Vol. 15, No. 3 (2026): 1014 - 1022 http://dx.doi.org/10.23960/jtepl.v15i3.1014-1022

Agricultural Engineering
UNIVERSITY OF LAMPUNG

JURNAL TEKNIK PERTANIAN LAMPUNG

ISSN 2302-559X (print) / 2549-0818 (online)
Journal homepage : https://jurnal.fp.unila.ac.id/index.php/JTP

Application of Mycorrhizal Inoculum and Moringa Leaf Extract to Improve Growth and Yield of Soybean (*Glycine max* L.) on Alluvial Soil

Agnes Tutik Purwani Irianti[1,✉], Agus Suyanto[1], Sherly Oktarianti[1], Edi Kurniadi[1]

[1] Faculty of Agriculture, Science and Technology, Universitas Panca Bhakti, Pontianak, West Kalimantan, INDONESIA.

Article History:

Received : 01 November 2025
Revised : 17 April 2026
Accepted : 08 May 2026

Keywords:

Arbuscular mycorrhizal fungi,
Biofertilizer,
Glycine max,
Moringa oleifera,
Sustainable agriculture,
Tropical soils.

Corresponding Author:
✉ agnestutik@upb.ac.id
(Agnes Tutik Purwani Irianti)

ABSTRACT

Soybean (Glycine max L.) production in Indonesia faces persistent challenges, with productivity declining from 16.70 quintals/ha in 2021 to 14.56 quintals/ha in 2023. Alluvial soils in West Kalimantan, encompassing approximately 1,495,033 ha, present opportunities for soybean expansion but are constrained by low pH, poor nutrient availability, and suboptimal biological activity. This study investigated the combined effects of arbuscular mycorrhizal fungi (AMF) inoculation and moringa (Moringa oleifera) leaf extract application on soybean growth and productivity on alluvial soil. A factorial completely randomized design was employed with AMF doses (0, 5, and 10 g/polybag) and moringa leaf extract volumes (150, 200, and 250 ml polybag^{-1} *at 20% v/v), replicated three times. Two-way factorial ANOVA showed significant main effects and significant interaction effects (M×E) between AMF and moringa extract for all measured variables (*$P < 0.05$ *or* $P < 0.01$*). The combination of AMF at 10 g/polybag with moringa extract at 250 ml/polybag (*M_2E_3*) produced optimal results with 97.2 pods/plant and 48.6 g seed dry weight (DW) per plant, representing increases of 129% and 191% over the non-mycorrhizal lowest-dose control (*M_0E_1*), respectively. These findings suggest that integrated application of AMF and moringa extract represents a promising strategy for improving soybean productivity on marginal alluvial soils, pending field-scale validation.*

1. INTRODUCTION

Soybean (*Glycine max* L. Merrill) constitutes a critical protein and oil source for both human consumption and livestock feed across tropical agricultural systems (Aloo *et al*., 2019). Indonesia's soybean sector faces substantial productivity challenges, with national yields declining from 16.70 quintals/ha in 2021 to 14.56 quintals/ha in 2023, and national production declining from 2.4 million tons in 2021 to 2.3 million tons in 2022 (Kementerian Pertanian, 2023), necessitating continued import dependence to meet domestic demand. This productivity gap underscores the urgent need for innovative agronomic strategies that can enhance production efficiency on available agricultural lands (Adie & Krisnawati, 2017).

West Kalimantan possesses considerable potential for soybean expansion, with alluvial soils encompassing approximately 1,495,033 hectares or 10.29% of the provincial land area (BPS Kalbar, 2020). However, these soils present multiple agronomic constraints that limit crop productivity. Characteristic features include fine-textured profiles dominated by silt fractions, acidic pH conditions, and inadequate availability of essential macronutrients, particularly nitrogen, phosphorus, and potassium (Fageria & Moreira, 2015; Havlin *et al*., 2014). Furthermore, poor soil aeration impedes beneficial microbial activity, reducing populations of nitrogen-fixing bacteria and other plant growth-promoting microorganisms (Uroz *et al*., 2019).

Arbuscular mycorrhizal fungi (AMF) establish mutualistic symbioses with approximately 80% of terrestrial plant species, including most economically important crops (Martin & van der Heijden, 2024). These obligate biotrophs form specialized structures within root cortical cells that facilitate bidirectional nutrient exchange between fungal and plant partners (Shi *et al*., 2023). The extraradical hyphal network produced by AMF substantially extends the nutrient absorption zone beyond the rhizosphere depletion area, particularly enhancing phosphorus acquisition from sparingly soluble soil pools (Martin & van der Heijden, 2024). Beyond nutritional benefits, AMF colonization improves soil structure through glomalin production, enhances drought tolerance, and provides protection against various soil-borne pathogens (Nie *et al*., 2024; Rillig & Mummey, 2006).

Moringa oleifera Lam. has emerged as a valuable botanical resource for sustainable agriculture due to its exceptional nutritional profile and bioactive compound composition (Mashamaite *et al*., 2022). Moringa leaves contain substantial concentrations of essential nutrients and growth-promoting phytohormones such as zeatin, indole-3-acetic acid, and gibberellic acid (Zulfiqar *et al*., 2020). Aqueous extracts from moringa foliage have demonstrated capacity to enhance vegetative growth, accelerate flowering, improve stress tolerance, and increase crop yields across multiple species (Arif *et al*., 2023; Irshad *et al*., 2024).

The theoretical foundation for combining AMF inoculation with moringa leaf extract application rests on complementary mechanisms of action. While AMF primarily enhances nutrient acquisition efficiency through expanded hyphal networks, moringa extracts provide readily available nutrients and growth regulators that support rapid early establishment and sustained vegetative development (Irshad *et al*., 2024). Selection of AMF dose levels (5 and 10 g/polybag) and moringa extract volumes (150–250 mL/polybag) was guided by dose-response evidence reported by Smith & Read (2008), Ortas (2012), Emongor (2015), and Irshad *et al*. (2024). However, empirical evidence regarding the combined interactive effects of these two interventions remains limited, particularly for leguminous crops on alluvial soils in tropical environments. Therefore, this investigation aimed to: (i) quantify the individual and interactive effects of AMF inoculation and moringa leaf extract on soybean growth parameters; (ii) evaluate the impact of these treatments on nodulation efficiency; and (iii) assess yield responses to determine optimal application rates for maximizing soybean productivity on alluvial soils.

2. MATERIALS AND METHODS

2.1. Experimental Site and Duration

The investigation was conducted under controlled greenhouse conditions at the Faculty of Agriculture, Science and Technology, Universitas Panca Bhakti Pontianak, located at Jalan Komyos Sudarso, West Pontianak District, Pontianak City, West Kalimantan Province, Indonesia (0°01'S, 109°20'E). The experimental period extended from January through May 2025, encompassing a complete soybean growing cycle.

2.2. Soil Collection and Characterization

Alluvial soil samples were collected from the surface horizon (0-20 cm depth) of agricultural land representative of local soil conditions. Composite samples were obtained, air-dried, manually crushed, and passed through a 5-mm sieve to remove coarse debris and ensure uniformity. Initial soil characterization revealed acidic pH (5.2), low organic carbon content (1.8%), deficient available phosphorus (4.2 mg/kg Bray-1 extractable P), and moderate cation exchange capacity (18.3 cmol/kg). The soil textural composition comprised 14% sand, 76% silt, and 10% clay, classified as silt loam according to the USDA textural classification system.

2.3. Experimental Materials

High-yielding soybean seeds of the Grobogan variety were obtained from certified commercial sources. Commercial AMF inoculum (Mycofir brand) containing a consortium of arbuscular mycorrhizal species (predominantly *Glomus* spp.) was procured from registered biofertilizer producers. The inoculum contained colonized root fragments, spores, and mycelial networks in a carrier medium; spore density was ≥ 50 viable spores per gram inoculum as stated by the manufacturer. Colonization success was confirmed through root staining and microscopic observation at 21 DAP. Fresh *Moringa oleifera* leaves were harvested from healthy, mature trees and processed into aqueous extract by blending 200 g fresh leaf material with 1 L distilled water, followed by filtration through double-layered muslin cloth. All moringa extract treatment levels used the same 20% v/v concentration; only the application volume per event differed among E_1, E_2, and E_3 treatments.

2.4. Experimental Design and Treatment Structure

The experiment employed a factorial completely randomized design (CRD) incorporating two treatment factors: Factor 1 – AMF Inoculation (M): M_0 = 0 g/polybag; M_1 = 5 g/polybag; M_2 = 10 g/polybag. Factor 2 – Moringa Leaf Extract Volume (E): E_1 = 150 ml/polybag; E_2 = 200 ml/polybag; E_3 = 250 ml/polybag. The factorial arrangement generated nine treatment combinations, each replicated three times with three plants per experimental unit (81 plants total). AMF dose levels were selected based on established effective rates for soybean and food crops in tropical environments (Smith & Read, 2008; Ortas, 2012). Moringa extract volume was based on dose-response evidence from Emongor (2015) and Irshad *et al*. (2024). The experimental unit was the polybag; after thinning to one vigorous plant per polybag at 10 days after emergence (DAE), each polybag and its individual plant were equivalent as observational units. Black polyethylene polybags (30 cm diameter × 35 cm height) were filled with 10 kg air-dried soil per unit.

2.5. Cultural Management Practices

2.5.1. Soil Amendment

Agricultural dolomitic limestone was incorporated at 14 g/polybag two weeks before planting to raise soil pH toward the optimal range for soybean cultivation and mycorrhizal colonization (Somasegaran & Hoben, 1994).

2.5.2. Planting and AMF Inoculation

Three viable seeds were sown 3 cm deep in each polybag. AMF inoculum was applied at planting by placing the prescribed amount in direct contact with seeds within the planting hole. Seedlings were thinned to one vigorous plant per polybag at 10 DAE.

2.5.3. Moringa Leaf Extract Application

Moringa extract was applied as soil drench at three growth stages: 14, 28, and 42 days after planting (DAP). The designated volume for each treatment level was applied in full at each application event (E_1 = 150 mL/plant, E_2 = 200 mL/plant, E_3 = 250 mL/plant per event), yielding cumulative seasonal totals of 450, 600, and 750 ml/plant for E_1, E_2, and E_3, respectively. All treatment levels used identical 20% v/v concentration; supplemental irrigation was applied uniformly to maintain comparable soil moisture across treatments.

2.5.4. Routine Maintenance

Plants received uniform management including manual weed removal, pest monitoring, and supplemental irrigation to maintain soil moisture at approximately 70–80% field capacity. No chemical fertilizers or pesticides were applied.

2.6. Data Collection

At 32 DAP some parameters were observed: plant height (cm); leaf number; branch number; root nodule number; shoot dry weight (g/plant); root dry weight (g/plant); and shoot-to-root dry weight ratio. AMF root colonization (%) was assessed by root staining and gridline intersection method at 21 DAP (McGonigle *et al*., 1990). At physiological maturity observation included pod number per plant; dry seed weight (g/plant, sun-drying to 12% moisture content).

2.7. Statistical Analysis

Data were subjected to two-way factorial ANOVA to partition total variation into main effects (Factor M and Factor E) and their interaction (M×E). Means were separated using Fisher's Least Significant Difference (LSD) test at 5% probability when F-tests indicated significant effects. Statistical analyses were performed using Microsoft Excel 2019 following the factorial CRD procedures described by Gaspersz (1991), and results are presented as mean ± standard error of mean.

3. RESULTS

Table 1 summarizes results of the ANOVA test of the effect of treatment factor combinations on seven vegetative parameters and two generative parameters of soybean. The analysis results show that the calculated F value is greater than the F table for all factors and their interactions. Therefore, it can be concluded that the AMF inoculation factor (M) and the moringa leaf extract factor (E) and their interactions have a significant to very significant effect on all observed parameters. Table 2 further shows the effect of the M*E interaction on all vegetative and generative parameters of soybean plants.

Table 1. Summary of two-way ANOVA showing F-values and significance for single factor (M and E), and their interaction (M×E)

Variable	F – Factor M	Sig.	F – Factor E	Sig.	F – M×E	Sig.	KK (%)
Plant Height (cm)	7.23	**	4.89	**	3.21	*	8.72
Leaf Number	9.41	**	6.13	**	4.07	*	7.94
Branch Number	14.32	**	8.76	**	5.43	**	11.23
Root Nodule No.	42.18	**	23.67	**	6.89	**	7.16
Shoot DW (g)	17.94	**	11.23	**	4.31	*	9.48
Root DW (g)	98.47	**	27.34	**	8.12	**	5.87
Shoot/Root Ratio	8.67	**	5.34	**	3.47	*	13.24
Pod Number	71.23	**	19.84	**	6.23	**	6.91
Seed DW (g)	22.41	**	14.87	**	5.16	**	12.83

Note: ns = not significant ($P > 0.05$); * = significant at $P < 0.05$; ** = significant at $P < 0.01$. df: Factor M = 2, Factor E = 2, M×E = 4, Error = 18. KK = coefficient of variation (%).

Table 2. Effects of AMF inoculation and moringa leaf extract on growth, nodulation, and yield of soybean grown on alluvial soil

Treatment	Plant Height (cm)	Leaf Number	Branch Number	Root Nodule No.	Shoot DW (g)	Root DW (g)	Shoot/Root Ratio	Pod Number	Seed DW (g)
M_0E_1	20.3±0.7^{a}	15.3±0.6^{a}	1.0±0.0^{a}	10.3±1.7^{a}	1.4±0.8^{a}	0.7±0.1^{a}	2.1±1.1^{c}	42.5±5.7^{a}	16.7±7.8^{a}
M_0E_2	22.9±2.4^{ab}	21.6±2.5^{bc}	1.4±1.0^{a}	12.1±2.4^{a}	2.3±0.4^{b}	1.3±0.2^{b}	1.8±0.4^{bc}	51.5±5.4^{b}	26.5±1.8^{bc}
M_0E_3	25.8±6.5^{bc}	21.0±2.2^{b}	2.3±0.6^{b}	12.5±0.4^{a}	2.4±0.2^{b}	1.6±0.2^{c}	1.5±0.3^{ab}	63.5±4.5^{c}	27.2±4.0^{bc}
M_1E_1	26.5±3.6^{bc}	22.0±4.3^{bc}	2.4±0.2^{b}	12.4±0.2^{a}	2.6±0.2^{bc}	2.3±0.2^{d}	1.1±0.2^{a}	67.3±5.2^{c}	21.0±6.9^{ab}
M_1E_2	26.9±5.2^{c}	21.7±2.1^{bc}	2.7±0.6^{bc}	16.8±1.3^{b}	2.8±0.1^{cd}	2.5±0.2^{e}	1.1±0.1^{a}	78.0±2.5^{d}	29.0±0.5^{bc}
M_1E_3	27.4±3.0^{c}	23.6±0.9^{bcd}	2.8±0.5^{bc}	17.8±2.9^{b}	3.1±0.2^{de}	2.8±0.3^{f}	1.1±0.1^{a}	83.2±4.0^{de}	32.0±3.0^{c}
M_2E_1	27.7±2.2^{cd}	23.9±1.3^{cd}	2.6±0.2^{b}	20.6±0.2^{c}	2.8±0.2^{cd}	2.6±0.2^{ef}	1.1±0.1^{a}	85.0±7.5^{e}	34.1±10.6^{c}
M_2E_2	26.0±0.7^{bc}	24.9±1.5^{d}	3.3±0.6^{cd}	23.6±0.9^{d}	3.3±0.3^{e}	3.0±0.0^{g}	1.1±0.1^{a}	94.0±3.7^{f}	35.3±8.8^{c}
M_2E_3	31.5±2.3^{d}	25.3±3.3^{d}	3.6±0.5^{d}	25.3±3.8^{d}	3.8±0.3^{f}	3.6±0.2^{h}	1.1±0.1^{a}	97.2±4.0^{f}	48.6±17.9^{d}

Note: Values represent mean ± standard error (n=3). Different superscript letters within each column indicate significant differences among treatment combinations based on LSD test at $P < 0.05$. DW = Dry Weight.

3.1. Mycorrhizal Root Colonization

AMF colonization was exclusively observed in treatments receiving mycorrhizal inoculum, with no detectable colonization in non-inoculated controls. Root infection intensity exhibited a dose-dependent response pattern. The M_1 treatment (5 g/polybag) produced moderate colonization levels ranging from 28.3% to 35.7% across moringa extract volumes, while M_2 treatment (10 g/polybag) achieved substantially higher colonization rates between 42.1% and 51.6%. The interaction between AMF dose and moringa extract volume significantly influenced colonization patterns (Table 1), with higher moringa volumes enhancing colonization intensity particularly at the M_2 dose level, indicating that moringa leaf extract augments the colonization capacity of AMF inoculum under alluvial soil conditions.

3.2. Vegetative Growth Responses

Two-way factorial ANOVA revealed significant main effects of both AMF inoculation and moringa leaf extract, and importantly, significant interaction effects (M×E) for all measured vegetative parameters (Table 1). Plant height showed progressive increases with both AMF and moringa applications. Non-mycorrhizal controls exhibited the shortest stature (20.3–25.8 cm), while plants receiving the highest AMF dose combined with maximum moringa volume achieved the greatest height (31.5 cm in M_2E_3), representing a 55% increase over the lowest treatment combination (Table 2). The significant M×E interaction for plant height ($F = 3.21$, $P < 0.05$) indicates that the effect of moringa extract volume on plant height differs depending on the AMF dose level, with the highest response observed only when both inputs were applied at their maximum levels. Similarly, leaf production and branching intensity responded positively to combined treatments, with M_2E_3 combination producing 25.3 leaves and 3.6 branches compared to 15.3 leaves and 1.0 branches in M_0E_1 treatment.

3.3. Root Nodulation Dynamics

Root nodule formation demonstrated substantial enhancement through the combined application of AMF and moringa leaf extract. Factorial ANOVA revealed highly significant interaction effects ($F = 6.89$, $p < 0.01$) between AMF dose and moringa extract volume on root nodule number (Table 1), indicating that the two inputs do not act independently but rather produce synergistic effects on nodulation. Non-mycorrhizal plants developed 10.3–12.5 nodules plant^{-1} across moringa volumes. AMF inoculation at 5 g/polybag increased nodule numbers to 12.4–17.8 per plant, while the dose 10 g/polybag elevated nodulation to 20.6–25.3 nodules/plant. The maximum nodulation (25.3 nodules/plant) occurred in M_2E_3, representing a 146% increase relative to M_0E_1 control (Table 2).

3.4. Biomass Accumulation Patterns

Both shoot and root dry weight (DW) accumulation exhibited significant positive responses to treatment interventions, with significant M×E interaction effects (shoot DW: $F = 4.31$, $p < 0.05$; root DW: $F = 8.12$, $p < 0.01$) confirming that combined application of AMF and moringa extract produces effects beyond simple additive responses (Table 1). Shoot DW increased from 1.4 g/plant in M_0E_1 to 3.8 g/plant in M_2E_3 (171% increase). Root DW showed even more pronounced responses, increasing from 0.7 g/plant to 3.6 g/plant (414% increase) across the same treatment range. AMF inoculation induced a shift in biomass partitioning patterns, evidenced by significantly reduced shoot-to-root ratios in mycorrhizal treatments (1.1) compared to non-mycorrhizal controls (1.5–2.1), indicating greater relative investment in root system development in AMF-colonized plants (Table 2).

3.5. Reproductive Development and Yield Performance

Pod production and seed yield demonstrated the most dramatic treatment responses, with significant interaction effects between AMF and moringa extract for both pod number ($F = 6.23$, $p < 0.01$) and seed dry weight (DW) ($F = 5.16$, $p < 0.01$) as shown in Table 1. Pod number per plant varied from 42.5 in M_0E_1 to 97.2 in M_2E_3, representing a 129% increase (Table 2). Analysis of combined treatment effects indicates that when AMF application at 10 g/polybag was combined with moringa extract at 250 ml/polybag, the interactive enhancement in pod number exceeded what would be expected from either input alone. Seed DW exhibited similar response patterns, ranging from 16.7 g/plant in M_0E_1 to 48.6 g/plant in M_2E_3 (191% yield increase). The significant M×E interaction for seed DW demonstrates that the combination of AMF and moringa extract at specific dose levels produces synergistic yield enhancement beyond additive effects. Using M_0E_1 (no AMF, lowest moringa volume) as the appropriate non-mycorrhizal baseline, the M_2E_3 combination produced a 129% increase in pod number and a 191% increase in seed DW, confirming the substantial yield advantage of optimally combined biofertilizer treatments (Table 2).

4. DISCUSSION

4.1. Mycorrhizal Colonization Establishment

The successful establishment of arbuscular mycorrhizal symbiosis confirmed the effectiveness of the commercial inoculum and the receptivity of the Grobogan soybean variety to AMF colonization. The dose-dependent colonization response aligns with established principles of mycorrhizal inoculation technology (Cely *et al*., 2016). The significant interaction between AMF dose and moringa extract volume on colonization percentage suggests that moringa extract components may enhance colonization efficiency, particularly at higher AMF doses. This finding aligns with findings by Ghorui *et al*. (2025) that plant-derived biostimulants modify the rhizosphere's chemical environment to promote mycorrhizal establishment, likely by altering root exudate composition and stimulating mycorrhizal helper bacteria.

4.2. Interactive Effects on Vegetative Development

The significant interaction effects observed for all vegetative growth parameters (Table 1) indicate that AMF inoculation and moringa leaf extract do not act independently but rather exhibit synergistic interactions that collectively promote plant development beyond what either input achieves alone. This interactive enhancement can be attributed to complementary mechanisms: AMF colonization expands the nutrient absorption zone through extraradical hyphal networks (Martin & van der Heijden, 2024), while moringa extract supplies exogenous growth regulators and nutrients that stimulate metabolic processes (Irshad *et al*., 2024). Together, these complementary effects create favorable conditions for enhanced vegetative expression. Beyond direct nutritional benefits, AMF colonization has been reported to induce systemic physiological changes including enhanced photosynthetic efficiency through improved stomatal conductance (Nie *et al*., 2024) and modified phytohormone profiles (Shi *et al*., 2023). The moringa extract, rich in zeatin-type cytokinins, may amplify these hormonal effects, explaining the significant interaction. These specific mechanisms were inferred from published literature and warrant direct verification in future studies.

4.3. Moringa Extract as an Interactive Biostimulant

The significant interaction between moringa extract volume and AMF dose for all growth parameters demonstrates that moringa extract functions not merely as a standalone biostimulant but as an interactive partner that enhances AMF effectiveness. The growth-promoting properties of moringa extracts include high concentrations of zeatin-type cytokinins (Zulfiqar *et al*., 2020), readily available nutrients, and diverse secondary metabolites with antioxidant and stress-mitigating properties (Arif *et al*., 2023). The interactive enhancement observed in this study exceeds the 46% yield increase reported by Irshad *et al*. (2024) for moringa extract applied alone, and the improvements documented by Emongor (2015) in leguminous crops, confirming that synergistic interaction with AMF produces greater benefits than either input independently. The application timing (14, 28, and 42 DAP) targeting vegetative establishment, reproductive commitment, and pod filling may have maximized the interactive effects by ensuring continuous biostimulant supply during critical developmental transitions.

4.4. Synergistic Enhancement of Root Nodulation

The highly significant interaction effect on root nodulation ($F = 6.89$, $p < 0.01$) represents one of the most biologically meaningful findings of this investigation. This synergistic interaction suggests that AMF colonization and moringa extract application act through complementary pathways that together create optimal conditions for rhizobial colonization and nodule development. It should be noted that rhizobial inoculation was not applied in this study; the observed nodulation reflects natural rhizobial populations responding to the improved rhizosphere environment created by combined AMF-moringa treatment. The interactive enhancement of nodulation likely occurs through multiple mechanisms: improved phosphorus nutrition via AMF hyphal networks alleviates the energy constraint on nitrogen fixation (Peng *et al*., 2025), while moringa extract supplies phytohormones that stimulate root hair development and rhizobial attachment sites. Several mechanisms may contribute to this: first, improved phosphorus nutrition mediated by mycorrhizal colonization directly supports nodule development and nitrogenase activity (Peng *et al*., 2025); second, AMF colonization may influence rhizosphere microbial communities in ways that favor rhizobial proliferation (Subaedah *et al*., 2024); and third, moringa-derived cytokinins may amplify nodule development signals, creating an interactive enhancement beyond what either factor achieves alone (Igiehon *et al*., 2021).

4.5. Biomass Partitioning and Interactive Resource Allocation

The significant interaction effects on both shoot and root dry weight (DW) confirm that AMF-moringa combined treatment produces synergistic biomass enhancement. The pronounced shift in shoot-to-root ratios observed in mycorrhizal plants (from 1.5–2.1 to 1.1) reflects fundamental changes in biomass partitioning driven by the interactive effects of both inputs. Studies employing ^{13}C isotopic tracing have demonstrated that AMF structures receive between 10% and 23% of current host photosynthate as carbon transferred to the fungal partner (Zheng *et al*., 2024). Moringa extract application likely compensates for this carbon cost by supplying exogenous carbon compounds and growth regulators that enhance photosynthetic capacity, enabling the plant to simultaneously support both mycorrhizal development and productive shoot growth. This interactive carbon economy explains the significant M×E interaction observed for biomass parameters.

4.6. Interactive Yield Enhancement and Agronomic Implications

The highly significant M×E interactions for both pod number (F = 6.23, $p < 0.01$) and seed dry weight (DW) (F = 5.16, $p < 0.01$) confirm that the combination of AMF and moringa extract produces synergistic yield enhancement that cannot be predicted from individual factor effects alone. This finding has important agronomic implications: integrated application of both inputs is essential to realize the full yield potential demonstrated in this study. When compared to the Grobogan variety description, which specifies an average productivity of 2.77 t/ha and plant height of 50–60 cm (Dinas Pertanian DIY, 2008), plant heights observed in this study (20.3–31.5 cm) were below varietal potential, indicating that polybag conditions constrained full vegetative expression. However, the per-plant seed DW in M_2E_3 (48.6 g), when extrapolated to field density, suggests productivity approaching varietal targets, with field-scale validation being essential to confirm these projections. The combined treatment produced a three-fold increase in seed DW (from 16.7 to 48.6 g/plant), substantially exceeding improvements achievable through either AMF alone (average increase from 19.0 to 37.1 g across moringa levels) or moringa alone (from 22.8 to 33.3 g across AMF levels). This demonstrates the agronomic value of integrated biofertilizer strategies over single-input approaches. Both inputs are accessible to smallholder farmers in West Kalimantan and represent low-cost, environmentally compatible alternatives to conventional synthetic fertilizers.

4.7. Sustainability Considerations and Environmental Benefits

Beyond immediate productivity gains, the integrated application of mycorrhizal inoculants and plant-based biostimulants offers important sustainability advantages over conventional fertilizer-intensive production systems. AMF symbioses contribute to long-term soil health through improved aggregate stability, enhanced carbon sequestration, and increased functional diversity of soil microbial communities (Rillig & Mummey, 2006). The local availability and renewable nature of moringa leaf extract resources provide additional sustainability benefits, particularly in tropical regions where *Moringa oleifera* grows vigorously as a perennial multipurpose tree. Smallholder farmers can produce moringa extracts on-farm with minimal capital investment, reducing input costs and improving economic viability of legume production on marginal lands.

4.8. Limitations and Future Research Directions

Several aspects warrant consideration when interpreting results. The controlled polybag environment differs substantially from field conditions where temperature fluctuations, precipitation variability, and soil heterogeneity introduce additional complexity. The relatively short experimental duration precluded assessment of residual effects. Future investigations should examine field-scale validation across multiple locations, rhizobial co-inoculation treatments, economic feasibility assessments, and molecular studies examining gene expression patterns and hormone profiles to elucidate mechanistic details of the observed interactive effects.

5. CONCLUSIONS

This investigation demonstrates that integrated application of arbuscular mycorrhizal fungi inoculation and moringa leaf extract produces substantial improvements in soybean growth, nodulation, and yield on alluvial soils. Two-way factorial ANOVA confirmed significant main effects of both AMF inoculation and moringa leaf extract, and crucially, significant interaction effects (M×E) for all measured parameters ($p < 0.05$), demonstrating that these two inputs interact synergistically rather than independently. The optimal treatment combination of 10 g AMF/polybag with 250 ml/polybag moringa extract (M_2E_3) achieved the highest values across all variables: 31.5 cm plant height, 25.3 root nodules, 3.8 g shoot DW, 3.6 g root DW, 97.2 pods/plant, and 48.6 g seed DW per plant, representing increases of 55%, 146%, 171%, 414%, 129%, and 191%, respectively, over the M_0E_1 control. These findings directly address the study objectives: (i) significant M×E interactive effects were confirmed for all growth and yield parameters; (ii) AMF and moringa combined treatment significantly enhanced root nodulation beyond individual factor effects; and (iii) M_2E_3 consistently produced the highest yield responses across all parameters. Field-scale validation studies are warranted to confirm these interactive effects under diverse agroecological conditions before broad agronomic recommendations can be established.

ACKNOWLEDGMENTS

The authors express sincere gratitude to Universitas Panca Bhakti for providing financial support and research facilities for this investigation. We thank the technical staff of the Faculty of Agriculture greenhouse facility for their assistance with experimental maintenance and data collection activities.

AUTHOR CONTRIBUTION STATEMENT

Author	C	M	So	Va	Fo	I	R	D	O	E	Vi	Su	P	Fu
ATPI	✓			✓		✓	✓		✓	✓	✓	✓		✓
AS		✓		✓	✓	✓		✓		✓	✓			✓
SO			✓			✓			✓	✓	✓		✓	
EK			✓		✓	✓		✓		✓	✓		✓	

C: Conceptualization	Fo: Formal Analysis	O: Writing - Original Draft	Fu: Funding Acquisition
M: Methodology	I: Investigation	E: Writing - Review & Editing	P: Project Administration
So: Software	D: Data Curation	Vi: Visualization	
Va: Validation	R: Resources	Su: Supervision	

REFERENCES

Adie, M.M., & Krisnawati, A. (2017). Variability of pod trichome and agronomic characters of several soybean genotypes. *Biodiversitas Journal of Biological Diversity, **18***(1). https://doi.org/10.13057/biodiv/d180153

Aloo, B.N., Makumba, B.A., & Mbega, E.R. (2019). The potential of Bacilli rhizobacteria for sustainable crop production and environmental sustainability. *Microbiological Research, **219***, 26–39. https://doi.org/10.1016/j.micres.2018.10.011

Arif, Y., Bajguz, A., & Hayat, S. (2022). *Moringa oleifera* extract as a natural plant biostimulant. *Journal of Plant Growth Regulation, **42***(3), 1–16. https://doi.org/10.1007/s00344-022-10630-4

BPS (Badan Pusat Statistik). (2020). *Provinsi Kalimantan Barat dalam angka 2020*. Badan Pusat Statistik Provinsi Kalimantan Barat. https://kalbar.bps.go.id/publication/2020/04/27/

Cely, M.V.T., de Oliveira, A.G., de Freitas, V.F., de Luca, M.B., Barazetti, A.R., dos Santos, I.M.O., Gionco, B., Garcia, G.V., Prete, C.E.C., & Andrade, G. (2016). Inoculant of arbuscular mycorrhizal fungi (*Rhizophagus clarus*) increase yield of soybean and cotton under field conditions. *Frontiers in Microbiology, **7***, 720. https://doi.org/10.3389/fmicb.2016.00720

Dinas Pertanian dan Ketahanan Pangan Daerah Istimewa Yogyakarta. (2008). *Deskripsi Varietas Kedelai Grobogan* (SK Mentan No. 238/Kpts/SR.120/3/2008).

Emongor, V.E. (2015). Effects of *Moringa oleifera* leaf extract on growth, yield and yield components of snap beans (*Phaseolus vulgaris*). *Current Journal of Applied Science and Technology*, ***6***(2), 114–122. https://doi.org/10.9734/BJAST/2015/14795

Fageria, N.K., & Moreira, A. (2011). The role of mineral nutrition on root growth of crop plants. *Advances in Agronomy, **110***, 251–331. https://doi.org/10.1016/B978-0-12-385531-2.00004-9

Gaspersz, V. (1991). *Metode Perancangan Percobaan untuk Ilmu-Ilmu Pertanian, Ilmu-Ilmu Teknik, Biologi*. Armico, Bandung.

Ghorui, M., Chowdhury, S., & Burla, S. (2025). Recent advances in the commercial formulation of arbuscular mycorrhizal inoculants. *Frontiers in Industrial Microbiology, **3***, 1553472. https://doi.org/10.3389/finmi.2025.1553472

Havlin, J.L., Tisdale, S.L., Nelson, W.L., & Beaton, J.D. (2014). *Soil Fertility and Fertilizers: An Introduction to Nutrient Management* (8th ed.). Pearson Education.

Igiehon, N.O., Babalola, O.O., Cheseto, X., & Torto, B. (2021). Effects of rhizobia and arbuscular mycorrhizal fungi on yield, size distribution and fatty acid of soybean seeds grown under drought stress. *Microbiological Research, **242***, 126640. https://doi.org/10.1016/j.micres.2020.126640

Irshad, S., Matloob, A., Ghaffar, A., Hussain, M.B., & Tahir, M.H.N. (2024). Agronomic and biochemical aspects of *Moringa* dried leaf extract mediated growth and yield improvements in soybean. *New Zealand Journal of Crop and Horticultural Science, **53***(5), 1510–1529. https://doi.org/10.1080/01140671.2024.2374066

Kementerian Pertanian. (2023). *Statistik Pertanian 2023*. Pusat Data dan Sistem Informasi Pertanian Kementerian Pertanian Republik Indonesia, Jakarta.

Martin, F.M., & van der Heijden, M.G.A. (2024). The mycorrhizal symbiosis: Research frontiers in genomics, ecology, and agricultural application. *New Phytologist,* ***242***(4), 1486–1506. https://doi.org/10.1111/nph.19541

Mashamaite, C.V., Ngcobo, B.L., Manyevere, A., Bertling, I., & Fawole, O.A. (2022). Assessing the usefulness of *Moringa oleifera* leaf extract as a biostimulant to supplement synthetic fertilizers: A review. *Plants,* ***11***(17), 2214. https://doi.org/10.3390/plants11172214

McGonigle, T.P., Miller, M.H., Evans, D.G., Fairchild, G.L., & Swan, J.A. (1990). A new method which gives an objective measure of colonization of roots by vesicular-arbuscular mycorrhizal fungi. *New Phytologist,* ***115***(3), 495–501. https://doi.org/10.1111/j.1469-8137.1990.tb00476.x

Nie, W., He, Q., Guo, H., Zhang, W., Ma, L., Li, J., & Wen, D. (2024). Arbuscular mycorrhizal fungi: Boosting crop resilience to environmental stresses. *Microorganisms,* ***12***(12), 2448. https://doi.org/10.3390/microorganisms12122448

Ortas, I. (2012). The effect of mycorrhizal fungal inoculation on plant yield, nutrient uptake and inoculation effectiveness under long-term field conditions. *Field Crops Research,* ***125***, 35–48. https://doi.org/10.1016/j.fcr.2011.08.005

Peng, Z.-C., Xing, Y.-X., Ma, Y.-H., Li, S.-H., Jia, Y.-X., Yang, H.-C., & Zhang, F.-H. (2025). Arbuscular mycorrhizal fungi enhance soybean phosphorus uptake and soil fertility under saline-alkaline stress. *Scientific Reports,* ***15***, 31792. https://doi.org/10.1038/s41598-025-15910-z

Rillig, M.C., & Mummey, D.L. (2006). Mycorrhizas and soil structure. *New Phytologist,* ***171***(1), 41–53. https://doi.org/10.1111/j.1469-8137.2006.01750.x

Shi, J., Wang, X., & Wang, E. (2023). Mycorrhizal symbiosis in plant growth and stress adaptation: From genes to ecosystems. *Annual Review of Plant Biology,* ***74***, 569–607. https://doi.org/10.1146/annurev-arplant-061722-090342

Smith, J.E. (2009). *Mycorrhizal symbiosis* (3rd ed.). *Soil Science Society of America Journal,* ***73***(2). https://doi.org/10.2136/sssaj2008.0015br

Somasegaran, P., & Hoben, H.J. (1994). *Handbook for rhizobia: Methods in legume-rhizobium technology* (1st ed.). Springer.

Subaedah, S., Netty., Nonci, M., Edy, & Sabahannur, S. (2024). Effect of application of arbuscular mycorrhizal fungi on growth and yield of soybean in different agroecosystems. *IOP Conference Series: Earth and Environmental Science,* ***1302***(1), 012039. https://doi.org/10.1088/1755-1315/1302/1/012039

Uroz, S., Courty, P.E., & Oger, P. (2019). Plant symbionts are engineers of the plant-associated microbiome. *Trends in Plant Science,* ***24***(10), 905–916. https://doi.org/10.1016/j.tplants.2019.06.008

Zheng, L., Zhao, S., Zhou, Y., Yang, G., Chen, A., Li, X., Wang, J., Tian, J., Liao, H., & Wang, X. (2024). The soybean sugar transporter GmSWEET6 participates in sucrose transport towards fungi during arbuscular mycorrhizal symbiosis. *Plant, Cell & Environment,* ***47***(4), 1041–1052. https://doi.org/10.1111/pce.14772

Zulfiqar, F., Casadesús, A., Brockman, H., & Munné-Bosch, S. (2020). An overview of plant-based natural biostimulants for sustainable horticulture with a particular focus on moringa leaf extracts. *Plant Science,* ***295***, 110194. https://doi.org/10.1016/j.plantsci.2019.110194