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Germination and Seedling Growth of Moringa (*Moringa Oleifera*) as Influenced by Seed Treatments and Sources in Southeastern Nigeria

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ABSTRACT

Seed source and treatment could play a significant role in the overall crop performance. This study was therefore conducted in the Greenhouse,AkanuIbiam Federal Polytechnic, Unwana, southeast Nigeria, to evaluate the effect of seed treatment methods and seed sources on germination and seedling growth of moringa. Three seed treatment methods used were hydro-primed, cracked and untreated seed and seed source locations were: Abia State (A1),Anambra State (A2), Ebonyi State (A3), Enugu State (A4), and Imo State (A5). The experimental design was a 3 x 5 factorial arranged in a split-plot in complete randomized design (CRD) with three replications.Forty five 10-liter capacity buckets, each filled with growth media (top soil, river sand and poultry droppings at 2:2:1, respectively) were used for the experiment. Three seeds of Moringa were sown in each bucket and data on germination were collected at 3, 4 and 5 days after planting, while growth parameters, including plant height, stem girth, leaf number, height of first branch and canopy spread were collected at 3,6 and 9 months after planting. Data analysis was done using analysis of variance with Genstat software. Results showed that seed treatment and sources interactively had a significant influence on the germination and seedlings performances of Moringa. Hydro-primed pre-treatment and seeds from Ebonyi consistently performed best in terms of germination and other growth parameters measured compared to other seed pretreatment method and other source locations. This study therefore recommends Hydro-primed pre-treatment and seeds from Ebonyi for Moringa to ensure both seedling growth and productivity.

Keywords: *Germination, Seedling growth, Seedling potential, Pre-treatment and Seed location.*

Introduction

Moringa (*Moringa oleifera* Lam) is one of the most useful multipurpose trees known to man with virtually every part is beneficial [1]. Moringa is believed to have originated from India but now spread and used in many tropical and subtropical countries of the world [2]. Moringa is used as an important food plant for both humans and animals. It is rich in health-promoting phyto-chemicals such as carotenoids, phenolics (chlorogenic acids), flavonoids (quercitin and kacmpherol glycosides), various vitamins and minerals [3], protein, some carbohydrates and fats.

In agriculture, moringa can be used in alley cropping, horny production as a good source of nectar, growth enhancer, organic fertilizer and bio-pesticide[4]. Moringa is often regarded as an agro-forestry plant, grown as a boundary tree and as live fence. Industrially, moringa is used as gum, water purification agent, bio-fuel, pulp and a domestic cleansing agent. Medically, all parts of moringa are traditionally used for different purposes such as treatment of several ailments including malaria, typhoid fever, parasitic diseases; boost the immune system[5],[6].Moringa leaves are good sources of sulphur, containing amino acids, methionine and cysteine which are often in short supply in human body. According to [7], moringa spp. contains high levels of nutrients and antioxidants, and nutrient content contained in moringa variesamong species. However, nutrient content vary with method of preparation, age and the time of harvest. Quick and uniform field emergencies of seeds are essential requirements to increase yield and quality of produce. Slow and delayed germination ability of some seeds results in smaller seedlings and consequently, plants. Seed priming has been a common treatment to increase the rate of uniformity of emergence in many plants, resulting in rapid and uniform germination when seeds are re-imbibed [8].

Priming, however, improves germination, plant establishment and the general physiological state of plant, and induces toleranceagainst diverse conditions such as biotic and abiotic stress during emergence and early seedling growth. Priming also increases yield and yield components of most field crops (including vegetables, fruits and tree species). Hydro-priming, matri-priming and hormonal priming synchronizing enhance seed emergence and improve plant vigour. Priming is a seed enhancement method that might improve seed performance under stress conditions, including drought or freshly harvested or aged seeds which might failed to germinate [9]. However, hydro-priming is one of the most effective priming methods of seed treatments to improve earlyemergence, short vigour and chlorophyll contents [10]. Despite the potential of moringa (*M. oleifera* Lam), the production and yield obtained by the local farmers in region is quite low.

Some of the reasons associated with low yield are poor germination and emergence (crop establishment). This problem could be address by seed priming. However, improve agronomic practices that will increase the quality and growth rate of seedlings which in turn allows for more emergence and crop establishment, is therefore paramount. Thus this work was conducted to study the effects of seed pre-treatment methods and location (source) on germination, emergence and seedling growth performance of moringa in southeastern Nigeria.

Materials and Methods

Experimental site: The experiment was carried out at the Teaching and Research farm of the Department of horticulture and Landscape Technology, AkanuIbiam Federal Polytechnic, Unwana, Afikpo, Ebonyi State, to evaluate the effects of pre-treatment methods and location on germination, emergence and seedling growth performance of moringa. Unwana is located on latitude $05^{0}.48^{1}$N and longitude $7^{0}.55^{1}$E with altitude of 400m above sea level. The study area lies within the humid tropical rain forest zone of southeastern Nigeria, which is tilted towards derived savanna [11]. The air temperature is generally high all year round and the current temperature range is 21^{0}C – 32^{0}C with total annual rainfall exceeding 3500mm [12].

Treatment and Experimental design: The experiment consisted of two factors. Factor A containing three pre-treatment methods (Hydro-primed, Cracked and Untreated seeds), while Factor B consisted of five location/sources of moringa seeds collections (Abia (A1), Anambra (A2), Ebonyi (A3), Enugu (A4) and Imo (A5)). The selection criteria were based on the provenance trial recommendations. The treatments were laid out in a split – plot design fitted in completely randomized design with eight treatments replicated three times. Black polythene bags measuring 7.5cm x 20cm were filled with well-decomposed potting mixtures (2 parts topsoil: 2 parts river sand: 1 part poultry droppings), and the stacked polythene bags were watered thoroughly prior to sowing to stabilize the soil mixture. Each treatment consisted of twenty-four polythene bags with 31cm x 50cm spacing between treatments and replications, respectively.

Sowing and Sowing Methods: Moringa collected from each location were sown with three seeds per bag at a depth of 3cm after being hydro-primed and cracked per each treatment. However, prior to sowing, seeds were soaked for 8hours (from 10.00pm to 6.00am local time), popularly called "overnight soaking". After soaking, the primed seeds were removed and air dried for 3hours (6.00am to 9.00am local time) and all the seeds for the experiment were treated with seed treatment chemical (Apron star 42WS containing 20%w/w thiamethoxam, 20%w/w metalaxyl-M and 2%w/w difenoconazole) at the rate of one sachet per 4kg of seed to avoid any seed borne infection during the experiment.

Agronomic Practice: To enhance vigorous seedling growth, seedlings were thinned down to one plant per stand per polythene bag two weeks after emergence (WAE). The nursery was kept free of weeds throughout the experiment.

Data Collection and Analysis: The parameters collected were emergence/germination percentage days after sowing (DAS), plant height, stem girth, leaf number, and height at first branching and canopy spread at three-month intervals.

The data generated were subjected to analysis of variance (ANOVA) and means separated using least significant difference (LSD) at 5% probability difference.

Results and Discussion

Effects of Pre-treatment methods and seed sources/ location on moringa germination/emergence

Percentage seed germination as influenced by seed pre-treatment methods and sources at 3, 4 and 5 days after sowing (DAS) is shown in table 1.

Table 1: Effects of Pre-treatment Methods and Seed Sources on Percentage Germination/Emergence at 3, 4 and 5 days after sowing (DAS)

3 DAS

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean	% Germination
Hydro-primed	4	4	7	3	4	4.40	73.33
Cracked	2	3	5	4	3	3.40	56.67
Untreated	3	2	6	3	4	3.60	60.00
Mean	3.00	3.00	6.00	3.00	3.67		

*$LSD_{0\cdot05}$ (Pre-treatment methods) = **0.10***
*$LSD_{0\cdot05}$ (Sources/location) = **0.50***
*$LSD_{0\cdot05}$ (Pre-treatment × Sources) = **ns***

4 DAS

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean	% Germination
Hydro-primed	4	4	10	5	6	5.80	96.66
Cracked	3	5	8	5	4	5.00	83.33
Untreated	5	5	9	4	5	5.60	93.33
Mean	4.00	4.66	9.00	4.66	5.00		

*$LSD_{0\cdot05}$ (Pre-treatment methods) = **1.20***
*$LSD_{0\cdot05}$ (Sources/location) = **2.67***
*$LSD_{0\cdot05}$ (Pre-treatment × Sources) = **0.06***

5 DAS

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean	% Germination
Hydro-primed	6	5	10	4	5	6.00	100.00
Cracked	4	5	9	5	4	5.60	93.33
Untreated	5	3	10	5	6	5.00	96.67
Mean	5.00	2.60	9.67	4.67	5.00		

*$LSD_{0\cdot05}$ (Pre-treatment methods) = **0.98***
*$LSD_{0\cdot05}$ (Sources/location) = **0.32***
*$LSD_{0\cdot05}$ (Pre-treatment × Sources) = **1.33***

Note:
% germ = Percentage germination
ns = Not significant

Results show that different pre-treatment methods and seed sources had significant (P≤0.05) effects on seed percentage germination. Relative to other seed sources, seeds obtained from A3 produced the highest mean values of 6.00%, 9.00%, and 9.67% germination at 3, 4, and 5 DAS, respectively. The effects of interaction between different pre-treatment methods and sources on germination was also significant (P≤0.05) at 4 and 5 DAS and A3 seeds in hydro-primed, producing the best interactive performance with a mean value of 10 seedlings at 4 and 5 DAS, respectively. However, the increase in percentage germination recorded in the hydro-primed method of pre-treatment could be attributed to increased metabolic activities such as stimulation of growth as a result of priming. This is in line with the work of [13], who maintained that, in tomato seeds, correcting enzymatic and substrate deficiencies at the initial stage of germination, stimulation of hydrolytic enzymes brings about the breakdown of stored food materials thereby increasing metabolic activity, providing energy, and sustaining embryo growth.

Effects of Pre-treatment methods and seed sources/ location on moringa Plant Height

Results (Table 2) indicated the effect of pre-treatment methods and sources on seedlings' plant height at 3, 6 and 9 months after planting (MAP). Result shows that there was a significant (P≤0.05) effect among the treatment used. However, A3 seed sources had the highest mean plant height values of 48.83cm, 117.47cm and 214.32cm at 3, 6 and 9MAP, respectively. Among the treatment used, hydro-primed pre-treatment methods produced the tallest plant height of 53.40cm, 121.70cm and 282.60cm at 3, 6 and 9 MAP, respectively. The effect of interaction between different pre-treatment methods and seed sources on plant height was significant with hydro-primed having the best interaction performance with a value of 53.40cm, 117.47cm and 214.32cm, respectively. The increase in seedling plant height as a result of hydro-primed method of treatment could be attributed to rapid cell division in the meristematic region of the stem as well as increased in cell elongation due to multiplication of various parts of the plant tissue that might have been due to rapid seedling emergence. This agrees with the work of [14], who reported that an increasein photosynthetic processes increased plant height.

Table 2: Effects of Pre-treatment Methods and Seed Sources on Moringa Plant Height (cm) at 3, 6 and 9 Months after Planting (MAP)

3 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	47.20	45.00	53.40	46.80	44.00	47.28
Cracked	45.10	45.01	47.77	44.35	44.00	45.25
Untreated	46.33	41.98	45.33	40.97	45.02	43.93
Mean	46.21	44.00	48.83	44.04	44.34	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***1.32***
$LSD_{0.05}$ *(Sources/location)* = ***1.40***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.02***

6 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	112.60	99.50	121.70	112.40	93.33	105.91
Cracked	109.70	99.12	120.66	110.33	102.67	105.50
Untreated	106.78	89.60	109.78	103.47	100.37	102.00
Mean	109.76	96.07	117.47	105.40	98.79	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***0.76***
$LSD_{0.05}$ *(Sources/location)* = ***2.33***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.26***

9 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	152.18	165.12	282.60	174.18	164.00	187.62
Cracked	149.60	154.31	186.77	193.00	150.72	166.88
Untreated	140.44	162.00	173.60	187.47	173.00	167.30
Mean	147.41	160.48	214.32	184.88	162.57	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***2.05***
$LSD_{0.05}$ *(Sources/location)* = ***1.78***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.60***

Effects of Pre-treatment methods and seed sources/ location on other moringa growth parameters

Result of the study (Table 3) shows the effects of pre-treatment methods and seed sources on moringa stem girth (cm) at 3, 6 and 9MAP. The result was significant (P≤0.05) among the treatment used. However, the hydro-primed method of pre-treatment produced the highest mean values of stem girth across seed sources with A3 producing the highest (2.41cm, 5.63cm and 10.45cm), while untreated method had the least (2.06cm, 4.31cm and 10.00cm) at 3, 6 and 9 MAP, respectively.

Table 3: Effects of Pre-treatment Methods and Seed Sources on Moringa Stem Girth (cm) at 3, 6 and 9 Months after Planting (MAP)

3 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	2.00	1.72	2.41	1.84	2.00	1.99
Cracked	1.95	1.52	2.15	2.00	1.93	1.91
Untreated	1.83	1.93	2.06	1.81	1.94	1.91
Mean	1.93	1.72	2.21	1.88	1.96	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***0.001***
$LSD_{0.05}$ *(Sources/location)* = ***0.03***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.10***

6 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	4.92	4.12	5.63	4.03	4.52	4.64
Cracked	4.31	3.95	4.92	4.73	4.44	4.47
Untreated	4.25	3.80	4.31	4.81	4.50	4.33
Mean	4.40	3.96	4.95	4.52	4.49	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***0.01***
$LSD_{0.05}$ *(Sources/location)* = ***1.22***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.06***

9 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	6.95	7.10	10.45	9.30	6.51	8.06
Cracked	6.90	6.70	10.18	7.45	7.90	7.83
Untreated	5.20	6.00	10.00	8.96	7.81	7.59
Mean	6.35	6.60	10.21	8.57	7.41	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***0.33***
$LSD_{0.05}$ *(Sources/location)* = ***1.08***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.60***

The result also revealed that leaf number was high with the hydro-primed pre-treatment method across all the treatments, even though there was no significant difference among the pre-treatment methods (Table 4) at 3MAP but significantly differed at 6 and 9 MAP, respectively. The results revealed that A3 seed sources produced the highest leaf number (8.33, 15.67 and 20.00) at 3, 6 and 9 MAP, respectively. The highest mean values of 7.56cm, 7.80cm and 7.69cm were observed on moringa plant height at first branching (Table 5), with hydro-primed pre-treatment methods an A3 seed sources at 3, 6 and 9 MAP, respectively. However, there was no significant difference (P≤0.05) among the treatment used at 3 and 9 MAP.

Table 4: Effects of Pre-treatment Methods and Seed Sources on Moringa Number of Leaves at 3, 6 and 9 Months after Planting (MAP)

3 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	7.66	7.20	8.33	8.00	7.33	7.70
Cracked	7.20	6.33	8.13	8.00	7.69	7.47
Untreated	7.40	7.03	8.27	7.01	7.00	7.34
Mean	7.42	6.85	8.24	7.67	7.34	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***ns***
$LSD_{0.05}$ *(Sources/location)* = ***1.12***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.03***

6 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	10.00	11.00	15.67	10.00	9.00	11.13
Cracked	9.20	10.00	12.60	11.00	10.00	10.56
Untreated	9.27	10.00	12.00	10.67	11.00	10.59
Mean	9.49	10.33	13.42	10.57	10.00	

$LSD_{0.05}$ *(Pre-treatment methods)* = ***0.11***
$LSD_{0.05}$ *(Sources/location)* = ***0.31***
$LSD_{0.05}$ *(Pre-treatment × Sources)* = ***0.06***

9 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	17.00	18.00	20.00	19.00	16.00	18.00
Cracked	19.00	15.00	18.88	18.43	19.00	18.27
Untreated	18.33	14.79	19.74	18.00	18.01	15.77
Mean	18.11	16.93	19.54	18.48	17.67	

*$LSD_{0.05}$ (Pre-treatment methods) = **0.06***
*$LSD_{0.05}$ (Sources/location) = **0.10***
*$LSD_{0.05}$ (Pre-treatment × Sources) = **0.11***

Notes:
MAP = Months After Planting
ns = Not significant

Table 5: Effects of Pre-treatment Methods and Seed Sources on Height of First Branch (cm) at 3, 6 and 9 Months after Planting (MAP)

3 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	6.38	7.07	7.95	6.76	7.01	7.03
Cracked	6.78	7.00	7.50	6.88	7.00	7.03
Untreated	6.23	6.34	7.23	6.56	7.01	6.67
Mean	6.46	6.80	7.56	6.73	7.01	

*$LSD_{0.05}$ (Pre-treatment methods) = **ns***
*$LSD_{0.05}$ (Sources/location) = **ns***
*$LSD_{0.05}$ (Pre-treatment × Sources) = **ns***

6 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	6.73	6.93	8.01	6.92	6.65	7.04
Cracked	7.37	6.95	7.52	6.73	7.00	7.11
Untreated	7.11	6.83	7.86	6.68	6.96	7.06
Mean	7.07	6.90	7.80	6.78	6.87	

*$LSD_{0.05}$ (Pre-treatment methods) = **1.23***
*$LSD_{0.05}$ (Sources/location) = **0.50***
*$LSD_{0.05}$ (Pre-treatment × Sources) = **ns***

9 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	7.00	6.73	7.91	7.26	6.70	7.12
Cracked	7.03	6.93	7.84	6.85	7.00	7.13
Untreated	6.93	6.83	7.31	6.90	6.96	6.99
Mean	6.99	6.82	7.69	6.94	6.84	

*$LSD_{0.05}$ (Pre-treatment methods) = **ns***
*$LSD_{0.05}$ (Sources/location) = **ns***
*$LSD_{0.05}$ (Pre-treatment × Sources) = **ns***

Notes:
MAP = Months After Planting
ns = Not significant

Results obtained (Table 6) shows the effects of pre-treatment methods and seed sources on canopy spread (cm) of moringa. It was observed that there was a significant effect on canopy spread at 3 and 6 MAP. The hydro-primed pre-treatment method had the highest canopy spread mean values of 16.64cm, 27.51cm and 46.75cm with A3 seed sources, while the least mean values of canopy spread was observed with A2 (12.79cm), A5 (22.45cm) and A4 (38.39cm) at 3, 6 and 9 MAP, respectively.
The increase in growth parameters observed in hydro-primed pre-treatment method could be attributed to seeds overcoming stress conditions, improved nutrient uptake as well as enhanced antioxidant activity during germination and seedling growth. This probably could have led to reduced seedling failure and better crop performance through faster germination, increased seedling vigor, enhanced root development and improved seedling establishment.

This work agrees with the work of [9], who reported hydro-priming as a more effective priming strategy to improve early emergence, shoot vigor chlorophyll and mineral contents.

Table 6: Effects of Pre-treatment Methods and Seed Sources on Moringa Canopy Spread (cm) at 3, 6 and 9 Months after Planting (MAP)

3 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	16.77	12.38	18.56	15.33	16.33	15.06
Cracked	14.89	13.25	15.80	14.78	15.01	14.75
Untreated	15.09	12.73	15.56	13.92	16.38	14.74
Mean	15.58	12.79	16.64	14.68	15.91	

*$LSD_{0.05}$ (Pre-treatment methods) = **0.01***
*$LSD_{0.05}$ (Sources/location) = **0.32***
*$LSD_{0.05}$ (Pre-treatment × Sources) = **0.02***

6 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	27.74	23.14	29.34	20.34	21.22	24.36
Cracked	23.84	20.98	28.84	22.44	23.11	23.84
Untreated	27.02	24.44	24.34	24.56	24.12	25.00
Mean	26.20	22.85	27.51	22.45	22.82	

*$LSD_{0.05}$ (Pre-treatment methods) = **1.32***
*$LSD_{0.05}$ (Sources/location) = **0.41***
*$LSD_{0.05}$ (Pre-treatment × Sources) = **0.07***

9 MAP

Pre-treatment Methods	A1	A2	A3	A4	A5	Mean
Hydro-primed	45.30	44.86	48.37	33.84	34.56	41.39
Cracked	44.21	43.72	44.55	43.50	44.00	44.00
Untreated	36.12	44.00	47.33	37.84	42.00	41.46
Mean	41.88	44.19	46.75	38.39	40.19	

*$LSD_{0.05}$ (Pre-treatment methods) = **ns***
*$LSD_{0.05}$ (Sources/location) = **ns***
*$LSD_{0.05}$ (Pre-treatment × Sources) = **1.34***

Notes:
MAP = Months After Planting
ns = Not significant

However, the results of the experiment showed that seedlings height, stem girth, leaf number, height at branching and canopy spread from seeds sourced from A3 (Ebonyi State) had the highest mean values of 126.87cm, 5.79cm, 13.73, 7.68cm and 30.30cm plant height, respectively. Seeds sourced from A2 (Anambra State) produced the least mean values of 100.18cm, and 11.37cm plant height and leaf number, respectively.
The results also indicated that the least stem girth, height at first branching and canopy spread (4.68cm, 6.81cm and 26.31cm) was recorded from seeds obtained from A5 (Imo State), A4 (Enugu State) and A5 (Imo State), respectively. The mean values of growth parameters sampled seedlings showed disparity from one to another among the five different sources. This could be attributed to differences in environmental conditions of the seeds of origin, possibly due to the variation that occurred in the natural ecosystem among individual plants even when planted in the same environment. This implies that the effect of geographical variations on the growth and development of species is paramount in seed study. [15], reported that local plant is subject to a wide variety of external influences that may modify the growth and development of the plant to a greater or a lesser degree.

Conclusion

Seed pre-treatment and sources had significant influence on the percent germination and seedlings performances of Moringa.

Hydro-primed pre-treatment and seeds from Ebonyi State consistently performed best in terms of germination/ emergence and other growth parameters measured compared to other seed pretreatment methods and other source locations. Therefore, the study recommends hydo-primed seed treatment and Ebonyi grown seeds for better Moringa production in southeast Nigerian.

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