



Enhancing Red-Chili (*Capsicum annuum* L.) Growth Using Black Soldier Fly (*Hermetia illucens*) Probiotics as Carrier

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Abstract

Red chili (*Capsicum annuum* L.) is a crucial horticultural commodity in Indonesia, necessitating expansion to meet the growing food demands. The formulation of carriers with probiotics is a promising solution for improving red chili growth. This study examines how the combination of carriers containing bacteria and *Trichoderma* from the intestines of *Hermetia illucens* larvae affects the growth of red chili cultivar Lembang-1. The research method used a Randomized Block Design (RBD) with six treatments. The treatment consisted of a group with the addition of a kaolin carrier with bacteria and *Trichoderma* (BTrK), a talc carrier with bacteria and *Trichoderma* (BTrT), a zeolite carrier with bacteria and *Trichoderma* (BTrZ), and a control group without a carrier (KtC), a positive control (K(+)), and a negative control (K(-)). The Least Significant Difference (LSD) test showed that the addition of carriers affected increasing plant height and the number of branches compared to the negative control. Specifically, BTrK demonstrated the highest number of leaves and branches. The result concluded that adding carriers with probiotics from the intestine isolate of BSF larvae contributes to the improved growth of red chili. The implementation of carrier addition with probiotics is considered to be a sustainable strategy in agriculture.

Keywords: Carrier, growth, *Hermetia illucens* larvae, probiotics, red chili

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Introduction

Red chili (*Capsicum annuum* L.) is a globally important horticultural commodity and is extensively cultivated in tropical countries, including Indonesia. Over the past five years, Indonesia has faced challenges related to declining chili availability. According to Center Agricultural Data and Information (2023), the availability of chili dropped drastically from 4.60 Kg/capita/year in 2019 to 1.23 Kg/capita/year in 2023, reflecting a stark contrast with the high demand for chili. Red chili, prized for its extended shelf life, holds particular importance in various processed foods in Indonesia (Wijaya *et al.*, 2020). Issues contributing to red chili production failures include attacks by plant-disrupting organisms that hinder optimal growth (Mellinia *et al.*,

2025). Applying beneficial microorganisms as biological agents represents a promising solution to foster environmentally friendly agricultural practices (Agriopoulou *et al.*, 2023).

The intestines of Black Soldier Fly or BSF (*Hermetia illucens*) larvae contain beneficial bacteria such as *Enterobacter* (Zhineng *et al.*, 2021). Utilizing bacteria and *Trichoderma* from the intestine isolate of BSF larvae has been shown to enhance tomato growth (Sanjaya *et al.*, 2023). However, relying solely on probiotics may not sufficiently maintain their viability to the target (Sun *et al.*, 2023). Incorporating a carrier into the formulation is a promising strategy to stabilize the inoculant during distribution and extend the shelf life. Carrier selection criteria include physical and chemical stability, non-toxicity to inoculants, biodegradability, absence of

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pollutants, good buffering capacity, and high moisture resistance (Dutta *et al.*, 2023).

Previous studies have highlighted that combining carriers with beneficial microorganisms can effectively enhance plant growth. The application of *T. afroharzianum* and *Azotobacter chroococcum* with a carrier in the form of Carboxymethyl cellulose resulted in increased growth of basil plants (Comite *et al.*, 2021). Similarly, the combining zeolite with *Arbuscular mycorrhiza* promoted enhanced growth of soybean plants (Sarmiento-López *et al.*, 2025). These carriers act as a nutrient source that supports the probiotic's survival for facilitating maximum activity.

This study focuses on combining carriers with probiotics as a solution to maximize inoculant activity and maintain the material's shelf life. The choice of carrier also considers the ease of obtaining it (Bolan *et al.*, 2023). Thus, kaolin, talc, and zeolite carriers were used in combination with bacteria and *Trichoderma* from the intestine isolate of *H. illucens* larvae to analyze how they affect the vegetative growth of red chili.

Materials and Methods

Study area

This research was carried out from December to April 2024 at the Botanical Garden, Universitas Pendidikan Indonesia in Sukasari District, Bandung City at an altitude of 923 asl with coordinates 6°51'44.2"S 107°35'41.8"E.

Sample preparation

The research design used a Randomized Block Design (RBD) consisting of 6 treatments with 4 replicates. The treatment is a combination of biological agents with the addition of carriers including Bacteria, *Trichoderma*, with Kaolin carrier (BTrK); Bacteria, *Trichoderma*, with Talc carrier (BTrT); Bacteria, *Trichoderma*, with Zeolite carrier (BTrZ); non-carrier control (KtC) or Bacteria, *Trichoderma* only; positive control with fungicides, and insecticides (K (+)); and negative control without treatment (K (-)). This research used red chili cultivar Lembang-1 as a population. In this study, the materials were acquired from The Vegetable Plant Research Institute Lembang, Indonesia.

The growth parameters assessed in this study include the plant height (cm), number of leaves per plant, and number of primary branches per plant. Climatic factors such as light intensity, temperature, and air humidity were also monitored. Additionally, edaphic factors such as soil moisture (Siregar *et al.*, 2024). Rainfall data were obtained from the online database of The National Geophysical Meteorology and Climatology Agency (BMKG). Data collection occurred weekly starting from 10 weeks after the red chili plants were planted.

The study commenced with seeding, planting, and maintenance. Red chili were planted in polybags measuring 40 x 40 cm. After 9 weeks of planting, red chili seedlings were prepared for treatment. The combination treatment of probiotics with carriers for each plant was carried out with a dose of 20 grams per plant (Sutarman & Prahasti, 2022). Treatment without carriers received a dose of 10 mL each of bacteria and *Trichoderma* (Kristi *et al.*, 2024). Applications were conducted around the roots of red chili every two weeks. Additionally, an insecticide with a fungicide is a positive control that adheres to labeled dosage guidelines. These applications were similarly scheduled every two weeks but stopped if the plants were healthy to avoid becoming resistant.

Data analysis

Data of the growth parameters were analyzed using IBM SPSS Statistic 25 software to interpret the data. Prerequisite tests were carried out, and the data of this study were normally distributed so that the analysis of mean differences between treatments was carried out using the Least Significant Difference (LSD) test with a significance level of 5%.

Results and Discussion

Plant growth is a critical phase that determines the survival and productivity of a crop. Various parameters are employed to assess plant growth conditions, including plant height, number of leaves, and number of dichotomous branches. Based on these parameters, observations began in the first week after treatment application, corresponding to the fifth week after transplanting (10 weeks after

sowing, WAS). Measurements of plant height and number of leaves were conducted weekly from 5 to 14 WAS and observations of the number of branches were carried out from 9 to 14 WAS.

The plant height data were sequentially ranked across treatments K (+), BTrK, KtC, BTrT, BTrZ, and K (-). The LSD test revealed that the average heights of red chili in the BTrK, BTrT, KtC, and K (+) groups were significantly different from those in the K (-) group. However, the BTrZ group showed a difference in red chili plant height compared to the K (-)

treatment, but the results were not significantly different.

The new findings on the number of leaves showed that the treatment with the addition of carrier had an average number of leaves that were noticeably different from the negative control red chili group. The best number of leaves was successively carried out by the positive control group, BTrK, KtC, BTrT, and BTrZ. The lowest number of leaves is shown by the negative control group. The LSD showed that there is no significant difference between treatments.

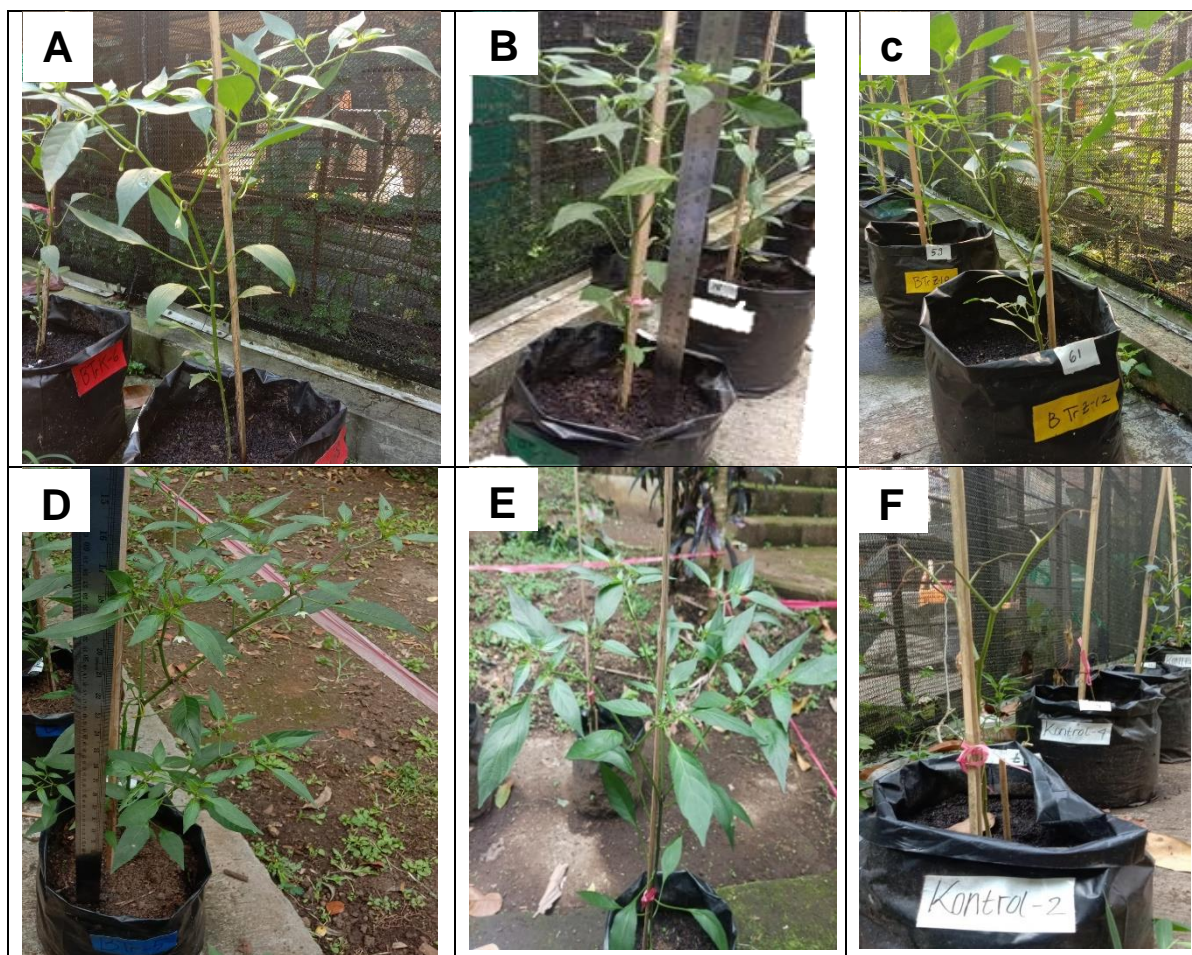


Figure 1. Growth red chilis at 19 weeks : A) BTrK; B) BTrT; C) BTrZ; D) KtC E) K (+); F) K (-).

Table 1. The plant height

| Treatments | The plant height per repeat (cm) | | | | Average |
|------------|----------------------------------|----|----|----|---------------------------|
| | 1 | 2 | 3 | 4 | |
| BTrK | 35 | 23 | 31 | 26 | 28.75±2.658 ^a |
| BTrT | 31 | 20 | 33 | 20 | 26.00±3.488 ^a |
| BTrZ | 29 | 20 | 22 | 22 | 23.25±1.974 ^{ab} |
| KtC | 27 | 32 | 33 | 21 | 28.25±2.750 ^a |
| K (+) | 35 | 33 | 19 | 40 | 31.75±4.498 ^a |
| K (-) | 11 | 12 | 15 | 13 | 12.75±0.854 ^b |

Table 2. The average number of leaves

| Treatments | The number of leaves per repeat | | | | Average |
|------------|---------------------------------|----|----|-----|---------------------------|
| | 1 | 2 | 3 | 4 | |
| BTrK | 58 | 31 | 65 | 65 | 54.75±8.087 ^a |
| BTrT | 50 | 24 | 74 | 31 | 44.75±11.191 ^a |
| BTrZ | 46 | 37 | 33 | 44 | 40.00±3.028 ^a |
| KtC | 39 | 76 | 66 | 24 | 51.25±11.982 ^a |
| K(+) | 40 | 49 | 26 | 108 | 55.75±18.048 ^a |
| K(-) | 7 | 10 | 12 | 15 | 11.00±1.683 ^a |

Table 3. The average number of dichotomy branches

| Treatments | The number of leaves per repeat | | | | Average |
|------------|---------------------------------|----|----|----|---------------------------|
| | 1 | 2 | 3 | 4 | |
| BTrK | 30 | 16 | 28 | 23 | 24.25±3.119 ^a |
| BTrT | 23 | 11 | 36 | 15 | 21.25±5.513 ^{ab} |
| BTrZ | 24 | 15 | 16 | 20 | 18.75±2.056 ^{ab} |
| KtC | 19 | 33 | 34 | 11 | 24.25±5.588 ^a |
| K(+) | 22 | 25 | 12 | 51 | 27.50±8.312 ^a |
| K(-) | 1 | 1 | 2 | 4 | 2.00±0.707 ^b |

This study proved that the addition of kaolin carrier with bacteria and *Trichoderma* carried out better red chili growth than KtC and was able to compensate for K (+). Kaolin has high absorbance, therefore it is easier to use as a nutrient storage place (Chen *et al.*, 2023). This causes the interaction of inoculants with the roots of red chili plants to occur optimally, which is shown by better red chili growth when compared to the negative control. In this study, kaolin is applied in powder form, thus the formulation is easily decomposed.

The structural properties of kaolin enable to absorb nutrients and assist in the delivery of inoculants to the root zone of red chili plants. Kaolin can increase nutrients such as Calcium (Ca), and Boron (B) in red chili (Salim *et al.*, 2019). Calcium is a mineral that plants need in large quantities to support growth. Boron is a micronutrient that is also important for plant survival. Calcium and Boron

play an important role in the occurrence of biochemical processes and plant physiology (Bhatla & Lal, 2023). Increasing calcium and boron stimulates plant height, the number of branches, and the number of red chili's (Salim *et al.*, 2019). This shows the effect of kaolin carrier on plant growth which is in line with the findings in this study.

BTrT treatment also showed better growth compared to K (-). The characteristics of talc which have a wide particle size allow probiotics to multiply and be able to maintain their viability (Basheer *et al.*, 2019). Powder form facilitates the decomposition of the formulation, by releasing bacterial inoculants and *Trichoderma* to interact with the roots of the red chili. Talc carriers of *T. viridae* with *Bacillus* showed the synergistic effect of the two inoculants in producing siderophores, and antifungal compounds to suppress pathogens so that the root growth of peanut plants is not

disturbed (Harman *et al.*, 2021). *Bacillus* and *Enterobacter* are involved in transporting silicon minerals in plants (Raza *et al.*, 2023). Silicon minerals play a role in water drought, heavy metal toxicity, and resistance to pathogens, so they can help increase plant growth (Cassel *et al.*, 2023). Silicon has been reported to increase plant growth, number of leaves, stem diameter, and fruit yield of red chili. Silicon is a nutrient that is also important in plant growth (Lob *et al.*, 2023).

The growth of red chili in the BTrZ treatment exhibited a relatively lower average compared to the BTrK and BTrT treatments but showed better results than the K(-) control group. The high cation exchange capacity (CEC) of zeolite enables it to absorb and release essential nutrients required by plants, such as potassium (K), calcium (Ca), and magnesium (Mg). This property allows zeolite to efficiently supply nutrients to chili plants, resulting in improved growth. Moreover, the use of zeolite in fertilization enhances the nutrient retention capacity in the soil, thereby improving nutrient availability (Cataldo *et al.*, 2021). This increased efficiency plays a crucial role in promoting the optimum growth of chili plants. In this study, zeolite as a carrier for inoculants sourced from the intestines of BSF larvae did not reach its maximum potential. This difference from the characteristics of zeolite applied in gravel form contrast with other carriers typically used in powdered form. The gravel form results in larger carrier particles, leading to a slower release of the inoculant over time. Zeolite is a carrier for biological agents in liquid form, which effectively enhances tomato growth (Prisa, 2020).

KtC red chili treated with bacteria and *Trichoderma* in liquid form exhibited favorable growth results following BTrK. The efficacy of this treatment can be attributed to the easy absorption of bacteria and *Trichoderma* when applied in liquid form (Sutarman & Prahasti, 2022). This underscores the positive impact of probiotics from BSF larvae on red chili growth. During the study, both the K (+) replicates and

the KtC were severely affected by mites, leading to a significant reduction in the average growth of these red chilies. Mites typically feed on young leaves, extracting plant juices and causing buds, flowers, and fruits to drop prematurely (Kleruk *et al.*, 2024). Interestingly, despite mite attacks, the treatment group with carriers did not experience losses of shoots. This resilience could be linked to the effectiveness of the inoculant combined with carriers in bolstering the plant's defenses against pests.

The positive growth outcomes observed in BTrK, BTrT, BTrZ, and KtC treatments highlight a synergistic interaction between *T. viride* and *T. harzianum* as plant growth-promoting fungi (PGPF), alongside *Bacillus subtilis*, *Enterobacter*, and *Micrococcus* as plant growth-promoting rhizobacteria (PGPR) in the root rhizosphere of red chili. This synergy involves competition for nutrients with pathogens and the production of metabolites that inhibit pathogenic activity (Poveda & Eugui, 2022). *Trichoderma* species employ micro-parasitism to compete directly with pathogens and stimulate the production of phytohormones in plant roots (Dutta *et al.*, 2023). Additionally, PGPR bacteria suppress pathogenic microorganisms by producing metabolites such as antibiotic compounds (Poveda & Eugui, 2022). Consequently, red chili are shielded from pathogen attacks, resulting in improved red chili growth parameters compared to the negative controls.

The growth of red chili without treatment (K (-)) is notably poor. This is due to the essential nutrients required for red chili growth and development (Bhatla & Lal, 2023). The results from this negative control treatment serve as a relevant comparison to show how the addition of carriers containing inoculants from *H. illucens* larvae effectively enhances chili growth. This underscores the necessity of adequate nutrients for supporting optimal chili growth. These essential nutrients, red chili in the untreated group lacked the necessary conditions for their growth processes to occur optimally.

Table 4. Environment condition

| | Temperature (°C) | Humidity (%) | RH (%) | Soil Moisture (%) | Rainfall (mm) |
|---------|---------------------|-----------------|-----------|----------------------|------------------|
| Average | 4180 | 28.1 | 68.4 | 61 | 32.1 |

Optimal environmental conditions are crucial for accurately evaluating the effects of different carrier types. The significance of growth enhancements in shallots under optimal conditions when using carriers with *Trichoderma*, resulting in notable differences between carrier types (Sutarman & Prahasti, 2022). In contrast, the environmental conditions in our study were suboptimal for red chili growth. The average light intensity at the study site was 3596 lux, whereas red chili typically require light intensities ranging from 8000 lux to 15000 lux for optimal growth (Minanda & Idris, 2022). Additionally, there was notably high rainfall of 32.1 mm (BMKG, 2024). During the study period, the average air temperature was 28°C with air humidity at 68%. Soil moisture averaged 61%, which is generally favorable for red chili growth (Kleruk *et al.*, 2024).

The implementation of probiotics combined with carriers is considered an innovative approach to improving plant growth while being environmentally friendly. The combination of probiotics and carriers plays a crucial role in improving soil health, increasing nutrient availability for plants, and reducing reliance on chemical fertilizers and pesticides that may harm ecosystems.

Conclusion

This study presents new findings regarding the effectiveness of kaolin, talc, and zeolite carriers in delivering probiotics from the intestine of BSF larvae to improve the vegetative growth of red chili. Formulation with the addition of carriers provides protection therefore the growth of red chili can increase. The results suggest that these formulations could be instrumental in sustainable agricultural practices aimed at improving the growth of plants for crop productivity.

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