

Germination and Seedling Vigor of Different Soybean (*Glycine max*) Varieties Subjected to Various Seed Priming Agents

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ABSTRACT

Soybean (*Glycine max* L.) is an economically important legume valued for its high protein and oil content. However, poor seed germination and weak seedling establishment remain major constraints to achieving optimum crop productivity. This study evaluated the effects of different seed priming agents on the germination and early seedling growth of three soybean varieties using a factorial experiment arranged in a Completely Randomized Design (CRD). The priming treatments consisted of moringa leaf extract (MLE), neem leaf extract (NLE), garlic extract, hydropriming, hormoprimer, and an untreated control. Germination percentage, shoot length, root length, seedling length, and seedling vigor index were evaluated. Results revealed that both soybean variety and seed priming treatment significantly affected all measured parameters. Among the varieties, Manchuria consistently exhibited the highest germination percentage and seedling vigor. Among the priming treatments, moringa leaf extract produced the best overall performance, followed closely by neem leaf extract, while untreated seeds consistently recorded the lowest values. The enhanced performance of MLE and NLE was reflected in higher germination percentage, longer shoot and root lengths, greater seedling length, and improved seedling vigor index. The findings demonstrate that botanical seed priming, particularly with moringa and neem leaf extracts, is an effective and environmentally friendly approach for improving soybean seed germination and early seedling establishment. These natural priming agents have the potential to enhance seed quality and support sustainable soybean production.

Keywords: priming agents, germination, vigor index, soybean

INTRODUCTION

Soybean (*Glycine max* L.) is one of the world's most important legume crops because of its high protein and oil content, making it valuable for food, feed, and industrial applications. Despite its economic importance, soybean production is often constrained by poor seed germination and weak seedling establishment, which can reduce plant population and ultimately affect yield. Improving seed quality and early seedling vigor is therefore essential for achieving sustainable soybean production.

Seed priming has emerged as a simple, low-cost, and environmentally friendly technique that enhances seed performance by initiating metabolic processes before germination. Primed seeds generally exhibit faster and more uniform germination, improved seedling growth, and greater tolerance to environmental stresses compared with untreated seeds (Paparella et al., 2015). Recently, interest has increased in the use of botanical seed priming agents because they are biodegradable, readily available, and contain naturally occurring growth-promoting compounds.

Among botanical extracts, *Moringa oleifera* leaf extract has gained attention due to its high concentrations of cytokinins, vitamins, minerals, amino acids, and antioxidants that stimulate germination and seedling growth (Leone et al., 2015; Gopalakrishnan et al., 2016). Likewise, neem (*Azadirachta indica*) leaf extract contains phenolic compounds and flavonoids with antioxidant and antimicrobial properties that may enhance seed germination and seedling development. However, information comparing the effectiveness of these botanical priming agents among different soybean varieties remains limited.

This study evaluated the effects of moringa leaf extract, neem leaf extract, and hydropriming on the germination and early seedling growth of three soybean varieties. Specifically, it aimed to determine the influence of these seed priming treatments on germination percentage, shoot length, root length, seedling length, and seedling vigor index to identify an effective and sustainable seed enhancement strategy for soybean production.

METHODOLOGY

Experimental Materials

The study utilized seeds of three soybean (*Glycine max* L.) varieties, namely Tiwala 8, Tiwala 10, and Manchuria. The seed priming materials included young moringa (*Moringa oleifera*) leaves, neem (*Azadirachta indica*) leaves, ProGibb® 40SG (gibberellic acid, GA₃), fresh garlic (*Allium sativum*) cloves, and distilled water. Other materials used were plastic germination boxes, filter paper, paper towels, measuring cups, blender, cloth filter, digital weighing scale, ruler, labels, and laboratory record notebook.

Experimental Design and Treatments

The experiment was arranged in a split-plot design under a Completely Randomized Design (CRD) with three replications.

The main plot factor consisted of three soybean varieties:

- A₁ – Tiwala 8
- A₂ – Tiwala 10
- A₃ – Manchuria

The subplot factor consisted of six seed priming treatments:

- B₁ – No priming (Control)
- B₂ – Hydropriming (Distilled water)
- B₃ – Hormopriming (GA₃)
- B₄ – Moringa leaf extract
- B₅ – Neem leaf extract
- B₆ – Garlic extract

The treatment combinations ($3 \times 6 = 18$) were replicated three times, resulting in 54 experimental units. Treatments within each replication were assigned randomly using a lottery method without replacement.

Preparation of Plant Extracts

Fresh, healthy, and disease-free young leaves of moringa (*Moringa oleifera* L.) and neem (*Azadirachta indica* A. Juss.) were collected from mature plants within the vicinity of the experimental area. The leaves were thoroughly washed under running tap water to remove adhering dust and debris, followed by a final rinse with distilled water to eliminate surface contaminants. The cleaned leaves were air-dried under shade at ambient room temperature (27–30°C) for approximately 12 hours to remove excess surface moisture while preventing the degradation of heat-sensitive bioactive compounds.

The partially dried leaves were ground separately into a fine powder using a sterilized electric blender. For each botanical extract, 50 g of leaf powder was mixed with 500 mL of distilled water (1:10 w/v ratio) in sterile glass

containers. The mixtures were allowed to stand for 24 hours at room temperature (27–30°C) to facilitate the extraction of water-soluble phytochemicals. The extracts were stirred intermittently every 6 hours to enhance extraction efficiency. After extraction, the mixtures were filtered through a double layer of muslin cloth followed by Whatman No. 1 filter paper to obtain a clear filtrate. The stock extracts were stored in sterile amber bottles under refrigeration at 4°C and were used within 48 hours to preserve their biological activity.

A 15% (v/v) working solution was prepared by diluting 150 mL of the stock extract with 850 mL of distilled water immediately before use.

Fresh garlic (*Allium sativum* L.) extract was prepared by crushing 10 g of peeled garlic cloves using a sterile mortar and pestle. The crushed cloves were homogenized with 100 mL of distilled water and allowed to stand for 30 minutes to maximize the release of sulfur-containing bioactive compounds. The mixture was then filtered through Whatman No. 1 filter paper, and the filtrate was used immediately as the priming solution to minimize the loss of volatile compounds.

For the hormone priming treatment, 0.2 g of ProGibb® 40SG (gibberellic acid, GA₃) was dissolved in 500 mL of distilled water following the manufacturer's recommended concentration. The solution was prepared immediately before seed priming to ensure its effectiveness.

Seed Priming Procedure

A total of 2,700 soybean seeds were used in the experiment. Prior to priming, the seeds were visually inspected, and damaged, cracked, malformed, and discolored seeds were discarded to ensure uniformity in seed quality. The remaining seeds were randomly assigned to the respective treatment combinations and placed in labeled 750-mL plastic containers containing sufficient volumes of the designated priming solutions to ensure complete seed immersion throughout the soaking period.

The seeds were soaked in their respective priming solutions for 8 hours at room temperature (27–30°C) under laboratory conditions. During soaking, the containers were loosely covered to minimize contamination while allowing adequate aeration. Upon completion of the soaking period, the seeds were rinsed three times with distilled water to remove residual botanical extracts or growth regulator solution. Excess water was removed using a sterile plastic strainer.

The primed seeds were then spread in a single layer on sterile paper towels and air-dried under shade for approximately 12 hours at room temperature (27–30°C) until they regained their original moisture content and free-flowing characteristics suitable for sowing. Direct exposure to sunlight was avoided to prevent heat stress and possible deterioration of seed viability.

Immediately after drying, the seeds were sown in accordance with the experimental design to minimize changes in seed moisture content before germination.

Environmental Conditions During Germination

Following priming, the seeds were subjected to germination tests under controlled laboratory conditions. The germination experiment was conducted at an ambient temperature of $28 \pm 2^\circ\text{C}$, with a relative humidity of approximately 70–80% and under natural photoperiod conditions. Seeds were placed on moistened germination paper (or paper towels) with sufficient distilled water to maintain adequate moisture throughout the experimental period. Moisture levels were monitored daily, and distilled water was added as needed to prevent drying of the germination medium while avoiding waterlogging. Germination observations were conducted daily until the final germination count, following the procedures prescribed by the International Seed Testing Association (ISTA, 2024).

Seed Germination Test

Fifty seeds were placed in each plastic germination box lined with moist paper towels. Seeds were arranged uniformly with adequate spacing to minimize competition during germination.

The germination medium was maintained at adequate moisture throughout the experimental period by periodic application of distilled water whenever necessary. Seedlings were evaluated seven days after sowing following standard germination testing procedures.

Data Gathering

Observations were recorded during the germination period. Germination and seedling parameters were evaluated seven days after sowing based on the prescribed experimental procedures. Data collected included germination percentage, germination rate, seedling vigor, root length, shoot length, fresh weight, dry weight, and vigor index.

Statistical Analysis

Data collected were subjected to analysis of variance (ANOVA) appropriate for a split-plot design under a Completely Randomized Design (CRD). Significant differences among treatment means were determined using Duncan's Multiple Range Test (DMRT) at the 5% level of significance. Statistical analyses were performed using STAR (Statistical Tool for Agricultural Research) software.

RESULTS AND DISCUSSIONS

Germination Percentage

Table 1 shows that germination percentage was significantly affected by both soybean variety and seed priming treatment. Among the three varieties, Manchuria consistently exhibited the highest germination percentage, while Tiwala 8 recorded the lowest. Among the priming treatments, moringa leaf extract (MLE) produced the highest germination percentage in Tiwala 8 and Manchuria, whereas neem leaf extract (NLE) was most effective in Tiwala 10. Conversely, untreated seeds consistently recorded the lowest germination percentages, indicating the beneficial effect of seed priming on soybean seed performance.

The superior performance of MLE may be attributed to its abundance of natural growth-promoting substances, including cytokinins, vitamins, minerals, amino acids, and antioxidants, which enhance enzyme activation, reserve mobilization, and membrane repair during germination. Likewise, NLE contains phenolic compounds and flavonoids with antioxidant and antimicrobial properties that may protect germinating seeds from oxidative damage and microbial infection, thereby promoting higher germination rates. These physiological responses enable primed seeds to germinate more rapidly and uniformly than untreated seeds.

The consistently higher germination observed in the Manchuria variety suggests that genetic differences among soybean cultivars influence their response to seed priming. Varieties with higher physiological seed quality are generally more responsive to priming because they possess greater metabolic efficiency and improved capacity for reserve mobilization during germination.

The present findings agree with previous studies showing that moringa leaf extract significantly enhances seed germination and early seedling establishment by improving physiological and biochemical processes associated with seed vigor (Khan et al., 2021; Khan et al., 2020). Similarly, Leone et al. (2015) and Gopalakrishnan et al. (2016) reported that the rich phytochemical composition of *Moringa oleifera* contributes to improved germination and seedling growth. These results indicate that MLE and NLE are promising, environmentally friendly seed priming agents that can improve soybean establishment and support sustainable crop production.

Table 1. Germination percentage of soybean as affected by variety and different seed priming agents.

SEED PRIMING AGENT	VARIETY
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	Tiwala 8	Tiwala 10	Manchuria
No Priming	61.33 ^c	61.00 ^d	70.33 ^d
Hydropriming	62.33 ^c	65.33 ^{cd}	74.00 ^d
Hormopriming	66.67 ^{bc}	72.00 ^{ab}	80.67 ^c
Priming in MLE	76.67 ^a	72.67 ^{ab}	92.67 ^a
Priming in NLE	71.33 ^b	74.67 ^a	88.33 ^{ab}
Priming in GE	65.33 ^c	67.33 ^{bc}	85.33 ^{ab}

Means with similar letter superscripts are not significantly different.

Shoot Length

Table 2 shows that shoot length was significantly influenced by both soybean variety and seed priming treatment. Among the three varieties, Manchuria produced the longest shoots, while Tiwala 8 had the shortest. Among the priming treatments, moringa leaf extract (MLE) resulted in the greatest shoot length (12.44 cm), followed by neem leaf extract (NLE) (11.87 cm), whereas untreated seeds produced the shortest shoots (8.93 cm). These findings indicate that botanical seed priming effectively enhanced the early vegetative growth of soybean seedlings.

The superior shoot growth observed in MLE-treated seedlings can be attributed to the presence of natural cytokinins, particularly zeatin, as well as essential nutrients, vitamins, and antioxidants that stimulate cell division, cell elongation, and chlorophyll synthesis during early seedling development. Likewise, the beneficial effect of NLE may be associated with its bioactive compounds, including flavonoids and phenolic compounds, which enhance metabolic activity and protect developing tissues from oxidative stress. These physiological responses promote faster seedling establishment and improved shoot development compared with untreated seeds.

The superior performance of the Manchuria variety suggests that genetic variation influences seedling growth and responsiveness to seed priming treatments. Varieties with higher physiological vigor are generally more efficient in utilizing stored food reserves during germination, resulting in greater shoot elongation and more vigorous early growth.

The present findings are consistent with previous studies reporting that moringa leaf extract significantly improves shoot growth by enhancing photosynthetic activity, nutrient uptake, and hormonal regulation (Khan et al., 2021; Khan et al., 2020). Similarly, Gopalakrishnan et al. (2016) and Leone et al. (2015) highlighted the growth-promoting properties of *Moringa oleifera*, while Yaseen and Hájos (2020) demonstrated that moringa leaf extract positively influenced vegetative growth in horticultural crops. These results suggest that MLE and NLE can serve as effective, sustainable seed priming agents for improving early soybean growth and crop establishment.

Table 2. Shoot length (cm) of soybean as affected by variety and different seed priming agents.

SEED PRIMING AGENT	VARIETY			SPA MEAN
	Tiwala 8	Tiwala 10	Manchuria	
No Priming	8.77	8.43	9.60	8.93 ^d

Hydropriming	9.60	10.67	10.80	10.36 ^c
Hormopriming	9.53	10.67	11.00	10.40 ^c
Priming in MLE	12.00	12.50	12.83	12.44 ^a
Priming NLE	11.43	11.83	12.33	11.87 ^a
Priming in GE	10.50	10.97	11.83	11.10 ^b
Variety Mean	10.31 ^c	10.84 ^b	11.40 ^a	
Means with similar letter superscripts are not significantly different.				

Root Length

Table 3 shows that root length was significantly influenced by both soybean variety and seed priming treatment. Among the three soybean varieties, Manchuria produced the longest roots (5.61 cm), while Tiwala 8 recorded the shortest root length. Among the priming treatments, moringa leaf extract (MLE) resulted in the longest roots (6.59 cm), followed by neem leaf extract (NLE) (6.33 cm), whereas untreated seeds produced the shortest roots (4.06 cm). These findings indicate that botanical seed priming effectively promotes early root development in soybean seedlings.

The enhanced root growth observed in MLE-treated seedlings may be attributed to its rich content of natural phytohormones, essential nutrients, amino acids, and antioxidants, which stimulate cell division and elongation while improving nutrient mobilization during germination. Similarly, the antioxidant and antimicrobial compounds present in neem leaf extract may enhance root development by protecting germinating seeds from oxidative stress and creating favorable conditions for early seedling growth. These physiological effects promote greater water and nutrient uptake, resulting in longer and healthier root systems.

The superior root growth of the Manchuria variety further indicates that genetic factors influence the response of soybean seeds to priming treatments. Varieties with higher seed vigor generally exhibit more efficient reserve mobilization and root development during germination, allowing seedlings to establish more rapidly under favorable conditions.

The results are consistent with previous studies demonstrating that moringa leaf extract significantly enhances root growth and early seedling development through improved physiological and biochemical processes (Khan et al., 2021; Khan et al., 2020). Likewise, Leone et al. (2015) and Gopalakrishnan et al. (2016) emphasized the biostimulant properties of *Moringa oleifera*, while Hasanuzzaman et al. (2020) reported that improved antioxidant defense during germination contributes to better root establishment. These findings suggest that MLE and NLE are effective natural seed priming agents for promoting vigorous root growth and improving soybean seedling establishment.

Table 3. Root length (cm) of soybean as affected by variety and different seed priming agents.

SEED PRIMING AGENT	VARIETY			SPA MEAN
	Tiwala 8	Tiwala 10	Manchuria	
No Priming	3.87	4.00	4.30	4.06 ^e
Hydropriming	4.13	4.37	4.63	4.38 ^d
Hormopriming	4.80	5.10	5.50	5.13 ^c

Priming in MLE	6.43	6.57	6.77	6.59 ^a
Priming NLE	6.10	6.30	6.60	6.33 ^a
Priming in GE	5.50	5.90	5.87	5.76 ^b
Variety Mean	5.14 ^b	5.37 ^{ab}	5.61 ^a	
Means with similar letter superscripts are not significantly different.				

Seedling Length

Table 4 shows that seedling length was significantly affected by both soybean variety and seed priming treatment. Among the soybean varieties, Manchuria produced the longest seedlings, while Tiwala 8 had the shortest. Among the priming treatments, moringa leaf extract (MLE) resulted in the greatest seedling length, followed by neem leaf extract (NLE), whereas untreated seeds consistently produced the shortest seedlings. These findings indicate that botanical seed priming effectively enhances the overall early growth of soybean seedlings.

The superior seedling length observed in MLE-treated seeds may be attributed to the presence of natural cytokinins, essential nutrients, amino acids, and antioxidants that stimulate cell division, cell elongation, and efficient mobilization of seed reserves during germination. Likewise, the bioactive compounds present in neem leaf extract may enhance metabolic activity and reduce oxidative stress, thereby supporting both shoot and root development. As seedling length reflects the combined growth of the shoot and root, improvements in both organs contributed to the greater overall seedling growth observed in primed seeds.

The consistently superior performance of the Manchuria variety suggests that varietal characteristics play an important role in determining seedling growth potential and responsiveness to seed priming treatments. Seeds with higher physiological vigor are generally more efficient in utilizing stored reserves, resulting in faster and more uniform seedling development.

These findings agree with previous studies reporting that moringa leaf extract significantly improves seedling growth through enhanced hormonal regulation, nutrient uptake, and antioxidant activity (Khan et al., 2021; Khan et al., 2020). Similarly, Leone et al. (2015) and Gopalakrishnan et al. (2016) recognized *Moringa oleifera* as an effective natural biostimulant capable of promoting early seedling growth. The results suggest that MLE and NLE are promising, sustainable seed priming agents for improving seedling establishment and early vigor in soybean cultivation.

Table 4. Seedling length (cm) of soybean as affected by variety and different seed priming agents.

SEED PRIMING AGENT	VARIETY			SPA MEAN
	Tiwala 8	Tiwala 10	Manchuria	
No Priming	12.33	12.43	13.90	12.99 ^d
Hydropriming	13.73	15.03	15.43	14.73 ^c
Hormopriming	14.33	15.77	16.50	15.53 ^c
Priming in MLE	15.77	19.07	19.60	18.14 ^a
Priming NLE	17.53	18.13	18.93	18.20 ^a

Priming in GE	16.00	16.87	17.70	16.86 ^b
Variety Mean	15.00 ^b	16.22 ^a	17.01 ^a	
Means with similar letter superscripts are not significantly different.				

Seedling Vigor Index

Table 5 shows that the Seedling Vigor Index (SVI) was significantly influenced by both soybean variety and seed priming treatment. Among the three soybean varieties, Manchuria consistently recorded the highest SVI, while Tiwala 8 had the lowest. Among the priming treatments, moringa leaf extract (MLE) produced the highest seedling vigor index, followed by neem leaf extract (NLE), whereas untreated seeds exhibited the lowest SVI. These results indicate that botanical seed priming substantially enhanced the overall vigor and establishment potential of soybean seedlings.

The higher seedling vigor observed in MLE-treated seeds can be attributed to its rich composition of natural cytokinins, essential nutrients, amino acids, vitamins, and antioxidant compounds that promote rapid germination, efficient reserve mobilization, and vigorous shoot and root growth. Similarly, the antioxidant and antimicrobial properties of neem leaf extract may improve seed metabolic activity while reducing oxidative stress during germination, resulting in healthier and more vigorous seedlings. Since the seedling vigor index integrates both germination percentage and seedling length, the superior performance of MLE and NLE reflects their combined positive effects on seed germination and early seedling development.

The consistently higher SVI recorded by the Manchuria variety suggests that varietal characteristics significantly influence seed vigor and responsiveness to seed priming. Seeds with greater physiological quality are generally more capable of utilizing stored reserves efficiently, resulting in stronger seedling establishment and improved early growth.

These findings are consistent with previous reports that moringa leaf extract enhances seed vigor by improving germination, seedling growth, and antioxidant activity (Khan et al., 2021; Khan et al., 2020). Likewise, Leone et al. (2015) and Gopalakrishnan et al. (2016) highlighted the biostimulant properties of *Moringa oleifera*, while Paparella et al. (2015) emphasized that seed priming improves seed vigor through the activation of physiological repair mechanisms before germination. Overall, the results demonstrate that MLE and NLE are effective, sustainable seed priming agents capable of improving seedling vigor and promoting successful soybean establishment under field conditions.

Table 5. Seedling vigor index of soybean as affected by variety and different seed priming agents.

SEED PRIMING AGENT	VARIETY		
	Tiwala 8	Tiwala 10	Manchuria
No Priming	7.75 ^e	7.59 ^d	9.78 ^f
Hydropriming	8.56 ^{de}	9.82 ^c	11.43 ^e
Hormopriming	9.54 ^{cd}	11.36 ^b	13.31 ^d
Priming in MLE	14.13 ^a	13.84 ^a	18.19 ^a
Priming in NLE	12.51 ^b	13.53 ^a	16.72 ^b
Priming in GE	10.45 ^c	11.36 ^b	15.10 ^c

Means with similar letter superscripts are not significantly different.

CONCLUSIONS

This study demonstrated that both soybean variety and seed priming treatment significantly influenced germination and early seedling growth. Among the soybean varieties evaluated, Manchuria consistently exhibited superior germination percentage and seedling vigor. Likewise, moringa leaf extract (MLE) and neem leaf extract (NLE) significantly improved germination percentage, shoot length, root length, seedling length, and seedling vigor index compared with hydropriming and untreated seeds, with MLE showing the best overall performance. These findings indicate that botanical seed priming, particularly using moringa leaf extract, is an effective, low-cost, and environmentally friendly strategy for enhancing soybean seed quality and early seedling establishment. Future studies should validate these results under field conditions and evaluate the effects of botanical seed priming on plant growth, yield, and stress tolerance across different soybean varieties.

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