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Study on PGPR-Amended Biochar Application on Chili (*Capsicum annuum*) Seedling Growth

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Abstract
The sustainable approach to agricultural productivity needs finding alternative resources and processes to increase soil fertility and crop productivity limiting use of harmful chemical fertilizers and pesticides. Use of biochar as soil amendment may provide a significant support by water and nutrient retention as well as site for microbial growth and activity effectively enhancing soil fertility. Further, plant growth promoting bacteria may augment the plant growth by phytohormone release, nutrient recycling and anti-phytopathogenic activity. In the present study, biochar was inoculated with a consortium of plant growth promoting rhizobacteria, previously isolated from rhizosphere soil of Mimosa pudica. Experimentally, the effect on germination and growth of Capsicum annuum (chili) plant with PGPR-inoculated biochar (5% w/w biochar to potting soil) was compared to non-inoculated biochar (5% w/w) and non-amended soil control under laboratory conditions for 30 days. The rate of transplanted Capsicum annuum seed germination was found to be 100 % (20 seeds per pot/treatment x triplicate) within 5day to 10day range while true leaves developed within 10day to 12days. PGPR-inoculated biochar showed significant improvement in plant growth parameters compared to the control, such as plant height (11%), leaf number (12%), root length (12%) and total fresh biomass (14%). These study results indicate that biochar inoculated with PGPR may offer a sustainable alternative to synthetic fertilizers can to enhance seedling growth, nutrient availability, and soil fertility leading to improved soil health and agricultural productivity.

Keywords: *Biochar, Capsicum annuum, PGPR, Soil fertility, Sustainable agriculture*

Introduction

The overuse of synthetic chemical fertilizers and over the years has gradually shifted soil quality soil compaction, nutrient imbalance, acidification, soil runoff and decline in soil microbial diversity leading to poor crop productivity. Hence development and adoption of sustainable alternatives are need of the hour to restore soil health (Lehmann et al., 2015). The microbial flora and organic components for soil amendments are suggested to mitigate harmful effects of chemical fertilizers as well as suggested as alternative cost-effective environment friendly initiatives (Zou et al., 2024). Biochar has been found to significantly enhances soil physical properties such as texture and soil particles aggregation, nutrient retention and promoting soil microbes (Nepal et al., 2023). Biochar is a carbon rich, porous, organic material derived through pyrolysis of biomass obtained under low-oxygen or oxygen-limited conditions (Lehmann and Joseph, 2009). Rice husk is mostly composed of cellulose and lignin, hemicellulose and silica (Ugheoke and Mamat, 2012). It is an easily available low-cost agriculture biomass which is obtained after the dehusking or milling process to obtain rice grains. Rice husk has been used to make biochar through pyrolysis. The resulting rice husk biochar was found to have high porosity and large specific surface area. (Dagarova, 2025). The rice husk biochar was found to significantly enhance the structure of the soil, improve water & nutrient retention (Dagarova, 2025). It was also implied that biochar acts as an active component that increases the biological activity of the soil (Joseph et al., 2010).

Plant Growth-Promoting Rhizobacteria (PGPR) are a heterogenous group of bacteria inhabiting the rhizosphere which is the region of the soil directly in contact with plant roots and affected by plant roots-microbe interactions (Mohanty et al., 2021). This region is also a high activity zone for microorganisms. These microorganisms can influence plant growth via a range of both direct and indirect mechanisms. Among the direct mechanisms are the synthesis of phytohormones, such as auxins (Indole Acetic Acid, IAA, etc) and gibberellins, which play roles in regulating root formation and plant aerial parts, respectively (Kang et al., 2014, Mohanty et al., 2021). Specific rhizosphere microorganisms have ability to do nitrogen fixation and nitrogenous compound metabolization. Some of the microorganisms are found to solubilize phosphorus, commonly found in an insoluble state within the soil (Izomor et al., 2025).

Some PGPR organisms have been shown to produce anti-microbial components to compete with other pathogenic microorganisms (Idowu and Babalola, 2021). The diverse functions of PGPR renders them as fundamental components of sustainable agricultural practices (Vejan et al., 2016; Basu et al., 2021). Some published literature have shown to have efficacy in chili crops in Indian agricultural studies (Parveen et al., 2024; Bora et al., 2024). Existing literature indicates that microbial consortia may provide additive or synergistic advantages resulting in to produce a overall positive growth of the plant (Santoyo et al., 2021). Thus, instead of single organism, a consortia would provide a more effective growth environment for plant as biofertilizer (Welmillage et al., 2021). In view of above, the present study was designed to study the cumulative impact of biochar inoculated with a PGPR consortium, on the growth response of chili (*Capsicum annuum*) seedling under laboratory settings.

Materials and Methods

1. Biochar Production

In the present experiment, biochar was made from rice husks obtained from nearby agricultural field and subjected to the process of pyrolysis at 500°C (Anbuganesan et al., 2022). Pyrolysis at low temperatures is still recognized as an age-old yet effective method to obtain a product that possesses attractive properties to fortify the soils (Kuo et al., 2023). It has been indicated as effective to activate the metabolism as well as the positive traits of some bacterial strains, including the production of antibiotics (Anbuganesan et al., 2022). It suggests that biochar offers an active environment that promotes microbial growth, extending the role beyond that of simply providing a physical habitat (Glaser et al., 2002). To execute the experimental treatments, the biochar was fortified with a PGPR consortium to create the PGPR-amended biochar product.

2. PGPR Consortium Isolation and Biochar Amendment

The *Mimosa pudica* rhizosphere soil was used for isolation of plant growth promoting rhizobacteria as described in Sánchez-Cruz et al., 2019. soil suspension (10g/100ml sterile DW) was stirred, diluted and plated on nutrient agar to isolate rhizosphere bacteria. The bacterial isolates based on morphological and biochemical properties were purified by streak plate method. The 68 different isolates were tested for gibberellin and Indole Acetic Acid production and phosphate solubilizing property. A consortium of 7 isolates was grown as co-culture and used in the present study. (Kang et al., 2014, Idowu and Babalola, 2021, Santoyo et al., 2021. Sterile biochar was inoculated with co-culture for overnight and decanted before use. The consortium inoculated biochar was mixed with sterile potting mixture in desired proportion for experiment.

a. Experimental Design

To study the cumulative effect PGPR-inoculated biochar on chili plant growth, three sets of treatment was designed viz. control (potting mix alone), non-amended Biochar (non-inoculated Biochar plus potting mix, 5% w/w), Enriched Biochar (PGPR-inoculated biochar with potting mix, 5% w/w) in triplicates. A summary of the experimental strategy as well as parameters is provided in Table 1.

Table 1. Experimental design and parameters

SN	Treatment Group	Biochar used (w/w)	PGPR	Parameters
1	Control	0%	No	Plant height, leaf number, root length , total biomass
2	Non-amended Biochar	5%	No	
3	Enriched Biochar	5%	Yes	

b. Chili Seedling Cultivation and Growing Conditions

The seeds of Chili (*Capsicum annuum*) were sowed in the standard potting medium under laboratory controlled environment maintained at temperature range of 25–30°C and humidity range of 60–70% with a light/dark cycle of 12 hr during 30 days (Debbarma et al., 2018, Yuniati et al., 2023) adequate time to observe the effect of the treatments during the initial growth and development of the seedling (Lorio and De Asis, 2021).

c. Growth Parameter Assessment and Statistical Analysis

At the end of the 30-day duration, important growth parameters were recorded to determine the impact of the modified biochar. These were plant height, number of leaves, length of roots, and cumulative biomass (fresh weight). Measuring these parameters followed standard horticultural research practice, thereby generating a believable and comparable dataset (Yuniati et al., 2023).

Results

1. Effect of the Use of Biochar on Seed Germination

The analysis revealed that chili seeds from all the treatment groups germinated within the expected time frame of 5–10 days, during which the appearance of true leaves was observed between days 10 and 20 (Yuniati et al., 2023). It is observed that the germination was slightly early in biochar inoculated with PGPR. Such initial growth push gives the plant the opportunity to establish a stronger root structure as well as absorb the nutrients more effectively, creating a strong foundation upon which the notable improvement in growth recorded later during the experiment (Lorio and De Asis, 2021).

2. Effect on Morphological Growth Parameters

Chili seedlings that were treated with PGPR-inoculated biochar presented significant improvement of vital morphological traits compared to the control. Amendment of the biochar increased the plant height by 11%, where the treated plants had their height ranging between 9–16 cm compared to the control specimens (8–14 cm). The number of true leaves increased by 12% in PGPR-inoculated biochar (4-6 leaves) compared to 4–5 leaves in the controls. Furthermore, the root length was found to be increased by 12% (9–13 cm) compared controls (8–12 cm). These results support earlier studies for using PGPR in biochar application in cabbage (Zou et al., 2024) and wheat (Ashry and Hassan, 2019; Parveen et al., 2024).

3. **Effect on Plant Biomass**

The present experimental data shows the total biomass increase in the weight of treated chili seedlings by 14%, with an average weight range 4.8 to 6.5 g, as compared to control group (4 to 5 g). This biomass increase may be attributed to enhancement in plant growth parameters parameters such as height, root length, number of leaves, etc. demonstrating the overall positive effect of the PGPR-inoculated biochar amendment in soil. (Çıg et al., 2021; Bora et al., 2024). The results are summarized in the following Table 2.

Table 2: Effects of PGPR-Inoculated Biochar on Chili Seedling Growth

SN	Growth Parameter	Control	Non-inoculated Biochar	PGPR-inoculated Biochar	Percent Increase
1	Plant Height	8–14 cm	8-13 cm	9–16 cm	11%
2	Leaf Number	4–5 leaves	4-5 leaves	4–6 leaves	12%
3	Root Length	8–12 cm	8-13 cm	9–13 cm	12%
4	Total Biomass	4–5 g FW	4-5.2 g FW	4.8–6.5 g FW	14%

Discussion

The significant positive effects determined during this study can be attributed to the physicochemical properties of biochar as well as the biological functions of PGPR bacteria. The highly porous structure of the biochar provides a safe-guarded micro-habitat for the PGPR, acting as a haven against stress factors from the environment, such as desiccation, temperature fluctuations, as well as competition among indigenous soil microbes (Zou et al., 2024). Such immobilization of PGPR on the biochar surface modifies their viability and survival after they are introduced into the potting mixture, facilitating their effective colonization among the roots of chili plants as well as the delivery of their desirable activities (Tao et al., 2023; Gryta et al., 2024). Biochar has also been explored as a vehicle among PGPR bioinoculant carriers (Basu et al., 2021). Moreover, biochar, as a dynamic micro-environment, could potentially activate microbial processes. It has been determined that biochar could potentially increase both the diversity as well as the quantity among desirable microbes, enriching significant taxa among the microbial community within the soils (Zou et al., 2024; Wu et al., 2025). Specific constituents among biochar, including manganese as well as silicon dioxide, were found to directly enhance the metabolism as well as the desirable activity among bacteria (Anbuganesan et al., 2022). Synergistic use of biochar as well as plant growth-promoting rhizobacteria (PGPR) will enhance microbial activity of rhizospheres to build a useful micro-environment (Tao et al., 2023; Welmillage et al., 2021). This contributes to soil properties useful to facilitate varied microbial communities (Hardy et al., 2019).

This study reveals biological activities of PGPR bacteria with biochar's characteristics. Plant health parameters elevation reveals an indication of PGPR's biological role. Mimosa pudica literature reveals their capacity to produce auxins (e.g., IAA) that facilitate root development (Idowu and Babalola, 2021; Mohanty et al., 2021). Growth-promotion by a consortium of PGPR (gibberellin and IAA- positive strains) indicate and promote cell extension and vertical development (Mohanty et al., 2021; Kang et al., 2014). Additionally, phosphate-solubilizing bacteria in this consortium promote phosphorus availability that is important for root health and transmission of energies (Izomor et al., 2025).

Root development can be significantly improved through Plant Growth-Promoting Rhizobacteria (PGPR) activity with phytohormones and mobilization of nutrients to improve root development (Vejan et al., 2016). Increased biomass summation indicates higher physiological activity. PGPR enhance nutrient availability through nitrogen fixing and phosphate solubilization (Mohanty et al., 2021). Biochar acts as a "nutrient reservoir" to sequester desired nutrients and prevent leaching from the rhizosphere (Lehmann et al., 2011). This symbiosis allows sustained availability of nutrients to stimulate expanded plant development (Haider et al., 2023). Experiment results and literature searches confirm that biochar in combination with PGPR is an effective agronomic input to provide a sustainable biofertilizer to stimulate an increment of crop yield.

Conclusion

The present study explores the cumulative effect of PGPR and rice husk biochar on the growth of chili (*Capsicum annuum*) seedlings. The present study comprised of studying effect of PGPR-inoculated biochar on plant growth stimulation and vigor. The present study findings suggest that the increased plant height (11%), leaf number (12%), root length (12%), and overall biomass (14%), shows a positive cumulative effect of PGPR-inoculated biochar on crop health. It also supports the interaction of PGPR with biochar as neutral support substrate for PGPR attachment and growth as well as creating necessary microenvironment for plant growth. The study parameters result suggest that biochar has conditioning effect on the soil by supporting beneficial microbial growth supporting soil fertility. Thus, biochar along with plant growth promoting rhizobacteria can be a sustainable alternative to chemical fertilizers. However, there remains a need to conduct more research on different types of PGPR and ratios of ratios of biochar-PGPR for better applications strategies on various crops to optimize their utilization.

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Conflicts of interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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