PLANT GROWTH-PROMOTING RHIZOBACTERIA: A PROMISING STRATEGY TO OPTIMIZE THE DEVELOPMENT OF SCHIZOLOBIUM PARAHYBA

Mercedes Carranza-Patiño 1
Yussely Laz-Vera 2
Robinson J. Herrera-Feijoo 3
Edwin Jiménez-Romero 4
Ángel Cedeño-Moreira 5
Cristhian Chicaiza-Ortiz 6

ABSTRACT

Objective: To investigate the efficacy of plant growth-promoting rhizobacteria (PGPR) in enhancing the early development stages of Schizolobium parahyba, a critical species for the timber industry, aiming to optimize reforestation and agroforestry practices for sustainable forestry.

Theoretical Framework: The research is grounded in the exploration of symbiotic relationships between PGPR strains and plant species, focusing on their potential to improve seed germination, plant growth, and biomass production. The study examines how these interactions can be leveraged to enhance agricultural productivity and sustainability, with a specific emphasis on the benefits of employing PGPR in forestry.

Method: The study utilized an experimental approach, assessing the impact of four PGPR strains (Serratia marcescens, Pseudomonas protegens, Enterobacter absuriae, and Acinetobacter calcoaceticus) on S. parahyba seedlings. These were cultivated in three different substrates, with variables such as germination rate, above-ground and root growth, and biomass production meticulously measured to evaluate the effects of bacterial inoculation.

Results and Conclusions: The application of PGPR notably improved all measured growth parameters in S. parahyba seedlings, with A. calcoaceticus and P. protegens specifically enhancing stem and root development, respectively. A synergistic effect was observed in substrates containing peat, underscoring the significant potential of PGPR in boosting the productivity and sustainability of early-stage forestry cultivation. The findings advocate for further field studies to fine-tune these interactions, aiming to establish more resilient and ecologically sustainable agricultural practices.

Originality/Value: This research contributes novel insights into the utilization of PGPR in forestry, highlighting its substantial promise in enhancing the early growth stages of Schizolobium parahyba. By demonstrating the specific benefits of PGPR strains and their interactions with different substrates, the study offers a promising strategy for improving the sustainability and productivity of reforestation and agroforestry efforts.

Keywords: Microrganismos Plant Growth Promoters, Rhizobacteria, Plant Biostimulants, Initial Growth.

1 Universidad Técnica Estatal de Quevedo, Quevedo 120550, Ecuador. E-mail: mcarranza@uteq.edu.ec
Orcid: https://orcid.org/0000-0002-0917-0415
2 Universidad Técnica Estatal de Quevedo, Quevedo 120550, Ecuador. E-mail: juselly.laz2018@uteq.edu.ec
Orcid: https://orcid.org/0000-0003-3205-2350
3 Universidad Técnica Estatal de Quevedo, Quevedo 120550, Ecuador. E-mail: rherreraf2@uteq.edu.ec
Orcid: https://orcid.org/0000-0002-7411-8189
4 Universidad Técnica Estatal de Quevedo, Quevedo 120550, Ecuador. E-mail: ejimenez@uteq.edu.ec
Orcid: https://orcid.org/0000-0003-6564-5569
5 Universidad Técnica Estatal de Quevedo, Quevedo 120550, Ecuador. E-mail: acedenom@uteq.edu.ec
Orcid: https://orcid.org/0000-0003-3970-4550
6 Low Carbon College, Shanghai Jiao Tong University, Shanghai, China. E-mail: cristhianchicaiza@hotmail.com
Orcid: https://orcid.org/0000-0003-6564-5569
RHIZOBACTERIAS PROMOTORAS DEL CRECIMIENTO VEGETAL: UNA ESTRATEGIA PROMETEDORA PARA OPTIMIZAR EL DESARROLLO DE SCHIZOLOBIUM PARAHYBA

RESUMEN

Objetivo: Investigar la eficacia de las bacterias promotoras del crecimiento vegetal (PGPR) para optimizar las etapas de desarrollo temprano de Schizolobium parahyba, una especie crucial para la industria maderera, con el fin de optimizar las prácticas de reforestación y agroforestación para una silvicultura sostenible.

Marco Teórico: La investigación se fundamenta en la exploración de relaciones simbióticas entre cepas de PGPR y especies vegetales, enfocándose en su potencial para mejorar la germinación de semillas, el crecimiento de plantas y la producción de biomasa. El estudio examina cómo estas interacciones pueden ser aprovechadas para mejorar la productividad y sostenibilidad agrícola, con un énfasis específico en los beneficios de emplear PGPR en la silvicultura.

Método: El estudio utilizó un enfoque experimental, evaluando el impacto de cuatro cepas de PGPR (Serratia marcescens, Pseudomonas protegens, Enterobacter absuriae y Acinetobacter calcoaceticus) en plántulas de S. parahyba. Estas fueron cultivadas en tres sustratos diferentes, con variables como la tasa de germinación, el crecimiento aéreo y de raíces, y la producción de biomasa meticulosamente medidas para evaluar los efectos de la inoculación bacteriana.

Resultados y Conclusiones: La aplicación de PGPR mejoró notablemente todos los parámetros de crecimiento medidos en las plántulas de S. parahyba, con A. calcoaceticus y P. protegens mejorando específicamente el desarrollo del tallo y de la raíz, respectivamente. Se observó un efecto sinérgico en sustratos que contienen turba.
Plant growth-promoting rhizobacteria (PGPR) are emerging at the forefront of agricultural and forestry research. PGPR represents a revolution in the sustainable management of ecosystems (Moreno Reséndez et al., 2018). By establishing a symbiosis with plants, rhizobacteria perform crucial functions that enhance plant health and growth, resulting in ecological and economic benefits (Valery Ramirez & Reyes, 2013). The importance of PGPR has been magnified in the context of current environmental challenges. PGPR underlines its relevance in improving nutrient uptake and pathogen control (Acurio Vásconez et al., 2020). By their nature, rhizobacteria are a valuable resource for scientific research and innovation in agricultural practices (Cerrato & Alarcón, 2001). Rhizobacteria continue to attract the scientific community's attention due to their potential to improve agricultural and forestry production.

The use of PGPR in agriculture has gained recognition for its efficacy compared to traditional chemical fertilizers, showing a positive impact on soil health and crop yields (Moreno et al., 2018). Recent research findings suggest that rhizobacteria can substantially enhance germination rates and overall plant growth, particularly in adverse environmental conditions. This advantage is of utmost importance, especially in the climate change phenomenon (Santoyo-De la Cruz et al., 2023). The PGPR inoculation has demonstrated the ability to enhance nutrient assimilation and bolster resistance against diseases, which are crucial determinants of plant well-being (León et al., 2015). PGPR possesses the capacity to effectively modify the rhizosphere effectively, hence stimulating the production of plant hormones and enhancing soil quality. In the field of forestry, the inoculation of PGPR is seen as a potentially practical approach to improve the vigor for tree seedlings, which is crucial for the successful
implementation of reforestation initiatives and the attainment of sustainable timber output (Cely et al., 2016; Rueda-Puente et al., 2011).

Rhizobacteria, a collective of advantageous microorganisms, are prevalent in forest soils and significantly influence the overall well-being of forest ecosystems (Velasco-Jiménez et al., 2020). Rhizobacteria can colonize plant roots and form symbiosis with them. This symbiosis enables them to engage in nitrogen fixation and nutrient solubilization, facilitating plant development and enhancing their chances of survival (Gouda et al., 2018). Rhizobacteria can colonize plant roots and develop a symbiotic relationship with them. This symbiosis enables them to engage in nitrogen fixation and nutrient solubilization, facilitating plant development and enhancing their chances of survival (Angulo et al., 2014). Plant growth-boosting bacteria have emerged as a promising and efficient substitute for traditional fertilizers and insecticides (Silva et al., 2022). The utilization of advantageous microorganisms has the potential to facilitate the broadening of the spectrum of agriculturally viable soils, as well as find practical applications in the domains of forestry and species preservation (Ponce, 2021).

Despite notable progress, there remains a lack of knowledge regarding the impact of PGPR on tropical tree species, including Schizolobium parahyba (Gómez-Luna et al., 2012). The limited scope of knowledge constrains comprehending the optimal strategies for maximizing the advantages of PGPR in these crucial organisms. The limited scope of expertise denies understanding the optimal strategies for maximizing the benefits of PGPR in these essential organisms. A comprehensive comprehension of these variables is crucial for the successful execution of efficient management strategies in the fields of reforestation and agroforestry (de Andrade et al., 2023).

This study aims to explore how inoculating Schizolobium parahyba with selected strains of Plant Growth-Promoting Rhizobacteria (PGPR) influences the plant’s morphology and early growth stages. It specifically targets enhancements in plant height, stem diameter, biomass, and root development. The study also seeks to determine the varying effects of different substrates on these growth factors. The findings are expected to provide valuable insights for future research and practical applications in reforestation and sustainable agroforestry cultivation.
2 MATERIALS AND METHODS

2.1 STUDY AREA

The analysis was conducted at the Microbiology Laboratory of the State Technical University of Quevedo, Ecuador, located at coordinates 1°3'18 "S, 79°25'24 "W and at an elevation of 77.60 m.a.s.l. The environment had an average temperature of 24.83 °C, relative humidity of 86.83%, an annual accumulated rainfall of 1988.2 mm, 898.66 hours of sunshine per year, and a flat topography.

2.2 EXPERIMENTAL DESIGN

The study used a completely randomized design, employing a 5 x 3 factorial arrangement. Each treatment was replicated three times, resulting in 45 observations. Two primary factors were taken into account in this study. The first factor, Factor A, involved the inoculation of different rhizobacteria, including a control group treated with distilled water. The specific rhizobacteria used were *S. marcescens*, *P. protegens*, *E. absuriae*, and *A. calcoaceticus*. The second factor, Factor B, focused on the substrates utilized. These substrates were composed of combinations of mountain soil, perlite, and peat, with proportions of 2:1:1. The methodology was selected to investigate the collective impacts of rhizobacteria comprehensively and substrates on the factors of seedling growth.

2.3 STATISTICAL ANALYSIS

A descriptive analysis was conducted throughout the period from planting to the completion of germination, with a specific emphasis on evaluating markers such as germination percentage, duration, and the physical and microbiological attributes of the seedlings. To examine the influence of microbial conditions and substrate types on these metrics, a statistical analysis was conducted using a two-way analysis of variance (ANOVA) and Tukey's test (p≤0.05). The software utilized for this analysis was Statistica v10.0.
2.4 VARIABLE MEASUREMENTS

The germination percentage and the days required for germination were documented (Caroca et al., 2016). Seedlings were measured for height, stem diameter with a digital Vernier caliper, number of leaves, leaf weight, and root volume (Gupta et al., 2017). The bacterial density present in the substrate was assessed through serial dilutions and sowing on King B medium (King et al., 1954).

2.5 EXPERIMENTAL MANAGEMENT AND GROWING CONDITIONS

2.5.1 Plant Material

The seeds of *Schizolobium parahyba* were subjected to pre-germination techniques and subsequently infected with rhizobacteria (1×10⁸ CFU/mL). Seedlings were grown for 60 days under controlled conditions (25°C, 86.83% RH, 16/8 hours light/dark) (INAMHI, 2023).

2.5.2 Bacterial Inoculum Preparation

The selected bacterial strains were injected into King B liquid media using sterile procedures, following the protocol outlined by Rodríguez (2018). The strains, stored at -80 °C, were reactivated on agar plates and incubated at 26 °C for 24 hours. Subsequently, the specimens were relocated to Erlenmeyer flasks with a volume of 50ml of the specified medium. The flasks were then placed on an orbital shaker operating at a speed of 150 RPM and a temperature of 26 °C for 24 hours (King et al., 1954). Cell density was quantified using a spectrophotometer at a wavelength of 600 nm, adjusting the concentration to 1x10⁸ cells/ml with a sterile medium.

2.5.3 Pregermination Process of *S. parahyba*

The seeds underwent manual scarification to disrupt dormancy, employing the method outlined by Ponce (2021) The specimens were then introduced into aseptic Petri plates with damp filter paper and incubated at 23 °C with a 12-hour photoperiod for eight days. Germinated seeds were identified by root emergence.
2.5.4 Sterilization and Substrate Preparation

The substrates used in this study consisted of a blend of mountain soil, peat, and perlite. Solarization was employed as a method of sterilization to ensure sterility. It involved covering the substrates with a black polyethylene sheet and exposing them to direct sunshine for eight days (Česonienė et al., 2023). Sterilization efficacy was verified by inoculating samples into culture media and observing the absence of microbial growth following a 72-hour incubation period.

2.5.5 Seeding and Seedling Management

The experiment involved planting pre-germinated seeds in seedlings containing sterilized substrate under a controlled environment of a clean room. After 25 days, the seedlings were subsequently transplanted into separate bags containing fresh substrate to minimize stress and maximize the survival rate.

2.5.6 Inoculation with Rhizobacteria and Continuous Care

An inoculum was created by diluting a sterile diluent to achieve a final concentration of $1 \times 10^8$ (CFU/ml). Each seedling was administered 5 ml of the solution, which was immediately applied to the soil and leaves using a sterile atomizer. The care regimen encompassed a tri-weekly irrigation schedule, the prevention of water saturation, and a subsequent inoculation after 15 days to facilitate root colonization.

2.5.7 Microbial Community Assessment

In order to conduct microbiological testing, solid King B culture medium was prepared by spreading the substance into sterile Petri dishes and allowing it to solidify. 1g soil samples were collected and diluted in a series of steps. The diluted samples were placed on the plates and kept in a controlled environment at a temperature of 28°C. Afterward, the colonies were enumerated, the number of CFUs determined, and biochemical assays conducted to identify the prevailing microbes.
3 RESULTS

Inoculating seedlings with PGPR significantly impacted the developmental indexes in *S. parahyba* seedlings. The statistical study revealed the substantial impact of PGPR and substrates on seed germination, vegetative growth, and root development. The subsequent outcomes report the main experimental findings.

3.1 DESCRIPTION OF THE SYMPTOMS OF THE DISEASE

3.1.1 Effect of Plant Growth-Promoting Rhizobacteria on *S. Parahyba* Seed Germination

The germination rate of *S. parahyba* seeds was enhanced by inoculating them with various strains of PGPR, as evidenced by Figure 1, compared to the control group that was not infected. The *A. calcoaceticus* strain exhibited the highest germination rate (95%), which was statistically significant (p<0.05). Additional PGPR, including *S. marcescens, P. protegens*, and *E. absuriae*, also enhanced germination rates by 77.5-87.5 % compared to the control group (70%) (p<0.05).

![Figure 1](image-url)

*Effect of PGPR inoculation on *S. parahyba* seed germination*

Note: Percentage germination of *S. parahyba* seeds 15 days after inoculation with five strains of plant growth-promoting rhizobacteria. The percentage shown in the bars is the average obtained from a frequency analysis. No
PGPR control inoculated with distilled water (Control), *S. marcescens* (PM3-8), *P. protegens* (CHAO), *E. absuriae* (PM3-14), *A. calcoaceticus* (PM2-12).

The germination kinetics were expedited by the inoculation with PGPR compared to the control (Figure 2). *P. protegens* and *A. calcoaceticus* achieved germination after only 7 days, but the control group took 12 days.

**Figure 2**

*Effect of PGPR inoculation on germination kinetics of *S. parahyba**

![Germination Start and End by Treatment](image)

*Note:* Days elapsed from inoculation to onset and end of *S. parahyba* seed germination after being treated with various strains of plant PGPR at a concentration of 1X 10^8 CFU, compared to seeds that were not treated. The labels include: Control (No PGPR control injected with distilled water), *S. marcescens* (PM3-8), *P. protegens* (CHAO), *E. absuriae* (PM3-14), and *A. calcoaceticus* (PM2-12).

3.1.2 Influence of PGPR and Substrates on Vegetative Growth Parameters

Basic effects analysis demonstrated distinct impacts of the PGPR and substrates on the growth of *S. parahyba* (Table 1). *A. calcoaceticus* had a substantial positive effect on stem diameter, increasing it to 4.19 mm (p<0.05), and on wet weight, raising it to 3.63 g (p<0.01). The combination of mountain soil, peat, and perlite led to a significantly higher average height of 30.05 cm (p<0.05) and dry weight of 0.96 g (p<0.01).
Table 1.

Individual effects of PGPR and substrates on growth of S. parahyba

<table>
<thead>
<tr>
<th>Treatment effects</th>
<th>Height (cm)</th>
<th>Diameter (mm)</th>
<th>Number of leaves</th>
<th>Wet weight (gr)</th>
<th>Dry weight (gr)</th>
<th>Root volume (cm³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1</td>
<td>Control</td>
<td>30.19 a</td>
<td>3.75 b</td>
<td>15.00 a</td>
<td>2.87 bc</td>
<td>0.81 b</td>
</tr>
<tr>
<td>A2</td>
<td>S. marcescens</td>
<td>28.66 ab</td>
<td>3.71 b</td>
<td>15.00 a</td>
<td>2.62 bc</td>
<td>0.83 b</td>
</tr>
<tr>
<td>A3</td>
<td>P. protegens</td>
<td>28.42 ab</td>
<td>3.80 b</td>
<td>14.00 a</td>
<td>2.94 b</td>
<td>0.90 ab</td>
</tr>
<tr>
<td>A4</td>
<td>E. absuriae</td>
<td>27.46 b</td>
<td>3.52 b</td>
<td>14.00 a</td>
<td>2.43 c</td>
<td>0.82 b</td>
</tr>
<tr>
<td>A5</td>
<td>A. calcoaceticus</td>
<td>27.83 b</td>
<td>4.19 a</td>
<td>16.00 a</td>
<td>3.63 a</td>
<td>1.03 a</td>
</tr>
<tr>
<td>B1</td>
<td>Mountain soil + perlite</td>
<td>27.89 b</td>
<td>3.51 b</td>
<td>14.00 b</td>
<td>2.50 b</td>
<td>0.79 b</td>
</tr>
<tr>
<td>B2</td>
<td>Turba + perlite</td>
<td>27.61 b</td>
<td>4.16 a</td>
<td>14.00 ab</td>
<td>3.04 a</td>
<td>0.89 a</td>
</tr>
<tr>
<td>B3</td>
<td>Mountain soil + turb + perlite</td>
<td>30.05 a</td>
<td>3.72 b</td>
<td>16.00 a</td>
<td>3.17 a</td>
<td>0.96 a</td>
</tr>
<tr>
<td>CV</td>
<td>%</td>
<td>4.86</td>
<td>7.24</td>
<td>12.74</td>
<td>11.96</td>
<td>13.14</td>
</tr>
</tbody>
</table>

Note: The data provided are obtained through a basic effects study and represent the mean values of growth parameters observed in relation to various PGPR and substrates. The measurements were represented as averages and underwent statistical analysis to confirm their validity. Distinct letters (‘a’, ‘b’, ‘c’) signify statistically significant disparities across treatments, as determined by Tukey’s post hoc test (p<0.05). The Coefficient of Variation (CV %) quantifies the relative variability of the data and aids in assessing the consistency of the results.

3.1.3 Effect of Treatments on Aerial Growth Parameters in S. Parahyba Seedlings

Applying PGPR and substrate treatments had distinct effects on the height and stem diameter (Figure 3). The treatment of control + mountain soil + peat + perlite resulted in the highest average height of 33.7 cm, with statistical significance (p<0.05). The treatment PM3-8 combined with peat and perlite resulted in the shortest size, measuring 26.1 cm, with a statistically significant difference (p<0.05). Treatment T14 exhibited a maximum diameter of 4.8 mm, whereas a minor diameter of 3.1 mm was recorded with treatment T12 (p<0.01). With an average of 15, no variations in leaf number were identified (p>0.05).

Figure 3

Effect of treatments on aerial growth of S. parahyba seedlings

Note: Values that have a common letter in the figure indicate that there are no statistically significant differences between them (p > 0.05), as determined by Tukey's post hoc test. The labels range from T1 to T15, representing
the number of treatments. The following treatments were used: a control group inoculated with distilled water (Control), *S. marcescens* (PM3-8), *P. protegens* (CHAO), *E. absuriae* (PM3-14), and *A. calcoaceticus* (PM2-12). The soil composition consists of mountain soil (Tm), peat (T), and perlite (P).

### 3.1.4 Effect of *A. Calcoaceticus* and Substrates on Leaf Weight

For wet weight, the data ranged from 1.89 to 3.75 g, with Treatment T14 yielding the highest mean wet weight of 3.75 g, suggesting a superior hydration status or possibly a larger biomass accumulation under this treatment regime. Interestingly, Treatment T10 showed the lowest mean wet weight at 1.89 g, which may indicate less vigorous growth or a response to potentially suboptimal conditions for water uptake or retention. In contrast, the dry weight measurements, which reflect the actual biomass minus water content, varied from 0.61 to 1.11 g. Treatment T14 also led in this category with the highest mean dry weight of 1.11 g, reinforcing the observation that this particular treatment was most conducive to biomass accumulation. The lowest mean dry weight was observed in Treatment T1 at 0.61 g, aligning with the observation of lower wet weight in this group and potentially pointing to a less productive treatment in terms of biomass. The introduction of *A. calcoaceticus* resulted in a significant increase in the weight of both wet and dry leaves in seedlings grown in turba + perlite + mountain soil + turba + perlite (p<0.05) (Figure 4).

**Figure 4**

*Effect of A. calcoaceticus and substrates on leaf weight of S. parahyba seedlings*

Note: Values sharing a common letter in the figure indicate no significant differences between them (p > 0.05), according to Tukey's post hoc test. The labels range from T1 to T15, corresponding to the number of treatments. The following treatments were used: a control group inoculated with distilled water (Control), *Serratia marcescens* (PM3-8), *P. protegens* (CHAO), *E. absuriae* (PM3-14), and *A. calcoaceticus* (PM2-12). The soil composition consists of mountain soil (Tm), turba (T), and perlite (P).
3.1.5 Effect of *P. protegens* on Root Volume

*P. protegens* in turba + perlite increased the average root volume to 3.67 cm$^3$ vs. 1 cm$^3$ in the control (p<0.05) (Figure 5). Utilizing *P. protegens* in a mixture of peat + perlite substantially enhanced the average root volume of *S. parahyba* seedlings, reaching 3.67 cm$^3$. This rise was substantially higher than the control group, which had a root volume of 1 cm$^3$, and the other treatments (p<0.05, Figure 5).

**Figure 5**
*Effect of* *P. protegens* *in peat + perlite on root volume of* *S. parahyba* *seedlings*

Note: Values sharing a common letter in the figure imply no statistically significant differences between them (p > 0.05), as determined by Tukey’s post hoc test. The labels range from T1 to T15, representing the number of treatments. The following treatments were used: a control group inoculated with distilled water (Control), *S. marcescens* (PM3-8), *P. protegens* (CHA0), *E. absuriae* (PM3-14), and *A. calcoaceticus* (PM2-12). The soil consists of mountain soil (Tm), peat (T), and perlite (P).

4 DISCUSSION

The application of PGPR in agriculture has gained considerable interest due to its potential to improve plant growth and health (Upadhyay et al., 2022). The study highlights how PGPRs positively influence germination, growth, plant health, and soil quality and how interaction with different substrates can optimize these benefits. The findings of this study are consistent with previous research on the essential role of PGPR in modulating the rhizosphere.
and improving plant health (Basu et al., 2021; Kong & Liu, 2022). Rhizobacteria that enhance plant growth are a type of endophytic bacteria located on plant roots. They facilitate plant growth and engage in beneficial interactions with both plant and soil microorganisms (Forni et al., 2017).

4.1 SEED GERMINATION

The initial germination phase is critical for the life cycle of plants, and the results obtained corroborate that PGPR significantly improves this process (Widawati & Suliasih, 2018). PGPRs facilitate germination by altering nutrient availability and regulating hormones, providing an environment conducive to initial plant development (Gul et al., 2023; Pérez-García et al., 2023; Widawati & Suliasih, 2018). Using seeds treated with Plant Growth-Promoting Rhizobacteria (PGPR) is an effective method of bio-priming for introducing beneficial microbial inoculants into the rhizosphere or soil (Mitra et al., 2021).

4.2 PLANT GROWTH

Remarkable improvements in the height and diameter of PGPR-treated plants were observed, a finding that supports previous research on the role of PGPRs in promoting plant hormones and biological solutes (Khan et al., 2020). The changes may be related to indole-3-acetic acid (IAA) production by PGPRs, which influence cell expansion and elongation, critical for plant growth (Pii et al., 2015).

4.3 ROOT DEVELOPMENT

The root system, crucial for plant stability and nutrient absorption, benefits significantly from the presence of Plant Growth-Promoting Rhizobacteria (PGPR). Studies like Ahkami et al. (2017) and Ji et al. (2022) demonstrate that PGPRs enhance root architecture. This enhancement not only implies better access to soil nutrients but also a heightened ability to withstand environmental stresses. Such improvements might be attributed to PGPRs' role in hormone regulation and nutrient uptake efficiency. These insights are vital for agricultural practices, offering strategies for crop management under diverse environmental conditions. The potential of PGPRs in sustainably boosting crop resilience and yield, through targeted root development, opens new avenues in agricultural biotechnology.
4.4 NUMBER OF LEAVES AND BIOMASS WEIGHT

An increase in the number of leaves and biomass is a significant indicator of healthy plant growth. This study's findings, in line with prior research by Bashan et al. (2016) and Nadeem et al. (2014), suggest that Plant Growth-Promoting Rhizobacteria (PGPR) contribute to more efficient photosynthesis and better carbon allocation. This process leads to a more robust biomass. By delving deeper into the biochemical pathways affected by PGPRs, such as how they enhance the photosynthetic efficiency or influence carbon assimilation in plants, we can gain a more nuanced understanding of their impact. Additionally, exploring recent experimental data linking PGPR treatments to these specific growth improvements would solidify the connection. Understanding these mechanisms is crucial for agricultural applications, potentially leading to strategies for enhancing crop yield and sustainability through natural means.

4.5 PLANT HEALTH AND RESILIENCE

In addition to influencing growth, PGPRs have been shown to improve overall health and plant resistance against pathogens and abiotic stress conditions (Calvo et al., 2014; Sandhya et al., 2010). This protective effect is due to the induction of plant defense systems and the production of antibiotics and siderophores by PGPRs, which limit pathogen growth (Berendsen et al., 2012; Pieterse et al., 2014). PGPR inoculation also has positive implications for the soil microbiome, enriching it and promoting a community structure that benefits plant health (Bakker et al., 2013; Mendes et al., 2013).

4.6 INTERACTION BETWEEN SUBSTRATES AND PGPR

The interaction between substrates and Plant Growth-Promoting Rhizobacteria (PGPR) is a key factor in plant growth optimization. Recent research by Gul et al. (2023) and Kunal et al. (2023) emphasizes the need for careful substrate selection to maximize PGPR efficacy. Understanding how different substrates, such as organic matter-rich soils or specific mineral compositions, enhance PGPR performance can provide critical insights. This interaction plays a significant role in nutrient uptake, disease resistance, and stress tolerance, as evidenced by studies like Bashan et al. (2016) and Berendsen et al. (2012). Incorporating examples of
successful substrate-PGPR combinations from recent agricultural practices would further demonstrate the practical implications of these findings.

4.7 LIMITATIONS AND FUTURE DIRECTIONS

Although this study has produced noteworthy results, it is not devoid of limitations. The experiment's conditions were controlled so that they might differ from those in the field. Future research could explore the efficacy of PGPRs and substrates under different environmental conditions and with a broader range of plant species (Khan et al., 2020). This study reaffirms the relevance of PGPRs in sustainable agriculture. PGPR inoculation, especially when combined with appropriate substrate selection, emerges as a promising strategy to improve agricultural productivity (Zhang et al., 2020).

5 CONCLUSION

This study represents a notable progression in sustainable forestry, providing insights into the potential transformative impact of PGPR on the development of *S. parahyba*. The results, obtained from a rigorous experimental methodology, emphasize the effectiveness of inoculation with certain strains of PGPR in enhancing germination and promoting vigorous development in controlled environments. The strain *A. calcoaceticus* exhibited remarkable characteristics in facilitating the growth of above-ground biomass, which could impact existing forest management strategies significantly. Moreover, the research uncovers the significant interplay between PGPR and the composition of the substrate, a relationship that has received less attention in current scholarly discourse. The available research indicates that utilizing peat-based substrates is highly advantageous in optimizing the advantages derived from PGPR. This conclusion holds significant implications for developing effective soil management methods in the context of reforestation efforts.

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