Soil Cover Influences the Effectiveness of Microbial Biofertilisers on the Growth and Yield of Papaya (*Carica papaya* L.)

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Article

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Abstract

In a field experiment with papaya (*Carica papaya* L.), the effects of three soil cover types (bare soil, living cover of *Canavalia ensiformis*, and senescent cover) and three microbial biofertilisers (*Bacillus subtilis*, *Pseudomonas putida*, and *Trichoderma viride*) on crop growth and yield were evaluated. Vegetative and reproductive variables were monitored over 187 days, and the data were analysed using generalised linear mixed models (GLMMs) and generalised linear models (GLMs). The results indicated that soil cover was the dominant factor, explaining the largest proportion of variation in plant growth and final yield ($p < 0.001$), whereas biofertilisers did not exhibit significant main effects when applied independently. However, significant interactions between soil cover and biofertiliser were detected ($p < 0.05$), demonstrating that inoculant efficacy was strongly context-dependent. *Trichoderma viride* increased stem diameter by approximately 7% under living cover only, while *Pseudomonas putida* showed a comparative advantage under bare soil conditions, increasing final fruit weight by approximately 32%. Principal component analysis (PCA) further confirmed that treatment groupings were primarily driven by soil cover type. These findings provide field-based evidence that the efficiency of microbial biofertilisers in promoting papaya growth depends on edaphic conditions shaped by soil cover management. A hierarchical management strategy is therefore proposed, in which establishing a favourable soil habitat through plant cover is a prerequisite for maximising the benefits of microbial inoculants in tropical fruit production systems.

Keywords: agroecosystem management; plant–microorganism interactions; soil cover; GLMM; sustainable fruit production; context-dependent efficacy

1. Introduction

Agricultural intensification has increased reliance on synthetic fertilisers, undermining the sustainability of production systems through soil degradation, environmental pollution, and elevated associated emissions [1]. In response, ecological management strategies are increasingly promoted to sustain productivity while reducing dependence on external inputs, particularly those centred on soil management as a fundamental component of agroecosystem functioning [2].

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Plant cover, whether living or senescent, functions as a key regulator of the rhizosphere habitat by modulating soil temperature and moisture, supplying organic matter, stimulating microbial activity, and enhancing nutrient cycling, thereby delivering multiple ecosystem services [3–5]. Through these integrated mechanisms, cover crops can exert hierarchical control over edaphic processes that ultimately shape crop responses.

Microbial biofertilisers, including plant growth–promoting rhizobacteria (PGPR) and fungi such as *Bacillus*, *Pseudomonas*, and *Trichoderma*, can enhance plant nutrition by solubilising nutrients, producing phytohormones, and stimulating root development [6,7]. However, these beneficial effects do not always translate into consistent performance under field conditions.

Despite their potential, the adoption of biofertilisers remains limited due to the high variability of their performance under field conditions, where effects are often inconsistent or even negligible [8,9]. This discrepancy between outcomes observed in controlled environments and those in real agricultural systems has reinforced the notion that inoculant efficacy is not an intrinsic trait, but rather a dependent function of prevailing abiotic and biotic soil conditions, a phenomenon commonly referred to as context-dependent effectiveness [10–12].

It has been suggested that targeted management of the rhizosphere habitat, particularly through plant cover, can create a favourable edaphic context that enhances the stability and effectiveness of microbial biofertilisers [13]. However, the specific interactions between different cover types and inoculant performance remain insufficiently understood, especially in perennial fruit systems, where long-term crop–soil dynamics play a critical role in shaping treatment outcomes.

Papaya (*Carica papaya* L.) cultivation provides an appropriate model system for addressing this issue due to its economic significance in tropical regions, rapid growth rate, and well-defined phenological cycle, which enables the assessment of sequential responses from vegetative establishment to reproductive output under field conditions [14,15].

Therefore, the objective of this study was to assess how soil cover type modulates the response of *Carica papaya* L. to different microbial biofertilisers under field conditions. We hypothesised that (i) soil cover constitutes the determinant factor of crop growth and yield, and (ii) the efficacy of microbial biofertilisers is strictly context-dependent, becoming evident only under specific combinations of soil cover and inoculant.

2. Materials and Methods

2.1. Experimental Site and Edaphoclimatic Conditions

The study was conducted from November 2024 to September 2025, in the district of Jaén (province of Jaén, Cajamarca region, Peru), located in a transitional zone between the northern highlands and the high jungle. The experimental site lies at an altitude of 720–900 m.a.s.l. within the middle basin of the Marañón River, an area characterised by undulating to rugged terrain, inter-Andean valleys, river terraces, and moderate to steep slopes [16]. The local climate is semi-arid, warm and humid throughout the year, with mean temperatures ranging from 29 to 33 °C and annual precipitation between 900 and 1200 mm, according to the Peruvian National Meteorological and Hydrological Service (SENAMHI) [17]. This seasonal pattern strongly influences soil moisture dynamics throughout the year.

Soils in the study area are highly heterogeneous. At the regional scale, associations of Andosols, Cambisols, Fluvisols, Regosols, and Leptosols predominate [16]. Within the experimental plot, initial physicochemical analyses indicated a sandy clay loam soil texture [18,19], with a pH of 8.0, electrical conductivity of 14.9 $mS \cdot m^{-1}$, 3.4% organic matter, 1.7 $mg \cdot g^{-1}$ total nitrogen, 13.87 $mg \cdot kg^{-1}$ available phosphorus, 206.6 $mg \cdot kg^{-1}$ available potassium, and 2.09% equivalent calcium carbonate. This combination of soil properties confers moderate water-retention capacity and adequate drainage, making it representative of the edaphic conditions under which tropical crops are commonly established in the region.

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The soil at the study site in the district of Jaén, Cajamarca region, exhibits a sandy clay loam texture, reflecting a balanced proportion of sand, silt, and clay that is generally favourable for agricultural production. Based on available soil information and in the absence of a detailed description of diagnostic horizons, the soil is provisionally classified as an Inceptisol according to the USDA Soil Taxonomy. This order comprises soils with incipient pedogenic development and limited horizon differentiation, which are widely distributed in tropical and subtropical agricultural regions [20]. Under the World Reference Base for Soil Resources (WRB) system, the studied soil is classified as Cambisol, a reference soil group representing young to moderately developed soils common in mountainous and cultivated areas of the Andean region of Peru [21].

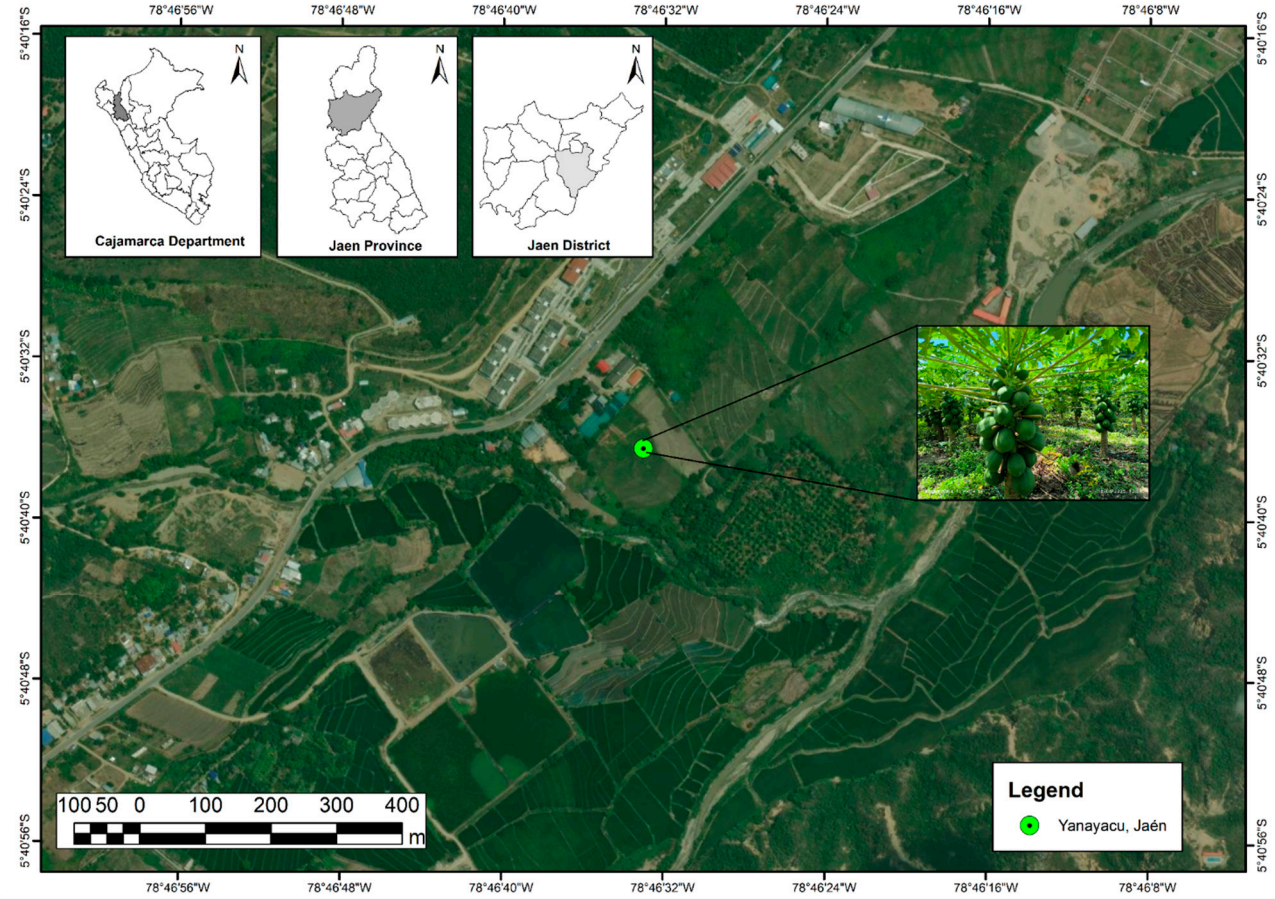

Figure 1. Geographic location of the experimental site. Study area: 48.5 × 45.5 m². UTM coordinates (WGS84): E = 746,342.4; N = 9,372,050.15; altitude = 645 m.a.s.l.

2.2. Experimental Design and Treatments

The experiment was established using a randomised complete block design with three blocks and nine treatments. Each treatment comprised an experimental plot with 14 papaya plants, yielding a total of 27 plots and 378 plants across the experiment. Each plant constituted an independent experimental unit. To minimise edge effects, only the central plants within each plot were used for data collection. Two experimental factors were evaluated: soil cover (bare soil, living cover of *Canavalia ensiformis*, and senescent cover) and microbial biofertilisers (*Bacillus subtilis*, *Pseudomonas putida*, and *Trichoderma viride*).

Table 1. Experimental treatments in the papaya field trial.

Treatment code	Soil cover	Biofertiliser	Repetitions
T1	Bare soil	*Bacillus subtilis*	3
T2	Bare soil	*Pseudomonas putida*	3

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T3	Bare soil	*Trichoderma viride*	3
T4	Living cover (*Canavalia ensiformis*)	*Bacillus subtilis*	3
T5	Living cover (*Canavalia ensiformis*)	*Pseudomonas putida*	3
T6	Living cover (*Canavalia ensiformis*)	*Trichoderma viride*	3
T7	Senescent cover (leaf litter)	*Bacillus subtilis*	3
T8	Senescent cover (leaf litter)	*Pseudomonas putida*	3
T9	Senescent cover (leaf litter)	*Trichoderma viride*	3

2.3. Crop Establishment and Management

2.3.1. Plant Material and Sowing

Certified seeds of the hybrid papaya variety Sinta F1 were obtained and soaked in water for 48 h before sowing. Seeds were sown in polyethylene nursery bags containing a substrate composed of soil, compost, and biochar in a 3:2:1 ratio, respectively. Germination commenced approximately 10 days after sowing, and seedlings were maintained under regular irrigation until they reached a height of 15–20 cm within 45 days. Subsequently, papaya seedlings were transplanted to the field at a spacing of 2.5 m between plants and 3.0 m between furrows.

2.3.2. Establishment and Management of Soil Cover

The senescent cover, composed of cocoa leaf litter and rice straw, was applied one week after the papaya plants were sown.

The living cover, consisting of *Canavalia ensiformis*, was also established one week after papaya plants were sown to prevent excessive growth that could otherwise overtop or suppress the papaya plants' growth. *Canavalia* was sown at 40 cm spacing between papaya plants, in accordance with the experimental treatments. The senescent cover was applied as a uniform mulch layer approximately 5 cm thick. The living cover was established at an average density of approximately 9 plants·m^{-2} and was managed through periodic cutting to minimise direct competition with papaya.

2.3.3. Preparation and Application of Biofertilisers

The microbial biofertilisers, supplied by the National Institute of Agrarian Innovation (INIA), consisted of fermented broth formulations of *Bacillus subtilis* strain BAC F (NCBI accession: MT982637), *Pseudomonas putida* strain P3 (NCBI accession: MT982624), and *Trichoderma viride* strain INIA (isolated from the INIA strain bank) [22]. The initial concentration of each formulation was 1 × 10^{8} CFU·mL^{-1}.

For each application, 1 L of each fermented broth was diluted separately in 200 L of water, with 2 kg of sugar added as a readily available carbon source to stimulate microbial activity. This procedure yielded an application suspension with a final concentration of approximately 1 × 10^{6} CFU·mL^{-1} for each microorganism. Sugar was added uniformly across all inoculated treatments to enhance microbial activity and, therefore, does not represent a confounding factor in treatment comparisons.

Two sequential applications were performed: the first at 15 days after transplanting (DAT) and the second at 90 DAT (approximately three months later). At each inoculation, a uniform volume of 1.5 L of the respective microbial suspension was applied per plant as a soil drench around the root collar, ensuring effective distribution within the rhizosphere.

2.3.4. Baseline Soil Management and Fertilisation

All plots received uniform agronomic management during the experimental period. Fertigation was applied every 15 days using urea (2,349 kg), diammonium phosphate (2,833 kg) and potassium chloride (3,422 kg), which were diluted in a tank with a capacity of 2,500 L of water, in accordance with local technical recommendations for papaya cultivation. This baseline management was implemented to ensure that the differences observed among the evaluated variables were attributable

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exclusively to the soil cover and biofertilisation treatments, and not to variations in nutrient availability.

2.4. Response Variables and Monitoring

2.4.1. Vegetative Growth and Development Variables

Plant height and stem diameter were measured monthly for 7 months (November 2024 to May 2025) following papaya plant sowing. Plant height was recorded using a measuring tape, and stem diameter was measured with a Mitutoyo digital vernier calliper.

Leaf number was recorded monthly for 5 months (November 2024 to March 2025), as lower leaves were pruned after March to improve canopy aeration and crop management.

2.4.2. Reproductive and Yield Variables

The number of flowers was recorded monthly over six months (December 2024 to May 2025). The number of fruits was assessed monthly over five months (January 2025 to May 2025). Yield was evaluated from May to September 2025. Fruits were harvested at marketable physiological maturity and weighed individually, and yields were quantified for each experimental treatment.

2.5. Data Analysis

Growth data evaluated at each sampling period were analysed using generalised linear mixed models (GLMMs), whereas final yield variables were analysed using generalised linear models (GLMs). In both cases, parameter estimation was performed using the *glmmTMB* package [23]. Statistical model selection was based on the Akaike information criterion (AIC) using the *MuMIn* package [24]. Model validation was conducted using the *DHARMa* package [25], assessing the assumptions of residual uniformity, absence of dispersion, homogeneity of variances, and absence of outliers. Subsequently, estimated marginal means were calculated using the *emmeans* package [26]. Statistical significance was evaluated at $\alpha = 0.05$, and multiple comparisons were adjusted using the False Discovery Rate (FDR) correction method, which controls the expected proportion of false positives while maintaining adequate statistical power under multiple testing conditions [27].

Additionally, principal component analysis (PCA) was performed using the *FactoMineR* package [28] after standardising the variables. The number of retained components was determined based on the Kaiser criterion (eigenvalues > 1) [29] and visual inspection of the sedimentation plot [30]. Component scores were plotted in a Cartesian plane, with variables and treatments simultaneously projected in a biplot, enabling examination of multivariate associations and identification of treatments associated with high or low values of the response variables.

All analyses were conducted using *R* software (version 4.5.0) [31] together with the *RStudio* development environment [32] for data processing and analysis. Both tools are freely available; R is open-source, and RStudio provides an environment that facilitates project management and the use of specialised packages for statistical analysis and data visualisation.

Variables evaluated at multiple time points were analysed using generalised linear mixed models (GLMMs), with plant and evaluation time included as random effects. Final yield variables were analysed using generalised linear models (GLMs). Model selection was based on Akaike's Information Criterion (AIC), and the significance of fixed effects was assessed using likelihood ratio tests.

3. Results

3.1. Temporal Dynamics of Vegetative Development

Stem diameter growth in *Carica papaya* L. exhibited pronounced temporal variation, with significant differences among treatments primarily observed during the early stages of development

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(Figure 2). During the initial period (7–36 days after intervention), the greatest increases in diameter were recorded in treatments with living and senescent cover, whereas treatments without cover (bare soil) generally showed lower growth rates. Between 36 and 67 days, differences among treatments diminished, although some covered treatments maintained higher values than uncovered treatments. From 67 days onward, stem diameter growth tended to become progressively homogenised, and no significant differences were detected between treatments from 98 to 187 days, indicating stabilisation of radial stem growth.

A similar pattern was observed for longitudinal growth (Figure 3). During the early developmental stages (7–36 and 36–67 days), significant differences among treatments were observed, with greater increases primarily associated with living cover treatments. However, from 67 days onward, longitudinal growth was statistically homogeneous among treatments, with occasional differences of low magnitude during intermediate periods and complete convergence toward the end of the evaluation period.

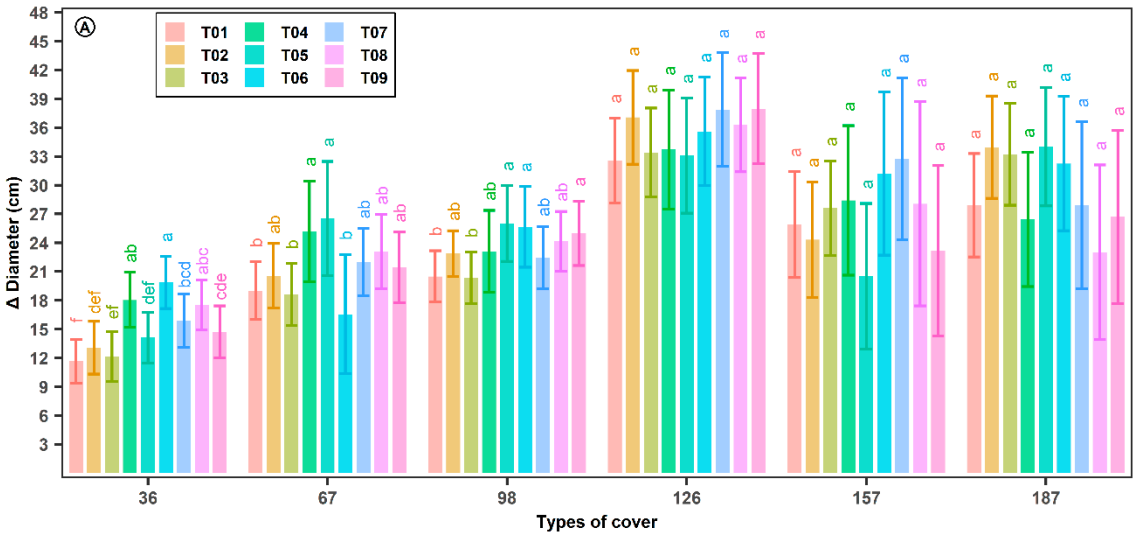

Figure 2. Stem diameter growth of *Carica papaya* L. across different evaluation periods.

T01, T02, and T03 corresponded to plots without cover, inoculated with the microbial biofertilisers *Bacillus subtilis*, *Pseudomonas putida*, and *Trichoderma viride*, respectively. T04, T05, and T06 included living cover with *Canavalia ensiformis* combined with the same biofertilisers in the indicated order. T07, T08, and T09 incorporated senescent cover (leaf litter) together with the microbial biofertilisers in the same order.

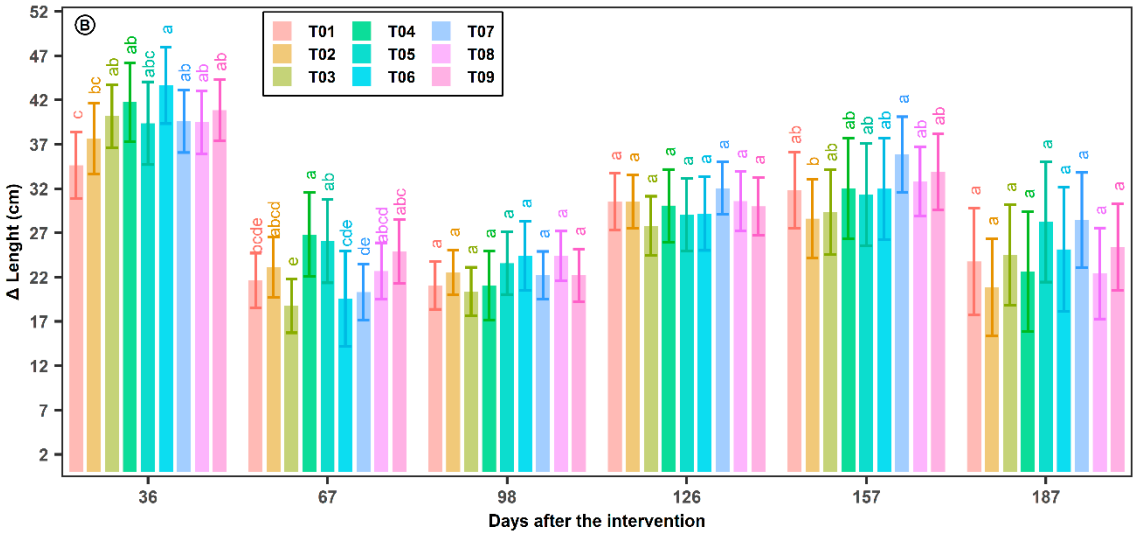

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Figure 3. Longitudinal growth of *Carica papaya* L. across different evaluation periods. Different letters among treatments indicate significant differences within each evaluation period at a significance level of 0.05, based on the FDR multiple comparison correction.

3.2. Leaf Development and Reproductive Transition

The number of flowers exhibited temporal fluctuations throughout the evaluation period (Figure 4). During the initial flowering stage (36–67 days) and the intermediate stage (98–126 days), treatments associated with soil cover showed greater increases compared with other treatments, whereas during the adjacent periods (67–98 days and 126–157 days), flower production increase was largely homogeneous across treatments. In the final period (157–187 days), a generalised decline in flower number was observed across all treatments, indicating a similar magnitude of flower loss regardless of the management strategy applied.

Regarding the number of leaves (Figure 5), a heterogeneous response pattern was observed during the early stages of vegetative development. During the initial periods (7–36 and 36–67 days), the largest increases in the number of leaves were primarily associated with treatments with living cover, whereas treatments without cover tended to exhibit the lowest values. In later stages (67–126 days), differences among treatments persisted at specific time points, although no consistent pattern was observed toward the end of the evaluation period.

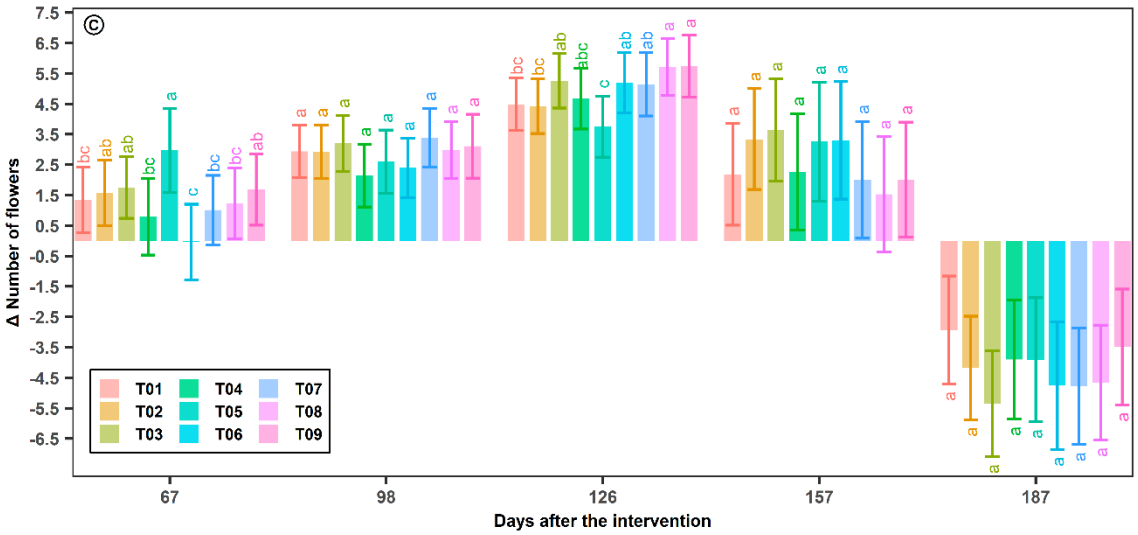

Figure 4. Increase in the number of flowers of *Carica papaya* L. across different evaluation periods. Different letters among treatments indicate significant differences within each evaluation period at a significance level of 0.05, based on the FDR multiple comparison correction.

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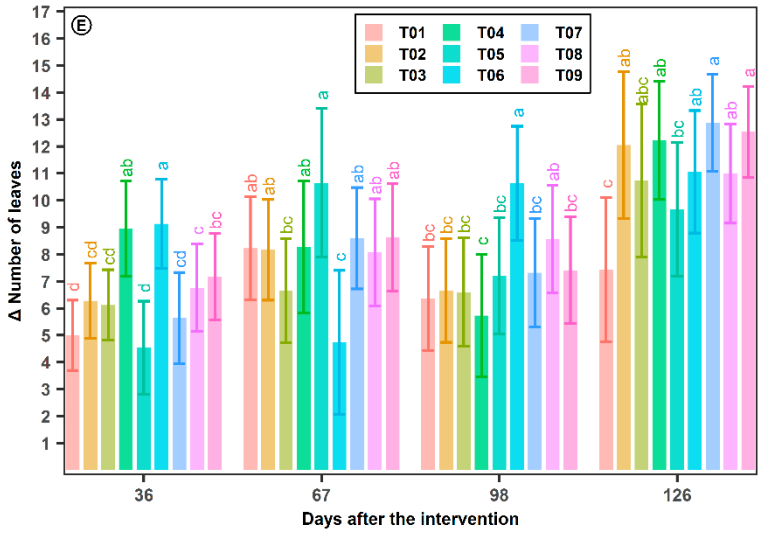

Figure 5. Increase in the number of leaves of *Carica papaya* L. across different evaluation periods. Different letters among treatments indicate significant differences within each evaluation period at a significance level of 0.05, based on the FDR multiple comparison correction.

3.3. Global Significance Analysis of the Evaluated Factors

Analysis using generalised linear mixed models (GLMMs) identified significant effects of the evaluated factors on the growth variables of *Carica papaya* L. throughout the experimental period. Soil cover, microbial biofertiliser, and time had different effects across the analysed variables.

Significant three-way interactions (cover × biofertilizer × time) were detected for stem diameter, length, number of leaves, and number of flowers ($p < 0.05$), indicating that crop responses varied over time depending on the combination of these factors. In contrast, the number of fruits did not exhibit a significant three-way interaction ($p > 0.05$); however, it showed a significant interaction between cover and time ($p < 0.05$) as well as a significant main effect of microbial biofertilizer ($p < 0.05$), as detailed in Table 2.

The inclusion of random effects associated with plant and time improved model fit primarily for the number of fruits, whereas for the remaining variables, the variability attributable to these random effects was low relative to the fixed effects.

For variables evaluated at the final measurement (stem diameter, plant length, number of leaves, number of flowers, number of fruits, and fruit weight), analysis of variance (ANOVA) indicated that soil cover had a significant effect on all analysed variables ($p < 0.05$), whereas microbial biofertiliser, when considered independently, showed no significant effects ($p > 0.05$). However, the interaction between soil cover and microbial biofertiliser was significant for stem diameter and number of leaves ($p < 0.05$) and showed marginal trends for plant length and fruit weight ($p < 0.1$), as detailed in Table 3.

Table 2. ANOVA of longitudinal growth of *Carica papaya* L.

Factors	Stem diameter (cm)	Plant length (cm)	Number of flowers	Number of fruits	Number of leaves
Fixed effects (p-values)					
Block	0.00	0.00	0.17	0.00	0.00
Cover	0.00	0.01	0.64	0.00	0.00
Biofertiliser	0.12	0.00	0.51	0.04	0.18
Days	0.00	0.69	0.00	0.00	0.00
Cover * Biofertiliser	0.00	0.00	0.00	0.41	0.00
Cover * Days	0.24	0.30	0.28	0.04	0.00

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Biofertiliser * Days		0.27	0.00	0.81	0.68	0.15
Cover * Biofertiliser * Days		0.00	0.03	0.01	0.19	0.00
Distribution selection						
Distribution		T	T	T	Normal	T
Link		Identity	Identity	Identity	Identity	Identity
Akaike Information Criterion (AIC)		17983	17237	9790	9315	9147
Random effects ($\hat{\sigma}$)						
Intercept	Plant	0.00	0.00	0.00	0.68	0.00
	Days	0.00	0.00	-	1.00	0.00

*C–M–P: Conway–Maxwell–Poisson distribution.

3.4. Multivariate Validation Using Principal Component Analysis

Principal component analysis (PCA) explained 82% of the total variance in the dataset across the first two components (Figure 6). The first principal component consistently accounted for the growth and production variables evaluated, including leaf number, stem diameter, plant length, fruit number, flower number, and fruit weight.

The projection of treatments onto the factorial plane (Figure 6B) revealed a clear separation between plots with soil cover and those without cover. Treatments without cover (T01–T03) clustered in the region associated with relatively low values of the number of leaves, stem diameter, plant length, and number of fruits. In contrast, treatments with living cover (T04–T06) and senescent cover (T07–T09) clustered in the region associated with higher values of these variables, as well as higher numbers of flowers and greater fruit weight, regardless of the microbial biofertilizer applied.

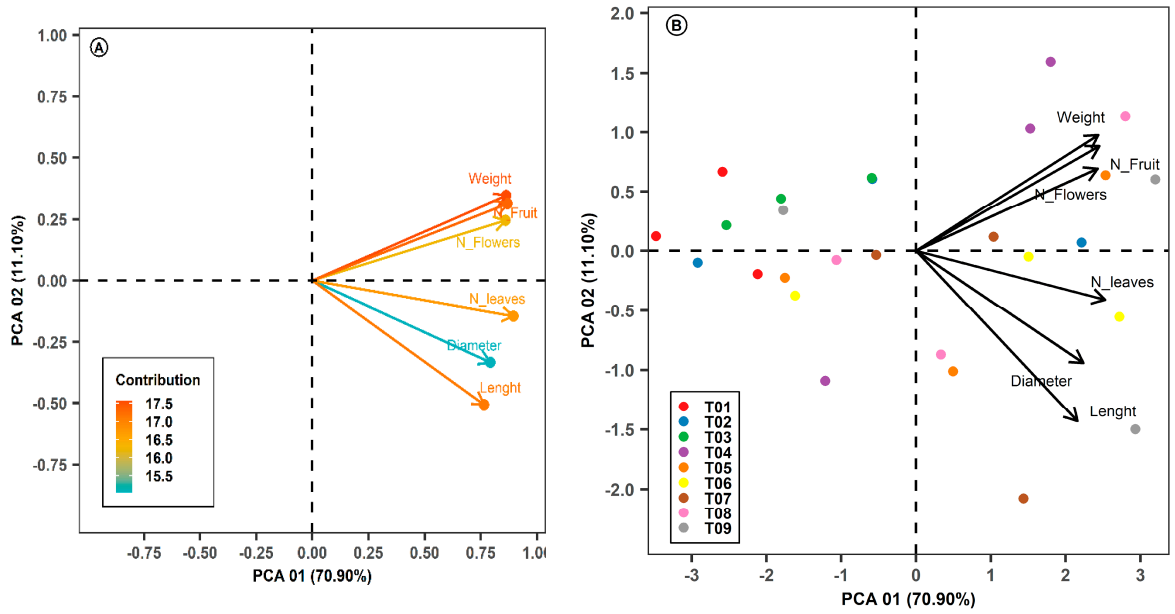

Figure 6. PCA of morphological and productive variables of papaya. Ⓐ Contribution of plant and fruit traits of *Carica papaya* L. to axis formation. Ⓑ Biplot of plant and fruit traits of *Carica papaya* L. T01, T02, and T03 correspond to plots without cover, inoculated with the microbial biofertilisers *Bacillus subtilis*, *Pseudomonas putida*, and *Trichoderma viride*, respectively. T04, T05, and T06 include living cover with *Canavalia ensiformis* combined with the same biofertilisers in the indicated order. T07, T08, and T09 incorporate senescent cover (leaf litter) together with the aforementioned biofertilisers in the same order.

3.5. Fruit Production and Weighted Yield

The number of fruits of *Carica papaya* L. showed a differentiated response throughout the experimental period depending on soil cover type (Figure 7A). Between 67 and 157 days after intervention, treatments with living and senescent cover exhibited significantly greater increases in

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the number of fruits compared with plots without cover. In contrast, during the final evaluation period (157–187 days), increases in the number of fruits were statistically homogeneous across the three soil cover types evaluated.

When the number of fruit increments was analysed according to the type of microbial biofertilizer applied (Figure 7B), no significant differences were detected among treatments throughout the experimental period.

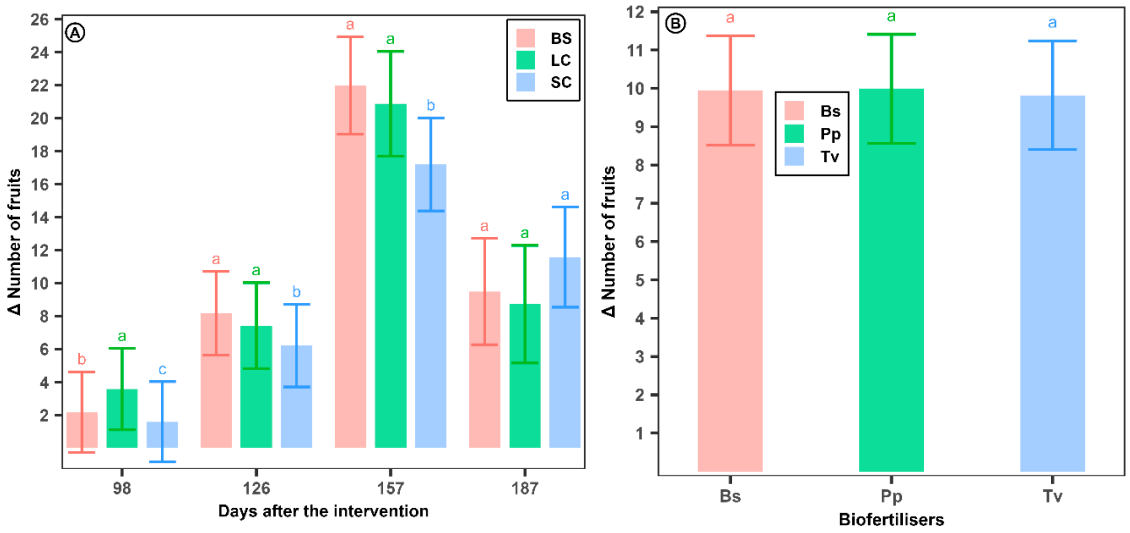

Figure 7. Increase in the number of fruits of *Carica papaya* L. across different evaluation periods and biofertilisers. BS: Bare soil (without cover); SC: Senescent cover; LC: Living cover; Bs: *Bacillus subtilis*; Pp: *Pseudomonas putida*; Tv: *Trichoderma viride*. Ⓐ Comparison of the number of fruit increments within each evaluation period; Ⓑ comparison of the number of fruit increments among the three microbial biofertiliser types. Different letters indicate significant differences at the 5% significance level using the FDR correction method.

Post hoc multiple comparisons following ANOVA revealed significant differences among specific treatments, even when interactions between living cover and microbial biofertilizer were not significant in the overall model (Table 4). At the final measurement, treatments with soil cover generally exhibited higher values for stem diameter, plant length, number of flowers, number of fruits, number of leaves, and yield compared with treatments without cover.

Within the soil-cover treatments, specific differences associated with the type of microbial biofertiliser were observed; however, these treatments clustered statistically, with comparable values among themselves and consistently higher than those of plots without cover. In contrast, treatments without cover exhibited the lowest values for most evaluated variables, whereas some treatments showed intermediate responses between the two groups (Table 4).

Table 4. Adjusted means of plant and fruit traits of *Carica papaya* L.

Factors	Stem diameter (cm)	Plant lenght (cm)	Number of flowers	Number of fruits	Number of leaves	Fruit weight (kg)
T01 (BS – Bs)	156.86d	195.00c	49.62c	36.76cd	38.03d	309.87c
T02 (BS – Pp)	170.85bc	198.52bc	54.34ab	37.66bcd	43.9ab	408.13abc
T03 (BS – Tv)	165.05cd	193.68c	53.42b	36.36d	38.93cd	336.93bc
T04 (LC - Bs)	171.12bc	204.4ab	54.69ab	46.23a	44.18ab	485.19a
T05 (LC – Pp)	171.12bc	209.35a	55.07ab	41.06abcd	42.03bc	413.56abc
T06 (LV – Tv)	182.23a	204.56ab	57.15ab	42.32ab	45.11ab	432.24ab
T07 (SC – Bs)	175.97ab	208.70a	54.77ab	42.07abc	45.77ab	379.84abc
T08 (SC – Pp)	172.23bc	204.36ab	56.07ab	41.68abc	44.93ab	413.16abc
T09 (SC – Tv)	172.1abc	212.03a	57.89a	43.81a	46.52a	444.88ab

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BS: Bare soil (without cover); LC: Living cover (*Canavalia ensiformis*); SC: Senescent cover (leaf litter); Bs: *Bacillus subtilis*; Pp: *Pseudomonas putida*; Tv: *Trichoderma viride*. Different letters indicate significant differences at the 5% significance level using the FDR correction method.

3.6. Interactions Between Living Cover and Microbial Biofertilizer

Simple effects analysis revealed differential responses of microbial biofertilisers across soil cover types (Figure 8). In plots with senescent cover, the three evaluated microbial biofertilisers showed similar responses across the agronomic variables analysed. In plots with living cover, specific differences among biofertilisers were observed, with greater stem diameter associated with treatments inoculated with *Trichoderma viride*. In the absence of cover, treatments inoculated with *Pseudomonas putida* were associated with higher values of stem diameter, number of flowers, and number of leaves, as well as greater fruit weight, although the latter did not reach statistical significance.

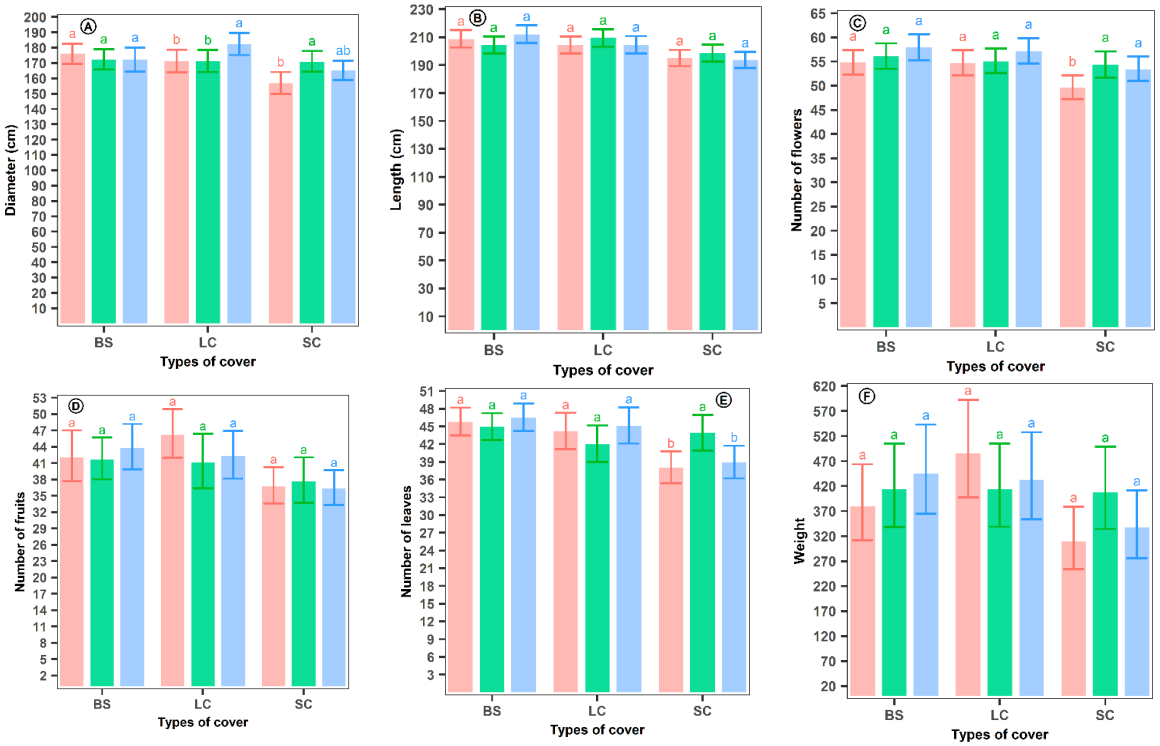

Figure 8. Estimated means of morphological and productive traits of papaya. Comparisons are based on simple effects, where different letters (a, b) indicate significant differences in morphological and productive traits among microbial biofertilisers within each soil cover type. Bar colours represent microbial biofertiliser types: red = *Bacillus subtilis*, green = *Pseudomonas putida*, and blue = *Trichoderma viride*.

4. Discussion

The results of this study demonstrate that soil cover management is the primary structuring factor influencing the response of *Carica papaya* L. under field conditions, consistently outweighing the isolated effects of microbial biofertilisers. Although the evaluated microbial biofertilisers exhibited specific effects, their efficacy was strongly dependent on the edaphic context created by soil cover, which explains the variability observed among treatments and evaluation periods. This finding is consistent with recent evidence indicating that the effectiveness of microbial biofertilisers in real agricultural systems is largely mediated by the physical, chemical, and biological conditions of the soil rather than by the intrinsic properties of the applied inoculants. In this context, the present study provides field-based experimental evidence supporting a hierarchical approach to

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agroecosystem management, in which soil habitat modification is a prerequisite for maximising the benefits of microbial biofertilisers in tropical fruit crops.

4.1. Soil Cover as a Structuring Factor of the Production System

The overall analysis showed that, among the fixed factors evaluated, microbial biofertilisation had a limited effect on most variables, whereas soil cover exerted a significant influence on most variables. Likewise, the interaction between these factors, evaluated both globally and across evaluation periods, significantly affected stem diameter and plant length, as well as increases in the number of leaves and flowers, except for the number of fruits. Collectively, these results indicate that soil cover is the primary regulator of the agronomic performance of *Carica papaya* L. under the conditions of the present study.

Soil cover may function as an edaphic regulator by reducing water losses through evaporation and maintaining higher moisture levels in the surface soil profile, thereby promoting vegetative crop development [33]. In contrast, biofertilisers play a modulatory role in the rhizosphere by enhancing nutrient bioavailability, inducing phytohormone synthesis, and strengthening plant–microorganism interactions [7]. However, their performance under field conditions is often variable, highlighting the need for standardised formulation and application protocols [34].

4.2. Temporal and Individual Variability: Interpretation of Random Effects

Regarding random effects, both plant identity and evaluation time significantly affected the number of flowers, whereas other variables were less influenced by these factors. This pattern suggests that the observed variability reflects, on the one hand, individual differences among plants and, on the other, environmental fluctuations along the growth cycle, including temperature, humidity, and nutrient availability. Kumar et al. [35] indicate that these factors can modulate phenotypic expression in papaya, although genetic variability remains a key determinant of fruit yield and quality.

4.3. Final Yield: Hierarchy of Effects Between Soil Cover and Biofertilisation

In the variables evaluated at the end of the production cycle (stem diameter, plant length, number of flowers, number of fruits, number of leaves, and fruit weight), soil cover was confirmed as the main factor explaining plant responses. Microbial biofertilisation applied in isolation had limited effects, primarily influencing stem diameter and leaf number, and produced only marginal effects on plant length and fruit weight when interacting with soil cover. These results are consistent with those reported by Gaat et al. [33], who observed that straw mulch significantly enhanced plant growth even under irrigated conditions, suggesting that soil cover improves water-use efficiency by reducing evaporation and preserving soil moisture. Similar effects have been documented in other agricultural systems [36,37].

4.4. Multivariate Validation: Structural Coherence of Treatments

Principal component analysis confirmed that both soil cover and microbial biofertilisers influenced the phenotypic characteristics of *Carica papaya* L.; however, the most consistent and robust separation was observed between plots with and without soil cover. Treatments without cover (T01–T03) were associated with lower growth and yield values, whereas treatments with living or senescent cover (T04–T09) clustered with higher values across the evaluated variables. These findings are consistent with those reported by Patel and Ramdevputra [36], who documented significant improvements in vegetative parameters under cover systems, even when combined with different fertilisation strategies. Taken together, the PCA results reinforce the hypothesis that soil cover constitutes the primary structural axis of the production system.

4.5. Temporal Windows of Response and Early Crop Establishment

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The temporal patterns observed for stem diameter and plant length indicate that treatment effects are not uniform across the phenological cycle but instead emerge during specific windows of heightened sensitivity, particularly in the early stages of cultivation. Soil cover–induced modification of the edaphic microhabitat enhances water retention and moderates soil temperature, thereby creating favourable conditions for root establishment and nutrient uptake [38,39]. During these initial stages, intensified root growth and cell division stimulate the release of root exudates, such as organic compounds, which function as chemical signals that promote microbial recruitment and facilitate nutrient mobilisation in the rhizosphere [40–43].

Under this scenario, the combination of soil cover and inoculation with *Trichoderma viride* promoted enhanced initial growth, particularly in stem diameter and plant length. This response is likely associated with phosphorus solubilisation mechanisms and the synthesis of phytohormones such as auxins [44,45]. Living cover extends beyond its physical role in thermal regulation; studies by Lombardi et al. [46] have demonstrated that root exudates actively attract *Trichoderma* spp. and serve as a readily available energy substrate. This continuous input of labile carbon favours fungal proliferation and increases its biological effectiveness. In contrast, senescent cover provides a more chemically complex carbon source, which limits its immediate availability and may constrain the short-term performance of the microbial biofertiliser.

4.6. Attenuation of Effects and Stabilisation of the Edaphic System

In later stages of development, differences among treatments tended to diminish, coinciding with the development of a deeper and more efficient root system that could explore larger soil volumes. This pattern suggests a progressive balance between the native soil microbiota and the inoculated microorganisms [47,48], a phenomenon widely documented in studies on biological soil management [49].

4.7. Context Dependency: Interpretation of Key Interactions

The central finding of this study is not the absence of a main effect of microbial biofertilisers, but rather the presence of significant interactions between soil cover type and microbial biofertilisation. These interactions demonstrate that the effectiveness of microbial biofertilisers is strongly context-dependent [13]. Under living cover, *Trichoderma viride* showed a significant advantage in stem growth, likely promoted by the continuous supply of root exudates from *Canavalia ensiformis*, which serve as readily available energy substrates supporting its metabolic activity [50,51]. In contrast, *Pseudomonas putida* exhibited a superior response in soils without cover, possibly due to its greater metabolic versatility and higher tolerance to abiotic stress [52]. In the absence of the physical and microclimatic protection provided by soil cover, plant performance may rely on stress-tolerant bacteria that enhance nutrient acquisition under suboptimal conditions [53]. Finally, under senescent cover, the abundance of structural carbon derived from leaf litter may have induced a microbial saturation effect, thereby attenuating functional differences among the microbial biofertilisers [54].

4.8. Agronomic Implications and Management Hierarchy

The results indicate that the implementation of cover crops is a priority and decisive strategy for maximising papaya yields. The application of microbial biofertilisers in the absence of a favourable soil environment is both inefficient and economically risky [55]. Once an adequate habitat has been established through soil cover, the selection of microbial biofertilisers can provide specific and incremental benefits. This approach redefines the sequence of agroecological management, positioning the soil as a primary environmental filter that conditions the effectiveness of biofertilisers by regulating microclimatic conditions and resource availability [56,57]. Accordingly, this hierarchy of effects should be considered the central axis for the development of technical recommendations and policy guidelines aimed at sustainable agriculture based on ecological processes.

4.9. Limitations

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Despite the statistical robustness of the experimental design, based on generalised linear mixed models (GLMM), several limitations should be acknowledged. First, the results are specific to sandy clay loam soils with slightly alkaline pH, and therefore may not be directly transferable to other edaphic contexts. Under different soil conditions, the competitive dynamics between microbial biofertilisers and native microbial communities may vary substantially. Second, the absence of direct measurements of soil microbial activity and root exudate composition required that the underlying biochemical mechanisms be inferred from existing literature rather than empirically verified. Future research should incorporate direct assessments of microbial community structure and function, including metagenomic or metatranscriptomic approaches, to determine whether the applied microbial biofertilisers establish persistent colonisation or merely induce transient stimulation effects.

5. Conclusions

This study demonstrates that soil cover is the dominant factor regulating the growth, production, and yield of *Carica papaya* L. under field conditions, widely surpassing the isolated effects of the applied microbial biofertilisers. Crop performance followed a distinct hierarchy of effects, in which the establishment of a favourable edaphic habitat through soil cover was crucial for enhancing the agronomic performance of the system.

Both living (*Canavalia ensiformis*) and senescent (leaf litter) soil coverings consistently increased stem diameter and plant length, as well as the number of leaves, flowers, and fruits, and overall weighted yield compared with bare soil. These effects were most pronounced during the early stages of cultivation, when the root system is highly sensitive to soil conditions and to the availability of water and nutrients.

Microbial biofertilisers did not exhibit a significant main effect on final yield; however, their effectiveness was conditional on both the type of soil cover and the stage of the crop cycle. Under living cover, *Trichoderma viride* promoted greater early vegetative growth, whereas *Pseudomonas putida* showed a comparatively better response in soils without cover. In contrast, under senescent cover, the three microbial inoculants evaluated displayed comparable performance.

These results provide empirical evidence of the context-dependent nature of biofertiliser efficacy, confirming that their performance is not an intrinsic property of the inoculant itself, but rather an emergent outcome of its interaction with the soil–plant–management system. Consequently, the key question for agronomic decision-making is not which biofertiliser is inherently superior, but under which soil and management conditions its potential can be effectively expressed.

From an applied perspective, these results indicate that the implementation of soil covers should be considered a priority and structural strategy in papaya production systems, whereas microbial biofertilisers use should be deployed as a complementary and context-specific tool. This hierarchical approach of agronomic management reduces production risk and enhances the efficiency and predictability of biologically based practices under field conditions.

Overall, this study provides both a conceptual and practical framework for the design of agroecological management strategies in papaya, highlighting the need to integrate soil conservation practices with the rational and context-dependent use of microbial biofertilisers to achieve more efficient, resilient, and sustainable production systems.

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