi *et al.*, 2012). Thus, priming enables repair processes to recover from storage damage and the previously described benefits. Seed priming has found beneficial effects on many field crops. Seed priming is a technique of regulating the germination process by managing the temperature and seed moisture content; the seed is taken through the first biochemical processes within the initial stages of germination. Germination occurs in three phases (I) imbibition, (II) 'pre-germination,' and (III) emergence (Bewley, 1997). During Phase II, the biochemical processes are activated and will eventually start the germination in Phase III, where (roots and hypocotyls emerge from the seed). The procedure of seed priming is known to trigger 'pre-germination.' The seed is taken partway through Phase II and then dried, before radicle emergence, under suitable temperature and moisture. Although the germination is not completed, metabolic activities that prepare seeds for radicle protrusion may be initiated during priming (Heydecker *et al.*, 1973; Passam and Kakouriotis, 1994). Phase III can continue, and germination occurs in a much shorter time. Hydropriming, halopriming, osmopriming, matrix priming, hormopriming, nutripriming, nanoparticle priming, heat/cold priming, and other priming strategies have been shown to increase crop germination and seedling establishment (Rashid *et al.*, 2006; Yu-Jie *et al.*, 2009; Sedghi *et al.*, 2010; Imran *et al.*, 2013; Singh *et al.*, 2014; Baier *et al.*, 2019). Seed priming is one of the most effective, feasible, and quick strategies to enhance seed vigour and germination synchronisation, which results in uniform stand and improved yield (Du and Tuong, 2002; Kaya *et al.*, 2006). Some authors have shown that germination and emergence of moringa seeds are enhanced with pre-sowing treatment (Tedonkeng *et al.*, 2004; Afzal *et al.*, 2008; Nouman, Siddiqui, Basra, Afzal *et al.*, 2012; Padilla *et al.*, 2012; Materechera, 2017; El-Absy *et al.*, 2019). It was recorded that 12 hrs

of hydropriming increased the germination of moringa seeds in the western highlands of Cameroon (Tedonkeng *et al.*, 2004). While soaking in water for 24 hours before sowing, incubate their germination on the field in Cuba (Padilla *et al.*, 2012). Nouman, Siddiqui, Basra, Khan *et al.* (2012) found that hydropriming, MLE priming, and matrix priming improved moringa seedlings' emergence and seedling vigour compared to unprimed seeds. Numerous factors can affect seed germination and vigour, including abiotic stresses such as saline, which can impair germination and initial plant establishment. The high content of salts in the soil and waters used in irrigation can compromise crop development. Nouman, Siddiqui, Basra, Khan *et al.* (2012) and Nóbrega *et al.* (2021) reported that moringa was tolerant to the effect of salinity. Santos *et al.* (2011) found that a salinity increase results in a decrease in germinability and delayed germination rate of *M. oleifera*. Meanwhile, osmopriming with NaCl at higher concentrations showed negative results in germination and emergence in moringa (Elhag and Abdalla, 2014; Fatima *et al.*, 2018). Eghobor *et al.* (2015) recorded that gibberellic acid at concentrations of 7.5 mg/L soaked for 24 hrs gave a higher germination percentage of moringa than untreated seeds. Seed priming is influenced by many factors such as water potentiality of the priming agent, priming agent duration, temperature, seed vigour, and seed storage condition (Ghassemi-Golezani *et al.*, 2010). There is a need to overcome germination issues in *M. oleifera* seeds for local climate conditions. To understand the responses of different pre-treatment to *M. oleifera* seeds, it may be useful to investigate the germination stage and seedling development changes. Water, KNO₃, and nutrient elements were chosen as different priming reagents in this work to evaluate the total germination, germination rate, and seedling response to various priming reagents and priming duration.

2. Materials and methods

2.1 Plant materials and seed treatments

Moringa oleifera seeds stored for three months before sowing was bought from a local shop in Selangor. All seeds were sterilised for 25 mins with 1% (w/v) sodium hypochlorite, then washed twice with distilled water (Fotouo-M, 2015). The sterile seeds were treated with hydropriming (distilled water), osmopriming (KNO₃) at 1% (w/v), and nutripriming (Humic acid and TE) at 1% (w/v) for 4, 8, and 12 hrs, respectively, with unprimed seeds, served as the control treatment. The priming solutions were 29°C in temperature. Before priming application, seeds were weighed. Following treatment, seeds were rinsed with distilled water, dried on paper towels, and ventilated until they returned to their original weight at room temperature.

2.2 Seed germination in the laboratory

For the germination test, 20 treated seeds for each treatment were placed in a petri dish with double layer Whatman No. 1 filter paper in a plant growth chamber (Hettich Model PRC1200SL) at 12 hrs photoperiod, 12 hrs light (30°C)/12 hrs dark (20°C), and 60% relative humidity (RH). The experiment was set up in a completely randomised design (CRD), with four replicates for each treatment. Seeds were moistened whenever necessary. Seed germination was observed and counted daily for up to 14 days. Once the radicle appeared, the seed was considered germinated. After each count, germinated seeds were removed from the petri dish. The germination percentage (G%) was calculated with the following formula:

$$G\% = \frac{\text{Number of germinated seeds}}{\text{Total number of seeds}} \times 100$$

The mean germination time (MGT) was estimated using the Ellis and Roberts equation (1981):

$$MGT = \frac{\sum(D * n)}{\sum n}$$

where n is the number of seeds that germinates on day D, and D is the number of days counted from the beginning of the germination test.

The time to 50% germination (T50) was determined using Coolbear *et al.* (1984), as modified by Farooq *et al.* (2005):

$$T50 = \frac{t_i \left\{ \left(\frac{N}{2} \right) - n_i \right\} (t_i - t_j)}{n_i - n_j}$$

Where N is the final number of emergence, and n_i and n_j are the cumulative numbers of seeds germinated by adjacent counts at times, t_i and t_j , respectively, when $n_i < N/2 < n_j$.

The germination index (GI) was calculated as described by the Association of Official Seed Analysts (1983) by the following formula:

$$GI = \frac{\text{No. of germinated seeds}}{\text{Days of first count}} + \dots + \frac{\text{No. of germinated seeds}}{\text{Days of final count}}$$

2.3 Root pouch assays

The priming treated seed germinated in a petri dish before being placed in a pouch with a tweezer by inserting the seeds with radicles into a pre-made slit growing pouch (PhytoTC.com). Each growing pouch had four seeds evenly placed along the seed line for each treatment and arranged in a completely randomised design (CRD) with six replications. The pouches were stacked vertically in a rack in a plant growth chamber (Hettich Model PRC1200SL) for 14 days with a photoperiod of 12 hrs light (30°C)/12 hrs dark (20°C) at

60% RH. During growing, root systems were shaded from light by covering each side of the pouch with black cardboard. At the end of the experiment, root morphological parameters such as total length, volume, number of tips, number of forks, and number of crossings were measured. The acquired root images were analysed using WinRHIZO Pro 2007b (Regent Instruments Inc., Quebec, Canada).

2.4 Greenhouse trials

Seedling emergence and seedling growth of *M. oleifera* were studied under greenhouse conditions with two different sowing media. The priming treated seeds were sown in non-woven pots (15 cm × 7 cm) filled with a mixture of sand: peat moss (9:1), and topsoil: coco coir dusk: peat moss (5:2:1) (volume by volume; v/v). Before seeding, the substrates were well mixed with the organic fertiliser BioRichar 5% N: 5% P₂O₅: 5% K₂O at a rate of 4 g per pot, equivalent to 0.2 t ha⁻¹. The experimental design was arranged in a randomised complete block design (RCBD) with two factors and three replications. The factors were the priming treatments and growing media. Twenty seeds were used per treatment per replication, with a single seed sown in each pot. The growth was observed daily for over 14 days. The seedling was watered daily throughout this period, which wetted the growing medium to field capacity. The morphological growth parameters measured were seedling height (cm), stem girth (mm), number of leaflets, and total leaf area. The total leaf area per plant was measured in cm² by destructive sampling using a leaf area meter (LI-3100, Li-COR Inc., USA).

2.5 Statistical analysis

All data were analyzed using analysis of variance (ANOVA), and the mean was separated by Duncan Multiple Range at $P \leq 0.05$ (SAS Institute, Version 9.4).

3. Results and discussion

3.1 Seed germination

The effects of various priming treatments on seed germination were first studied under laboratory conditions. Seed priming improved germination percentage, mean germination time, time to reach 50% germination, and germination index. The soaking procedure partially moistened the seeds, allowing the metabolic process of germination to begin. The seed priming approach has proven effective in increasing the rate of germination. Regardless of priming substrates, the soaking time of 4 hrs was the best for the germination of *M. oleifera*. Under this soaking duration, germination percentage reached 63%, 61%, and 60% for hydropriming, nutrpriming, and osmopriming,

respectively. Figure 1 illustrates germination curves that show the germination rate of *M. oleifera* seed under various priming applications and durations. A narrow distribution curve indicates rapid germination rate and uniformity, whereas a broader curve demonstrates low germination homogeneity. The mean germination time (MGT) for the specific priming treatments is the time (days after sowing) when the curve peaks. The seed of hydropriming, nutripriming, and osmopriming curves demonstrate that primed treatments with a length of 4 hrs had the maximum germination rate and consistency. Compared to unprimed seeds, all pre-germination treatments exhibit significantly earlier mean germination time (MGT), time to 50% germination (T50), and higher germination index (GI) (Table 1). It is well known that the higher germination index denotes a higher percentage and rate of germination. Generally, most priming treated seeds germinated starting from day 4 with a mean germination time (MGT) of 5-6 days and time to reach 50% germination of 4-5 days compared to unprimed seeds that took 8 days for MGT and time to reach 50% germination.

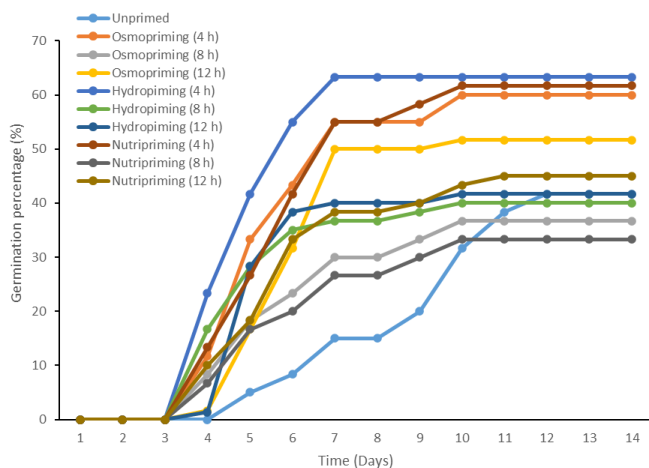


Figure 1. Germination percentage of Moringa seed under different priming applications and duration over 14 days

Table 1. Mean germination time (MGT), time to reach 50% germination (T50) of *M. oleifera* seed with different priming applications, and duration under laboratory condition

Treatments	MGT (days)	T50 (days)	Germination index (GI)
Unprimed	8.96 ^a	8.79 ^a	0.76 ^c
Osmopriming (4 hrs)	6.02 ^b	5.40 ^b	1.66 ^a
Osmopriming (8 hrs)	5.15 ^b	4.40 ^b	1.93 ^a
Osmopriming (12 hrs)	5.76 ^b	4.99 ^b	1.66 ^a
Hydropriming (4 hrs)	6.09 ^b	5.12 ^b	1.21 ^{abc}
Hydropriming (8 hrs)	5.26 ^b	4.60 ^b	1.26 ^{abc}
Hydropriming (12 hrs)	6.24 ^b	5.65 ^b	1.29 ^{ab}
Nutripriming (4 hrs)	6.25 ^b	5.58 ^b	0.87 ^{bc}
Nutripriming (8 hrs)	5.25 ^b	4.38 ^b	1.23 ^{abc}
Nutripriming (12 hrs)	5.95 ^b	5.40 ^b	1.02 ^{bc}
SE±	0.33	0.38	0.10
P Value	<0.001	<0.001	<0.001

Values with different superscripts are significantly different based on Duncan's multiple range test at 5%.

Enhanced germination because of improved water imbibition and faster metabolic process initiation, which determine radical protrusion through the softened seed coat and speed up the process of seed germination (Bray *et al.*, 1989; Basra *et al.*, 2005; Hafsat and Singh, 2009). During priming, metabolic activity transforms stored reserves into metabolites required for seed germination (Gallardo *et al.* 2001). Increased RNA and protein synthesis (Fu *et al.*, 1988; Ibrahim *et al.*, 1983) and uniformity have been linked to metabolic repair during imbibition (Bray *et al.*, 1989). Khaliq *et al.* (2015) reported that the pre-sowing treatment might be associated with a better performance of seeds through hydration trigger mechanisms to repair membranes, which could act over deteriorated seeds. Hydropriming of 4 hrs may be sufficient to partially hydrate the seeds to the point where the metabolic process of germination begins, hence enhancing the germination rate. We found that the germination percentage decreased with a longer time of priming. Lee and Kim (1999; 2000) and Farooq *et al.* (2006) support these findings that longer priming durations may negatively affect germination percentage.

3.2 Root morphology

Nutrient and water absorption depends on root morphology, indicative of plant response to the growing condition. Growth pouch experiments were conducted to examine the effects of priming on the root morphology of *M. oleifera* in a controlled environment (Table 2). Root scanning with the WinRHIZO optical scanner (Regent Instruments, Inc.) is one of the effective methods for analysing images and studying root morphological features. Priming treatments showed a significant effect on root morphology via root scan analysis. Parameters such as the number of root tips, forks, and crossings play a significant role in root architecture since they can improve penetration through

Table 2. Effects of different priming treatments and duration on root characteristics of *M. oleifera* after 14 days in growth pouches

Treatments	Length (cm)	Root Vol (cm ³)	Tips No	Fork No	Crossing No
Unprimed (Control)	163.64 ^{bcd}	0.292 ^b	260.60 ^{bc}	271.47 ^{cd}	28.53 ^c
Osmopriming (4 hrs)	137.84 ^{de}	0.278 ^b	240.20 ^c	311.07 ^{cd}	26.33 ^c
Osmopriming (8 hrs)	126.39 ^e	0.273 ^b	233.73 ^c	219.27 ^d	25.67 ^c
Osmopriming (12 hrs)	147.29 ^{cd}	0.314 ^b	228.53 ^c	275.60 ^{cd}	30.27 ^{bc}
Hydropriming (4 hrs)	240.52 ^a	0.467 ^a	344.20 ^{ab}	517.80 ^a	59.20 ^a
Hydropriming (8 hrs)	192.05 ^b	0.453 ^a	373.87 ^a	441.07 ^{ab}	46.07 ^{ab}
Hydropriming (12 hrs)	170.88 ^{bcd}	0.329 ^b	270.20 ^{bc}	370.00 ^{bc}	36.40 ^{bc}
Nutripriming (4 hrs)	183.80 ^{bc}	0.328 ^b	260.73 ^{bc}	322.00 ^{bcd}	34.60 ^{bc}
Nutripriming (8 hrs)	173.65 ^{bcd}	0.342 ^b	277.87 ^{bc}	310.13 ^{cd}	36.53 ^{bc}
Nutripriming (12 hrs)	195.69 ^b	0.301 ^b	268.47 ^{bc}	303.53 ^{cd}	33.33 ^{bc}
SE±	13.16	0.03	27.95	41.38	5.32
P Value	<0.001	<0.001	<0.001	<0.001	<0.001

Values with different superscripts are significantly different based on Duncan's multiple range test at 5%.

soil layers, positively affecting nutrient uptake. In the current study, hydropriming treatments had higher densities of root tips, forks, and crossings than unprimed treatments. Hydroprimed of 4 hrs seeds had shown the best results in enhancing root growth. The length of the roots (240.52 cm), root volume (0.47 cm³), number of forks (517.80), and number of crossing (59.20) of the hydropriming of 4 hrs seedlings were 47%, 60%, 91%, and 107% significantly more than unprimed seedlings, respectively. Results showed that osmopriming and nutripriming treated seedlings did not significantly differ from unprimed seedlings on the root morphology of *M. oleifera*. Wijewardana *et al.* (2015) reported that deeper roots and the high number of root forks and crossings lead to an improved root growth system of corn during the uptake of nutrients and water. Root branching order is the primary determinant of root trait variation among species (Picon-Cochard *et al.*, 2012). It is an essential indicator of robust seedling establishment, as fewer and shorter roots cannot cope with even minor stress, resulting in crop yield declines. It is important to understand the root system architecture and root characteristics that could compete better at the transplanting stage and aid in tolerating abiotic stress.

3.3 Seed germination and seedling growth under greenhouse conditions

The effects of the different priming treatments and growing media were observed on seedling emergence percentage, mean germination time (MGT), and germination index (GI), as shown in Table 3. A seedling was regarded to have emerged when it appeared visibly from the media surface. Results indicated that the media growing and priming treatments significantly affected seedling emergence percentage, mean germination time, and germination index. All pre-germination treatments have significantly shown early mean germination days

(MGT) at day 6 compared to the unprimed seeds except for osmopriming of 4 hrs, which was similar to unprimed treatments with delayed one day. The highest emergence percentage was recorded on seed subjected to 8 hrs hydropriming (61%), followed by hydropriming of 12 hrs (58%), osmopriming of 4 hrs (55%), and hydropriming of 4 hrs (53%). Higher germination index (1.06-1.16) and the least time to 50% germination (5.5 days) were recorded in all hydropriming treatments.

Meanwhile, unprimed seeds have the lowest emergence percentage (40.5%), germination index (0.667), and longer mean germination time (7.45 days). The seed absorbed the required amount of water during priming treatments for hydration and metabolic activity. Primed seeds complete their germination processes early compared to non-primed seeds (Dezfuli *et al.*, 2008). Due to moisture availability, primed seeds emerge rapidly after sowing and seek additional soil nutrient resources, resulting in robust seedlings with uniform crop stands. Seed priming promotes synchronised and uniform germination by reducing the lag time to imbibition, activating enzymes, accumulating germination-enhancing metabolites, and improving osmotic adjustment (Lee and Kim, 2000; Farooq *et al.*, 2006; Brocklehurst and Dearman, 2008; Hussain *et al.*, 2015; Ullah *et al.*, 2019). On the other hand, osmopriming and nutripriming affect seed hydration and other germination-related processes due to the absorption of exogenous ions/hormones, consequently confusing the effects of imbibition versus that of ions/hormones. Under stressful conditions, oxidative damage to mRNA inhibits protein synthesis and degradation, resulting in a disruption of protein functions due to changes in enzymatic and binding properties. As a result, seed germination may be delayed or suppressed.

Growing media of sand: peat moss (9:1) positively

Table 3. Emergence percentage, mean germination time (MGT), germination index (GI), and time to reach 50% germination (T50) of *M. oleifera* seedlings with different priming applications and growing media

Treatments	Emergence Percentage %	MGT (days)	GI	T50 (days)
Growing Media (M)				
Sand:peat moss (9:1)	53.87 ^a	6.80 ^a	1.040 ^a	6.11 ^a
Top soil:coconut coir:peat moss (5:2:1)	47.03 ^b	6.45 ^b	0.856 ^b	5.79 ^b
SE±	1.09	0.08	0.02	0.06
P value	<.001	<.001	<.001	<.001
Priming (P)				
Unprimed (Control)	40.50 ^d	7.45 ^a	0.667 ^c	6.83 ^a
Osmopriming (4 hrs)	55.33 ^{abc}	7.09 ^{ab}	0.980 ^{ab}	6.42 ^{ab}
Osmopriming (8 hrs)	47.50 ^{bcd}	6.74 ^{abc}	0.877 ^{bc}	5.98 ^{bc}
Osmopriming (12 hrs)	48.67 ^{bcd}	6.77 ^{ab}	0.887 ^{bc}	6.15 ^{bc}
Hydropriming (4 hrs)	52.83 ^{abc}	6.08 ^c	1.060 ^{ab}	5.50 ^d
Hydropriming (8 hrs)	61.17 ^a	6.53 ^{bc}	1.165 ^a	5.66 ^{cd}
Hydropriming (12 hrs)	58.00 ^{ab}	6.12 ^c	1.165 ^a	5.50 ^d
Nutripriming (4 hrs)	47.33 ^{bcd}	6.58 ^{bc}	0.885 ^{bc}	5.90 ^{bc}
Nutripriming (8 hrs)	47.33 ^{bcd}	6.38 ^{bc}	0.920 ^b	5.76 ^{cd}
Nutripriming (12 hrs)	45.83 ^{cd}	6.52 ^{bc}	0.877 ^{bc}	5.80 ^{bc}
SE±	2.43	0.18	0.05	0.14
P value	<0.001	<0.001	<0.001	<0.001
Interaction (M×P)	0.58	0.04	0.60	0.02

Values with different superscripts are significantly different based on Duncan's multiple range test at 5%.

affect emergence percentage, mean germination time, germination index, and time to 50% germination of *M. oleifera* seedlings. This growing media consists of mostly sand with larger particles which increases the aeration and drainage of the potting medium, allowing *M. oleifera* seeds to germinate faster. The growth media becomes more permeable by incorporating sand into the medium (Bhardwaj, 2014). Pardos (2004) reported that the excess humidity in the soil might provoke germination losses and diminish the root growth of some tree species. Growing media substrate needs to consider variations in physical, chemical, and biological properties. Additionally, the ideal substrate should cost low and be available in large quantities. Choosing an unsuitable substrate can affect the germination of seeds and the establishment of seedlings, leading to a reduction in plant quality. Our findings are in line with Abdulhamid and Dau (2016), which found that germination and early growth rate of *M. oleifera* seeds in topsoil river sand show better growth potential on average, than seeds planted in a mix of topsoil, river sand, and cow dung manure and mix of topsoil, river sand, and poultry manure. The use of coarse components as growth media provided excellent aeration and drainage of the growing media and facilitated germination and seedling emergence (Baiyeri and Aba, 2007). Germination results obtained in greenhouse environments differed from those obtained in laboratory conditions, where *M. oleifera* demonstrated better germination. It could be attributed to differences in media, culture, and environmental factors.

The different priming treatments and durations significantly affected the seedling growth of *M. oleifera* grown in a pot with two different growing media (Table 4). A faster rate of germination allowed the emerging seedlings to accumulate more biomass. Almost all treated seeds resulted in taller seedlings above 20 cm when compared to unprimed seeds with hydropriming of 4 hrs treated seeds had the tallest seedling (25.17 cm), 46% greater than unprimed seedlings at (17.17 cm). Whereas nutripriming for 4 hrs produced a robust stem girth (2.34 mm) 12% greater than unprimed seedlings. A higher number of leaves were found in hydropriming treated seeds; 12 hrs (5.5), 4 hrs (5.2), and 8 hrs (5.1). A similar trend was found in unprimed seeds; it showed the least plant height, stem girth, and the number of leaves among the treatments. Seedlings treated with nutripriming of 8 hrs had the highest total leaf area at 197.46 cm², 92% greater than unprimed seed; however, they statistically showed no differences ($P>0.05$) compared to 4 hrs hydropriming seedlings. In general, seedlings treated with hydropriming of 4 hrs gave higher values in most parameters statistically at par with seeds treated with nutripriming of 8 hrs. Similarly, Basra et al. (2003) and Nouman, Siddiqui, Basra, Afzal et al. (2012) discovered that hydropriming promotes *M. oleifera* germination and stand establishment, decreases the time between the planting and emergence and, induced resistance to unfavourable environments like abiotic stress, particularly during early growth. Hydropriming is a straightforward form of priming treatment. It does not need any technical equipment since water is used as a

Table 4. Vegetative growth measurement of *M. oleifera* seedlings treated with different growing media and priming treatments at 14 days after sowing

Treatments	Plant height (cm)	Stem girth (mm)	Number of leaves	Total leaf area (cm ²)
Growing Media (M)				
Sand:peat moss (9:1)	23.09	2.27 ^a	4.89	146.09
Top soil:coconut coir:peat moss (5:2:1)	21.76	2.20 ^b	4.81	131.92
SE±	0.53	0.02	0.10	4.10
P value	0.07	<0.001	0.55	0.09
Priming (P)				
Unprimed (Control)	17.17 ^c	2.09 ^d	4.00 ^d	102.54 ^d
Osmopriming (4 hrs)	23.83 ^a	2.31 ^{ab}	4.94 ^{abc}	106.91 ^{cd}
Osmopriming (8 hrs)	18.94 ^{bc}	2.29 ^{ab}	4.61 ^{bc}	112.93 ^{cd}
Osmopriming (12 hrs)	21.67 ^{ab}	2.27 ^{abc}	4.33 ^{cd}	107.76 ^{cd}
Hydropriming (4 hrs)	25.17 ^a	2.29 ^{ab}	5.17 ^{ab}	168.82 ^{ab}
Hydropriming (8 hrs)	23.67 ^a	2.19 ^{bcd}	5.11 ^{ab}	158.45 ^b
Hydropriming (12 hrs)	23.06 ^a	2.18 ^{cd}	5.50 ^a	141.94 ^{bcd}
Nutripriming (4 hrs)	24.56 ^a	2.24 ^{abc}	4.78 ^{bc}	146.06 ^{bc}
Nutripriming (8 hrs)	22.44 ^a	2.34 ^a	5.06 ^{ab}	197.46 ^a
Nutripriming (12 hrs)	23.72 ^a	2.13 ^d	5.00 ^{ab}	147.21 ^{bc}
SE±	1.19	0.04	0.22	5.80
P value	<0.001	<0.001	<0.001	<0.001
Interaction (M×P)	0.74	0.04	0.01	0.05

Values with different superscripts are significantly different based on Duncan's multiple range test at 5%.

priming medium. Our findings indicate that pre-soaking with water affects the germination process and the post-germination behaviour, which likely affects field establishment.

4. Conclusion

This study indicated that *M. oleifera* seeds are not an exception to the possibility of increasing emergence rate and vigour by using a seed priming technique. Seed germination and early growth phases of seedlings are vital for growing a successful crop because they significantly decide the crop stand density and affect the crop yield. Successful crop yield and productivity depend on practical methods that speed up germination and obtain more consistent germination of seeds sown. The study revealed that seed priming techniques using different solutions could significantly improve *M. oleifera* seedling's performance. Most of the priming strategies enhanced the emergence rate, synchronised the emergence, and improved the seedling growth of *M. oleifera*. However, hydropriming (4 hrs) was more effective in improving germination percentage, germination rate, and seedling growth. This priming technique is cost-effective, environmentally friendly, and easily adaptable for farmers in humid tropical Malaysia to grow *M. oleifera* from seed, which may contribute to their commercialisation as significant medicinal and nutritional crops; however, further study under field conditions could be suggested to determine the consistency of the positive effects of hydropriming on *M.*

oleifera plant growth.

Conflict of interest

The authors declare no conflict of interest.

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