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Effects of commercial bioformulations on tomato seedlings growth promotion

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ABSTRACT

Tomato (*Solanum lycopersicum* L.) holds global importance due to its culinary versatility and nutritional value. This study evaluated the effect of different commercial bioformulations on the growth promotion of tomato seedlings. The formulations used were based on i) *Trichoderma asperellum* strain CCT 2165; ii) *Paecilomyces lilacinus*; iii) a Microbial consortium composed of *Lactobacillus* spp. and actinomycetes; iv) non inoculated plants. Seedlings were grown for 25 days in polystyrene trays with 128 cells, using Carolina Soil® substrate. Inoculation was performed prior to sowing, following the furrow application dosage recommended by the manufacturers. The following parameters were evaluated: emergence speed index (ESI), emergence rate, seedling height, collar diameter, fresh and dry mass of shoot and root, total dry mass, and Dickson Quality Index (DQI). The experiment was laid out in a randomized block design (RBD), comprising 4 treatments (inoculations) and 10 replicates per treatment. P. *lilacinus* and the microbial consortium yielded the highest EVI values, 22.84 and 23.02 emerged plants day^{-1}, and emergence rates of 100% and 98.75%, respectively. P. *lilacinus* provided the best seedling development and a DQI of 0.002. In the principal component analysis (PCA), which explained 78.64% of the total variation, the dominance of P. *lilacinus* across variables was evident. However, the dendrogram revealed a similarity in the effects caused by the microbial consortium and P. *lilacinus*. It is concluded that the use of P. *lilacinus* is the most recommended treatment; albeit the application of other microorganisms also improves seedling growth.

Keywords: Microbial inoculants, Actinomycetes, *Trichoderma asperellum*, *Paecilomyces lilacinus*, *Solanum lycopersicum.*

Efeito de bioformulados na promoção de crescimento de mudas de tomateiro

RESUMO

O tomate (*Solanum lycopersicum* L.) tem importância global, pela versatilidade culinária e valor nutricional. Objetivo desse trabalho foi avaliar o efeito de diferentes bioformulados na promoção de crescimento das mudas do tomateiro. Foram utilizados produtos formulados à base dos microrganismos i) *Trichoderma asperellum* cepa CCT 2165; ii) *Paecilomyces lilacinus*, iii) Consórcio microbiano com *Lactobacillus* e Actinomicetos; e iv) plantas sem aplicação. As mudas foram cultivadas durante 25 dias em bandejas de isopor compostas por 128 células com substrato Carolina *Soil*®. A inoculação foi realizada antes da semeadura na dose de aplicação de sulco recomendada pelos fabricantes. Foram avaliados, índice de velocidade de emergência (IVE), emergência, altura, diâmetro do colo, massa verde e seca da parte aérea e raiz, massa seca total e o índice de qualidade de Dickson (IQD). Foi utilizado delineamento de blocos ao acaso (DBC), cada bloco composto por 4 tratamentos e 10 repetições por tratamento. O P. *lilacinus* e o consórcio microbiano resultaram no melhor IVE 22,84 e 23,02 plantas emergidas dia^{-1} e emergência 100% e 98,75% respectivamente. O P. *lilacinus* proporcionou o melhor desenvolvimento e IQD 0,002 das mudas, na análise de componentes principais (PCA), na qual explica-se 78,64% da variação total, fica mais evidente a dominância do P. *lilacinus* nas variáveis, mas o dendrograma, mostra uma proximidade nos efeitos causados pelo consórcio microbiano ao P. *lilacinus*. Concluindo que a utilização de P. *lilacinus* é mais recomendada, embora a utilização dos demais microrganismos tenham efeito no crescimento das mudas.

Palavras-chave: Microrganismos, Actinomicetos, *Trichoderma asperellum*, *Paecilomyces lilacinus*, *Solanum lycopersicum*.

1. Introduction

The tomato (*Solanum lycopersicum* L.) is a cosmopolitan vegetable widely used for its culinary versatility and nutritional value (Furquim et al., 2023). High-quality seedlings account for about 60% of tomato plant production success, and the use of microorganisms in seedling production can help promote plant growth and development, ensuring quality and reducing the impacts of biotic and abiotic factors, resulting in less costly management (Krause et al., 2017; Carvalho et al., 2023).

The use of biological inputs to promote plant growth increases the resistance of cultivated plants, ensuring the sustainability of production systems due to their low cost and the socioeconomic benefits provided by this management approach, which is a viable alternative for producers (Glick, 2014; Yu et al., 2021).

Plant growth promoting bacteria and fungi have a range of mechanisms of action, stimulating growth, acting on induced systemic resistance (ISR) and induced systemic tolerance (IST), increasing the production of hormonal compounds (auxins, gibberellins, cytokinins), solubilizing nutrients, and producing siderophores (Ahemad and Kibret, 2014).

Fungal microorganisms can be used as biostimulants for growth. Fungi of the genus *Trichoderma* are widely known for their potential in biocontrol, however, some species, such as *Trichoderma asperellum*, have been recorded as producers of abscisic acid (ABA), auxins (IAA), and gibberellins (GA) and promote plant growth (Illescas et al., 2021). The nematophagous fungus *Purpureocillium lilacinus* (syn: *Paecilomyces lilacinus*) can produce indole-3-acetic acid (IAA), an auxin, in addition to solubilizing phosphorus (Baron et al., 2020).

Bacterial microorganisms of the genus *Bacillus* and the actinomycetes group are morphologically distinct Gram-positive prokaryotes capable of forming resistance structures that aid survival in extreme conditions. They are widely used in agriculture and agricultural research because they have the potential to produce metabolites that are homologous to plant hormones auxins and gibberellins, act in the production of exopolysaccharides and in the solubilization of nutrients such as phosphorus, promoting plant growth (Souza et al., 2020; Naamala et al., 2023).

Apprehending the many mechanisms of action of plant growth promoting microorganisms is foundational. Selecting an unsuitable microorganism can cause an antagonistic effect by inhibiting development, releasing growth-inhibiting metabolites, competing with the plant for substrate resources, and causing an excessive amount of microorganism inoculum in the area. It is recommended to select, identify, and quantify which microorganisms have efficacy on seedling production (Glick, 2014; Blanco et al., 2022).

Bioformulations could yield beneficial outcomes on plant production, yet the expression of their mechanisms of action is contingent on multiple factors, including the inoculum nature, formulation, operational parameters, environment and the recipient crop. In this sense, it is important to assess the specific conditions and effects of the products to be used (Ahemad, 2014; Yu et al., 2021; Naamala et al., 2023). Thus, the objective of this study was to evaluate the effect of bioformulations on promoting the growth of tomato seedlings.

2. Material and Methods

This study was conducted in the plant production area of the Universidade Estadual de Mato Grosso do Sul, Aquidauana campus (UEMS/UAQ), at latitude 20º 20’ S, longitude 55º 48’ W, and an average altitude of 178 m. The climate of the region, according to the Köppen classification, is type Aw (tropical with a dry season in winter and rains in summer), average annual rainfall is 1,200 mm, maximum temperature is 33 °C, and minimum temperature is 19.6 °C.

The bioformulations selected are manufacturers-recommended for use in tomato cultivation. The microorganisms were selected based on documented plant growth-promoting potential as reported in the scientific literature. There is documented potential of *Lactobacillus helveticus* EL2006H CFS (Naamala et al., 2023), Actinomycetes (Sousa et al., 2020) *Paecilomyces lilacinus* (Baron et al., 2020) and *Trichoderma asperellum* (Yu et al., 2021).

Bioformulation inoculations were carried out by preparing 200 mL of microbial suspension per treatment, using the manufacturer-recommended dose. Each suspension was obtained by diluting the commercial formulations (Table 1) in distilled water. Subsequently, 1 mL of the prepared suspension was directly sprayed onto each individual cell of the seedling tray. Non-inoculated control plants were treated with pure distilled water to ensure baseline comparison.

The experiment utilized the 'Gaucho' tomato genotype, characterized as an indeterminate, salad-type cultivar with a phenological cycle ranging from 100 to 110 days.

Table 1. List of commercial products and microorganisms used in the production of tomato seedlings.

Product	Species	CFU mL^{-1}*	Recommendation
Tríppel	*Trichoderma asperellum*	1,0 x 10^5	Biopesticide
Nemakill	*Paecilomyces lilacinus*	1,0 x 10^5	Biopesticide
Flex Roots	Microbial consortium (*Lactobacillus* spp. +actinomycetes)	**	Organic fertilizer

* Colony forming units. ** Concentration of microorganisms, not disclosed by the company.

Seeds were sown on August 6, 2024, in 128-cell polystyrene trays filled with Carolina Soil® commercial substrate. Each tray cell received two seeds, and thinning was performed seven days post-emergence to retain a single seedling per cell. Manual irrigation was carried out twice daily, in the morning and late afternoon. At 25 days after sowing (DAS), ten plants were randomly selected from the central area of each treatment plot, comprising 32 treated plants, for evaluation.

The emergence speed index (ESI) was estimated by counting the number of seedlings that emerged, based on the number of days after sowing. A daily record of the number of emerging plants was kept until 7 DAS, when the first plant stand was complete. Subsequentially, the ESI was determined using Equation 1. For accurate measurement, only seedlings with open cotyledons were considered emerged (Maguire, 1962). Where: N - number of seedlings emerged observed on the day of counting; and D - number of days after sowing on which the count was made.

$$ESI = ((N1/D1) + (N2/D2) + \ldots + (NnDn)7) \qquad (1)$$

Seedling emergence rate was estimated at 7 days after sowing (DAS), based on Equation 2 (Souza et al., 2021a). Where: E% - emergence (%); NE - number of emerged seedlings; and NCE Total - number of seeds placed to emerge.

$$E\% = (NE/NCE\ Total) \times 100 \qquad (2)$$

Tomato seedlings were considered suitable for transplanting upon reaching the stage of 5 to 6 fully developed compound leaves, typically occurring between 25 and 35 days after sowing (DAS). At this developmental stage, the root system is well-established, vegetative growth is physiologically balanced, and the plants exhibit increased resilience to environmental stressors. This is the ideal period for evaluating the development and quality parameters of the seedlings (Issa et al., 2021).

Seedling development was characterized by analyzing morphological traits, including height (H), collar diameter (CD), fresh mass of the shoot (FMS) and root (FMR), dry mass of the shoot (DMS) and root (DMR), and total dry mass (TDM). Seedlings quality was assessed using the Dickson Quality Index (DQI), calculated according to Equation 3 (Dickson et al., 1960).

$$DQI = (MST/((H/DM) + (MSA/MSR))) \qquad (3)$$

The experiment was conducted using a randomized block design (RBD), consisting of 5 blocks, 4 treatments, and 10 replicates per treatment. Data were subjected to the Shapiro–Wilk and O'Neill–Mathews tests, confirming normality and homoscedasticity at a 5% significance level. Data for green root mass and dry root mass were subjected to square root transformation ($\sqrt{x}$) to meet assumptions of normality and homoscedasticity. Analysis of variance (ANOVA) was performed, and treatment means were compared using Tukey's test at a 5% significance level, implemented through the '*ExpeDes.pt*' package in R (Ferreira et al., 2023).

Principal components analysis (PCA) was applied to a matrix composed of mean values for height, collar diameter, fresh and dry mass of shoot and root, total dry mass, and the Dickson Quality Index (DQI), across five treatments resulting from the combination of tomato genotypes and inoculation conditions. The biplot was generated using the '*fviz_pca_biplot*' function from the '*Factoextra'* package in R (Kassambara, 2023). The dendrogram was generated using the Unweighted Pair Group Method with Arithmetic Mean (UPGMA), based on a matrix of Euclidean dissimilarity distances derived from growth response variables across treatments. All statistical analyses were performed using R software, version 4.3.3. (R Core Team, Vienna, Austria).

3. Results and Discussion

Treatments involving microbial bioformulations led to significant improvements ($p < 0.05$) in the emergence speed index (ESI) and seedling emergence rate, as presented in Table 2. The most promising treatments for seedling development were *Paecilomyces lilacinus*, which achieved an emergence speed index (ESI) of 22.84 plants day^{-1} and 100% emergence, and the microbial consortium composed of *Lactobacillus* spp. and actinomycetes, which yielded an ESI of 23.02 plants day^{-1} and 98.75% emergence. In comparison, *Trichoderma asperellum* resulted in an ESI of 15.77 emerged plants days^{-1} and emergence of 88.10%, while the control treatment (without microbial application) showed an ESI of 14.70 plants day^{-1} and 82.40% emergence.

Table 2. Effect of the application of microorganisms used as growth promoters on the emergence speed index (ESI) and emergence of tomato seedlings.

Treatments	ESI (emerged plants day^{-1})	Emergence (%)
Microbial consortium	23.02 a	98.75 a
P. lilacinus	22.84 a	100.00 a
T. asperellum	15.77 b	88.10 b
Non inoculated	14.70 b	82.40 c
F Treatments	**58.88 ($p < 0.001$)**	**51.35 ($p < 0.001$)**
F. Blocks	**3.94 ($p < 0.029$)**	1.01 ($p = 0.440$)
CV (%)	6.82	2.87

The bioformulations associated with the highest ESI and total emergence were the microbial consortium and *P. lilacinus*, both of which promoted uniform emergence and accelerated initial development of tomato seedlings (Table 2). The results confirm that bioformulations contribute to improved seedling development; however, the effectiveness varies depending on the microorganism employed. Each microbial species elicits distinct physiological responses in the plant, influenced by the nature of the compounds they release and their specific interactions with plant metabolism.

Treatment with the microbial consortium resulted in uniform emergence and accelerated growth of tomato seedlings. These outcomes align with those reported by Naamala et al. (2023), who reported similar effects in soybean cultivation using the strain *Lactobacillus helveticus* EL2006H CFS. Similarly, Khan et al. (2024) documented improvements in the early development of wheat (*Triticum aestivum* L.) following the application of *Lactobacillus agilis*, L. *acidophilus*, and *L. plantarum*.

Experiments involving actinomycetes in wheat, conducted by Sousa et al. (2020), also resulted in uniform emergence and accelerated initial growth of seedlings. Bacterial microorganisms have shown the potential to stimulate rapid and homogeneous crop development and can be applied either individually or in combination with other beneficial microbes.

The responses observed following the inoculation with *P. lilacinus* in this experiment are consistent with those reported by Baron et al. (2020), who applied the fungus as a bioinoculant in soybean (*Glycine max* (L) Merrill), corn (*Zea mays* L.), and bean (*Phaseolus vulgaris* L.) crops, and found that it promoted uniformity in the initial growth of the plants.

The fungus *P. lilacinus* along with some bacterial strains of the genus *Lactobacillus* and actinomycetes, releases secondary metabolites and phytohormones–such as indoleacetic acid (IAA), a key auxin, into the growth environment. These bioactive compounds contribute to uniform and accelerated seedling germination and emergence (Giassi et al., 2016; Baron et al., 2020; Naamala et al., 2023).

Application of *T. asperellum* resulted in an ESI of 15.77 and an emergence rate of 88.10%, both lower when compared to the other microbial treatments and comparable to the control without inoculation (Table 2). These results align with those documented by Barbosa et al. (2018), who, when applying a strain of *T. aureoviride* in association with bovine manure, did not detect uniform or accelerated initial growth of lettuce (*Lactuca sativa* L.) seedlings, results comparable to both the control and the treatment with *Pseudomonas putida*. Santos et al. (2020), evaluating two *Trichoderma* isolates, found that the *Trichoderma* sp. – Tc isolate reduced emergence speed and did not promote uniform seedling development, while the *T. asperellum* TAM03 isolate did lead to homogeneous and rapid initial growth.

The effect caused by *Trichoderma* may be associated with two factors. The first is the specific interaction between the plant and the fungal isolate. This was shown by Souza et al. (2021b), who evaluated three lettuce (*Lactuca sativa* L.) cultivars treated with Quality® and Organic® formulations, both based on the *T. asperellum* strain URM5911. Despite sharing the same inoculum source, Quality® increased germination time by 3.17 days–delaying seedling emergence–while Organic® reduced germination time by 1 day, accelerating emergence, both compared to the control. The second factor may be related to the amount of inoculum used. Rokni et al. (2021) reported that increasing the concentration of *Serendipita indica* during the inoculation of bell pepper (*Capsicum annuum* L.) seeds led to a reduction in germination–86.1% at the highest concentration and 69.8% at the lowest. Additionally, seedling emergence decreased by 6.8% at the highest concentration and 5.5% at the lowest, showing that excessive inoculum levels can negatively affect early seedling development.

The bioformulations used in the treatments had significant effects ($p < 0.05$) on all components of tomato seedling development, as presented in Table 3. The commercial products also significantly influenced the Dickson Quality Index ($p < 0.05$), reinforcing their impact on overall seedling quality.

All tested bioformulations promoted improvements in seedling morphological traits of seedling development and the Dickson Quality Index (DQI) compared to the control treatment. Both the microbial consortium and *T. asperellum* enhanced seedling development compared to the untreated group, as shown in Table 3. Nonetheless, the microbial treatment using *P. lilacinus* outperformed the other microbial formulations in promoting seedling development Table 3.

The results found in this study are consistent with those reported by Nesha and Siddiqui (2017), who applied *P. lilacinus* alone or in combination with *Aspergillus niger*, resulting in increased carrot (*Daucus carota* L.) growth–13.30 cm in length, 11.00 g in fresh weight, and 6.67 g in dry weight. Similarly, Baron et al. (2020) noted that different strains of *P. lilacinus* enhanced plant height in corn and soybean, along with gains in root dry mass and total biomass in soybean. Under water stress conditions, Musa et al. (2023) confirmed that *P. lilacinus* promoted tomato root development (3.16 cm), increased fresh weight (1.4 g), and improved dry weight (0.05 g), confirming its growth-promoting potential even under abiotic stress. A comparable effect on plant growth was documented in the present experiment.

The results obtained from the treatment with the microbial consortium are in agreement with those reported by Khan et al. (2024), who used three strains of *Lactobacillus* and found that *L. gilis* and *L. plantarum* promoted increases in plant height, stem diameter, root length, and both fresh and dry mass in wheat. Similarly, Sousa et al. (2020) confirmed improvements in the same growth parameters in wheat seedlings using two products formulated with actinomycetes.

Naamala et al. (2023), applying *L. helveticus* EL2006H to three different crops under saline stress conditions, also reported enhanced root development in potatoes, corn, and soybeans. These authors attribute the observed improvements to the ability of *Paecilomyces lilacinus*, *Lactobacillus* spp., and actinomycetes to stimulate the production of indole-3-acetic acid (IAA). In addition, these microorganisms may produce secondary metabolites that mimic plant growth hormones, promoting cell division and elongation, and thereby enhancing overall seedling development.

The results related to seedling development indicate that, although *Trichoderma asperellum* is marketed primarily as a biofungicide, it can also contribute to improved seedling quality. This effect was evident even in the presence of delayed germination and emergence, suggesting a mode of action comparable to that of the microbial consortium.

Trichoderma species can function as either growth biostimulants or antagonists, depending on the specific strain and environmental context. Variability occurs not only between species but also among individual isolates, with their effects modulated by the plant species involved. As noted by Yu et al. (2021), different *Trichoderma* species elicit distinct responses in seed germination and plant growth, suggesting that metabolites produced by individual isolates may stimulate development in certain plant species while remaining ineffective in others. Expanding on this, Santos et al. (2021) demonstrated that inoculum concentration is directly proportional to the degree of plant growth stimulation, underscoring the importance of dosage in maximizing beneficial outcomes.

When *Trichoderma harzianum* was used to inoculate tomato plants under drought conditions, Mona et al. (2017) reported enhanced growth and stress resistance, correlating with increased levels of secondary metabolites, proline, and key phytohormones such as indoleacetic acid (IAA), indole butyric acid (IBA), and gibberellic acid (GA).

The results of this study, along with supporting evidence from literature, show that the genus *Trichoderma* has potential as a growth biostimulant in specific environmental contexts. However, its effectiveness is highly variable, depending on the species, strain, and host plant involved. Certain isolates can produce phytohormones, which contribute to enhanced root development, cell elongation, and overall plant vigor. Yet, when inoculum concentrations are excessive, the overproduction of these compounds may lead to inhibitory effects, particularly during germination and seedling emergence. This duality highlights the importance of selecting appropriate strains and optimizing application rates to harness the biostimulant potential of *Trichoderma* without compromising early plant development.

Tomato seedlings produced using *P. lilacinus*, the microbial consortium, and *T. asperellum* are all recommended for field transplanting due to their higher DQI, which can enhance seedling survival after transplantation. This observation is supported by the findings of Santos and Scalon (2020) and Lima et al. (2017), who, in evaluating the DQI of vegetable seedlings, affirmed that the higher the index, greater quality, vigor, robustness, and biomass of the seedling – traits that increase the likelihood of survival following transplanting in the field.

The DQI results following treatments with *P. lilacinus*, the microbial consortium, and *T. asperellum* were all below 0.02, yet higher than those observed in the control treatment. It is important to highlight that DQI threshold values vary across species. Silva et al. (2018) evaluated bell pepper seedlings produced in different substrates and reported DQI values greater than 0.02. In contrast, Freitas et al. (2019), when producing lettuce seedlings with varying doses of organomineral fertilizer, observed DQI values ranging from 0.0047 to 0.0160, noting that seedlings with higher DQI exhibited superior quality.

Table 3. Results of development components and quality index in tomato seedlings after using microorganisms on growth promotion.

Morphological traits of seedling development and Dickson Quality Index				
Microorganisms	**H (cm)**	**CD (cm)**	**FMS (g)**	**FMR (g)**
Microbial consortium	6.82 ab	0.21 b	0.46 b	0.45 c
P. lilacinus	7.17 a	0.23 a	0.56 a	0.63 a
T. asperellum	6.70 b	0.21 b	0.48 b	0.52 b
Without application	6.72 b	0.20 b	0.39 c	0.32 d
Fcal. Treatments	**4.31 (p< 0.001)**	**10,24 (p< 0.001)**	**21.54 (p< 0.001)**	**60.65 (p< 0.001)**
Fcal. Blocks	**9.05 (p< 0.001)**	2 (p< 0.113)	**13.30 (p<0.001)**	**13.92 (p< 0.001)**
CV (%)	10.83	12.16	22.18	23.65
Microorganisms	**DMS (g)**	**DMR (g)**	**TDM (g)**	**DQI**
Microbial consortium	0.033 ab	0.13 b	0.053 b	0.0015 b
P. lilacinus	0.036 a	0.15 a	0.060 a	0.0020 a
T. asperellum	0.030 b	0.13 b	0.049 b	0.0015 b
Without application	0.025 c	0.11 c	0.038 c	0.0011 c
F_{cal}. Treatments	**25.21 (p< 0.001)**	**18.35 (p< 0.001)**	**30.63 (p< 0.001)**	**37.13 (p< 0.001)**
F_{cal}. Blocks	**2.96 (p< 0.001)**	**6.53 (p< 0.001)**	**7.28 (p< 0.001)**	**1.52 (p< 0.001)**
CV (%)	21.39	18.82	22.79	24.30

Averages followed by the same letter do not differ statistically from each other according to the Tukey test at 5%. Where: H - Height; CD - Collar diameter; FMS – Fresh mass of shoot; FMR – Fresh mass of root; DMS - Dry mass of shoot; DRM - Dry mass of root; TDM - Total dry mass; and DQI - Dickson Quality Index. Values in bold are significant at the 5% F-test.

These findings reinforce that while DQI values are species-specific, the index remains a reliable parameter for assessing seedling growth, quality, and survival potential. Principal component analysis (PCA) revealed that PC1 and PC2 accounted for 68.56% and 11.56% of the total variance, respectively, explaining 80.12% of the variation in seedling development. The variables analyzed included height, collar diameter, fresh and dry mass of shoot and root systems, total dry mass, DQI, emergence, and emergence speed index (ESI), across treatments with *P. lilacinus*, the microbial consortium, *T. asperellum*, and non-inoculated plants, as shown in Figure 1.

The first two principal components accounted for 80.13% of the total variance, supporting the use of two-dimensional dispersion for interpreting treatment effects.

According to Seymen et al. (2019), graphical interpretation is considered valid when the first two components explain at least 70% of the accumulated variation, ensuring that most of the data structure is captured in the biplot. In this study, treatments with *P. lilacinus*, the microbial consortium, and *T. asperellum* showed distinct separation from the control, driven primarily by variables such as DQI, dry mass, and emergence.

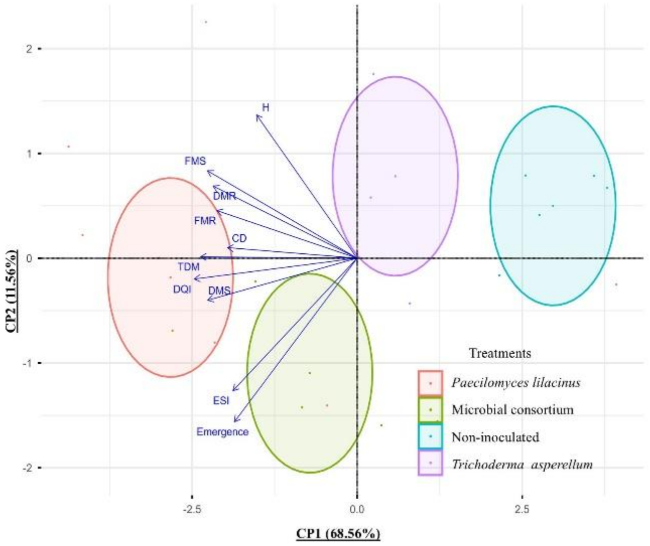

Figure 1. Two-dimensional dispersion of microbial treatments based on principal component scores derived from ten biometric variables. Components 1 and 2 explain 80.12% of the total variance.

Analysis of the biplot reveals that PC2 (11.56%) is more strongly associated with seedling development components and the Dickson Quality Index (DQI), particularly in treatments with *P. lilacinus* and the microbial consortium. In contrast, PC1 (68.56%) captures greater variation in treatments with *T. asperellum* and the non-inoculated control. The spatial distribution of treatments in the PCA indicates that the overall responses in seedling development and quality are predominantly influenced by *P. lilacinus*. Notably, the variables emergence and emergence speed index (ESI) are more closely aligned with the microbial consortium, suggesting its dominant effect on early seedling performance (Figure 1). Data collected following microbial treatments with *P. lilacinus* and the microbial consortium demonstrates that their secondary metabolites enhanced seedling quality and stimulated growth. These treatments represent viable, low-cost alternatives for vegetable growers in tomato seedling production.

As shown in Figure 2, distinct groupings emerged among treatments, suggesting consistent patterns in their effects. Moreover, the PCA analysis (Figure 1) revealed direct relationships between specific biometric responses and the microorganisms applied, with *P. lilacinus* and the microbial consortium showing strong influence on key variables. These findings were further supported by the dendrogram in Figure 2, which confirmed the clustering behavior observed in the PCA.

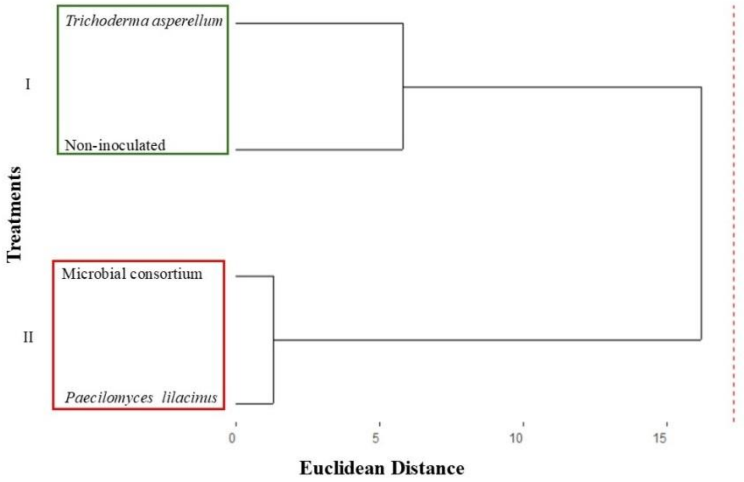

Figure 2. Dendrogram showing hierarchical clustering of microbial treatments based on Euclidean distances, illustrating group formation among seedling growth promoters.

A dissimilarity matrix was computed using Euclidean distance among treatments, followed by hierarchical agglomerative clustering via the UPGMA (Unweighted Pair Group Method with Arithmetic Mean) algorithm. The resulting dendrogram (Figure 2) reveals two distinct clusters, indicating that the treatments exert significantly different multivariate effects on seedling development. One cluster includes *P. lilacinus* and the microbial consortium, while the other comprises *T. asperellum* and the non-inoculated control. This separation reflects divergent influences of the microbial treatments on biometric traits. Within each cluster, the relatively short linkage distances suggest high internal similarity, underscoring consistent patterns in how each microorganism modulates seedling growth and quality.

The clade comprising *P. lilacinus* and the microbial consortium exhibits high intracluster similarity, suggesting analogous effects on seedling growth responses. In contrast, the second clade, formed by *T. asperellum* and the non-inoculated control, reflects greater intercluster dissimilarity relative to the other microbial treatments. Notably, the linkage height within this second cluster indicates a degree of intracluster heterogeneity, highlighting differential effects between *T. asperellum* and the control. These hierarchical relationships, as depicted in the dendrogram, corroborate the multivariate patterns observed in PCA, reinforcing the influence of microbial treatments on the morphological traits of seedling development.

4. Conclusions

All evaluated bioformulations demonstrated potential as plant growth promoters in tomato seedlings, enhancing key developmental parameters relative to the non-inoculated control. Accordingly, all treatments contributed to seedling growth promotion. However, the bioformulation based on *Paecilomyces lilacinus* exhibited the most pronounced effects, indicating superior efficacy and greater suitability for use in tomato seedling production.

Authors' Contribution

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