

## PLANT GROWTH-PROMOTING BACTERIA IN THE CONTROL OF *FUSARIUM SP*

## BACTÉRIAS PROMOTORAS DE CRESCIMENTO VEGETAL NO CONTROLE DE *FUSARIUM SP*

## BACTERIAS PROMOTORAS DEL CRECIMIENTO VEGETAL EN EL CONTROL DE *FUSARIUM SP*



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### ABSTRACT

The use of plant growth-promoting bacteria (PGPB) has emerged as a sustainable alternative for agricultural management and control of phytopathogens. This study aimed to evaluate the potential of PGPB in controlling *Fusarium sp.* and promoting plant development. The experiment was conducted in vitro at the Laboratory of Microbial Interactions and Biotechnology (LIMBio/IF Goiano – Campus Ceres), using the bacteria *Bacillus amyloliquefaciens*, *Bacillus subtilis*, and *Priestia aryabhatai*. The antagonism assay was performed by co-inoculation in PDA medium, and the mycelial growth of the fungus was measured to calculate the relative control index. The data were subjected to analysis of variance and Tukey's test ( $p < 0.05$ ). All treatments showed a significant effect in controlling the pathogen, with *Bacillus amyloliquefaciens* standing out, exhibiting the highest antagonism index (69.87%), statistically differing from *Bacillus subtilis* (31.57%) and *Priestia aryabhatai* (15.30%). The results indicate that *B. amyloliquefaciens* has high potential as a biocontrol agent for *Fusarium sp.*, and may contribute to reducing the use of agrochemicals and developing more sustainable agricultural practices.

**Keywords:** Biocontrol. *Fusarium Sp.* *Bacillus*. Sustainability. Microorganisms.

### RESUMO

O uso de bactérias promotoras de crescimento de plantas (BPCP) tem se destacado como alternativa sustentável para o manejo agrícola e controle de fitopatógenos. Este estudo teve como objetivo avaliar o potencial de BPCP no controle de *Fusarium sp.* e na promoção do desenvolvimento vegetal. O experimento foi conduzido in vitro no Laboratório de Interações

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Microbianas e Biotecnologia (LIMBio/IF Goiano – Campus Ceres), utilizando as bactérias *Bacillus amyloliquefaciens*, *Bacillus subtilis* e *Priestia aryabhatai*. O ensaio de antagonismo foi realizado por co-inoculação em meio BDA, e o crescimento micelial do fungo foi mensurado para cálculo do índice relativo de controle. Os dados foram submetidos à análise de variância e teste de Tukey ( $p < 0,05$ ). Todos os tratamentos apresentaram efeito significativo no controle do patógeno, com destaque para *Bacillus amyloliquefaciens*, que apresentou maior índice de antagonismo (69,87%), diferindo estatisticamente de *Bacillus subtilis* (31,57%) e *Priestia aryabhatai* (15,30%). Os resultados indicam que *B. amyloliquefaciens* possui elevado potencial como agente de biocontrole de *Fusarium sp.*, podendo contribuir para a redução do uso de agroquímicos e para o desenvolvimento de práticas agrícolas mais sustentáveis.

**Palavras-chave:** Biocontrole. *Fusarium Sp.* Bacillus. Sustentabilidade. Microrganismos.

## RESUMEN

El uso de bacterias promotoras del crecimiento vegetal (BPCP) se ha destacado como una alternativa sostenible para el manejo agrícola y el control de fitopatógenos. Este estudio tuvo como objetivo evaluar el potencial de las BPCP en el control de *Fusarium sp.* y en la promoción del desarrollo vegetal. El experimento se llevó a cabo *in vitro* en el Laboratorio de Interacciones Microbianas y Biotecnología (LIMBio/IF Goiano – Campus Ceres), utilizando las bacterias *Bacillus amyloliquefaciens*, *Bacillus subtilis* y *Priestia aryabhatai*. El ensayo de antagonismo se realizó mediante coinoculación en medio BDA, y el crecimiento micelial del hongo fue medido para el cálculo del índice relativo de control. Los datos fueron sometidos a análisis de varianza y prueba de Tukey ( $p < 0,05$ ). Todos los tratamientos presentaron un efecto significativo en el control del patógeno, destacándose *Bacillus amyloliquefaciens*, que presentó el mayor índice de antagonismo (69,87%), diferenciándose estadísticamente de *Bacillus subtilis* (31,57%) y *Priestia aryabhatai* (15,30%). Los resultados indican que *B. amyloliquefaciens* posee un alto potencial como agente de biocontrol de *Fusarium sp.*, pudiendo contribuir a la reducción del uso de agroquímicos y al desarrollo de prácticas agrícolas más sostenibles.

**Palabras clave:** Biocontrol. *Fusarium sp.* Bacillus. Sostenibilidad. Microorganismos.

## 1 INTRODUCTION

The use of biological inputs based on beneficial soil microorganisms, such as Plant Growth Promoting Bacteria (BPCP), stands out as one of the most promising technologies to achieve the sustainability of agricultural systems. These bacteria can promote significant improvements, from traits linked to shoot and root growth to enzymatic protection against biotic and abiotic stresses (Kaushal; Wani, 2016; Porto et al., 2022).

These innovations are relevant to today's agriculture, as they help to address challenges associated with unsustainable agricultural practices. Among the main problems are the excessive use of agrochemicals, deforestation, monoculture, soil exhaustion, agriculture in inadequate areas, inadequate use of water, waste of resources, and pollution of soil and water, which compromise the health of ecosystems and productive sustainability (Kaushal; Wani, 2016; Porto et al., 2022).

Microorganisms also act on the availability of phosphorus to crops, converting forms of ions into forms that can be assimilated by plants. In addition, they produce enzymes and organic acids in the region near the roots, favoring the dissolution of phosphorus present in the soil in mineralized mineral and organic forms (Porto et al., 2022).

In crop management, different microorganisms act as soil and plant pathogens, causing damage and losses. Thus, control strategies have been widely investigated, with emphasis on more sustainable alternative methods. Among the alternatives, *Bacillus subtilis* has mechanisms of action that influence plant development, with benefits associated with improved nutrition (biological nitrogen fixation and phosphate solubilization) and growth promotion related to phytohormone production (Swarnalakshmi et al., 2020). In a study with passion fruit, the application of *Bacillus subtilis* on leaves demonstrated a preventive and curative effect in the control of the phytopathogen *Fusarium* sp. (Chen, Lee, and Huang, 2021).

*Bacillus amyloliquefaciens* stands out as an important biocontrol agent, being able to suppress phytopathogens by mechanisms related to fungal cell wall degradation and changes in cell permeability. In addition, contact with the pathogen can induce the production of antifungal compounds, such as lipopeptides, intensifying the antagonist action (Abreu et al., 2022).

*Priestia aryabhatai*, in turn, acts as a promoter of plant growth and contributes to tolerance to water stress through the production of exopolysaccharides, which help retain water in the soil and induce physiological adaptations in plants, favoring development in conditions of water deficit (Embrapa, 2021). Additionally, the use of bacteria can reduce the need for chemical fertilizers, while also favoring growth and increasing productivity.

Therefore, this work aims to evaluate the potential of plant growth-promoting bacteria in the control of *Fusarium* sp. and in the promotion of plant development, through the *in vitro* analysis of the bacteria *Bacillus subtilis*, *Bacillus amyloliquefaciens* and *Priestia aryabhatai*, in order to compare their efficiency in antagonism to the phytopathogen.

## 2 MATERIAL AND METHODS

The experiment was conducted at the Laboratory of Microbial Interactions and Biotechnology (LIMBio), located at the Federal Institute of Education, Science and Technology of Goiano (IF Goiano – Ceres Campus), in the municipality of Ceres, Goiás, Brazil (15°21'02"S latitude, 49°35'55"W longitude, and altitude of 564 m). The bacterial strains used were commercial products: *P. aryabhatai*, *B. subtilis*, *B. amyloliquefaciens* CCT 7986 (BioHidric –  $1.0 \times 10^8$  colony-forming units (CFU) mL<sup>-1</sup>, SoluBio, Jataí, Brazil), UFV S1 (Bio Raiz Pro –  $1.0 \times 10^8$  CFU mL<sup>-1</sup>, SoluBio, Jataí, Brazil) and CCT 7994 (SoluStrong –  $1.0 \times 10^8$  CFU mL<sup>-1</sup>, SoluBio, Jataí, Brazil).

To perform the antagonism assay to the phytopathogenic fungus, the bacterial suspensions were co-inoculated *in vitro* with the phytopathogenic fungus *Fusarium* spp. 5 mm discs of fungal mycelium were inoculated in the center of Petri dishes containing potato-dextrose-agar culture medium (B.D.A) and, at 2 cm equidistant from the disc, 5  $\mu$ L of bacterial culture were deposited. The control consisted only of the mycelial disc of the fungus deposited in the center of the plate.

The plates were incubated at 28 °C in a B.O.D. incubator, and the experiment was conducted in triplicate. The diameter of the mycelium was measured with the aid of a caliper until the control treatment reached the edge of the plate. The percentage of suppression for each treatment was calculated using the relative index (RI) according to Trivedi et al. (2008).

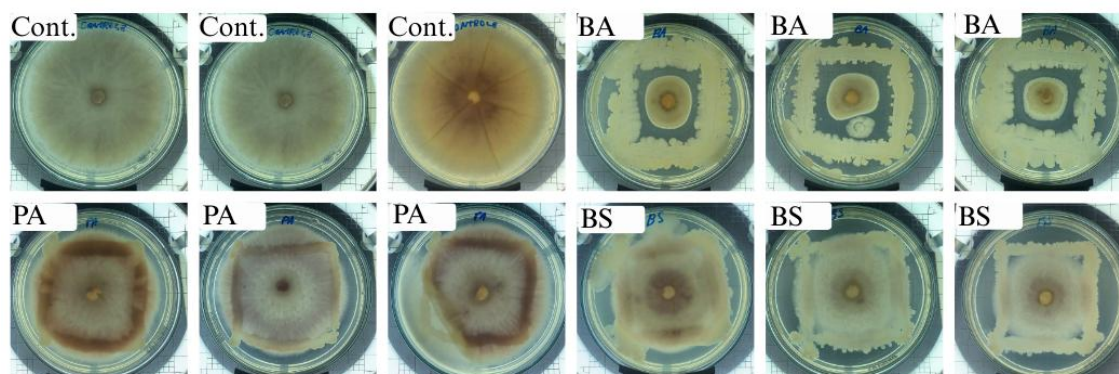
All data collected were initially tested for normality. If the prerequisites were met, the results were submitted to analysis of variance (ANOVA) followed by Tukey's test, using R and RStudio software (R Core Team, 2024). All results were considered significant at  $p < 0.05$ .

## 3 RESULTS AND DISCUSSIONS

All three bacteria tested showed a significant antagonistic effect against *Fusarium* sp. (Figure 1).

**Figure 1**

*In vitro* bacterial antagonism test to *Fusarium* sp.



Legend: Cont.: Control; BA: *Bacillus amyloliquefaciens*; PA: *Priestia aryabhatai*; BS: *Bacillus subtilis*. Source: Prepared by the authors.

The analysis of variance indicated a significant effect of the treatments on the evaluated variable ( $p < 0.0001$ ), evidencing differences between the means, as shown in Table 1. According to Tukey's test, the treatment with *Bacillus amyloliquefaciens* presented the highest relative control index, with 69.87%, differing statistically from the treatments with *Bacillus subtilis* (31.57%) and *Priestia aryabhatai* (15.30%), which also differed from each other, forming distinct response groups. These results demonstrate that the treatment with *Bacillus amyloliquefaciens* provided significantly superior performance compared to the others when controlling *in vitro* the phytopathogenic fungus *Fusarium* sp.

**Table 1**

Relative Index (RI) of *in vitro* bacterial antagonism to *Fusarium* sp.

Treatments	IR (%)
<i>Bacillus amyloliquefaciens</i>	69.86667 to
<i>Bacillus subtilis</i>	31.56667 b
<i>Priestia aryabhatai</i>	15.3 c
F.V	p-value
Treatment	0,001**
CV (%)	26,11

Legend: Different letters in the column differ significantly by Tukey's test. \*Significant at the level of 5% by Tukey's test; \*\*Significant at the level of 1% by Tukey's test; nsNot significant. F.V.: source of variation; C.V. (%): coefficient of variation. Source: Prepared by the authors.

These results corroborate the findings of Elanchezhiyan *et al.* (2018), which obtained a control percentage of 48.3% in the *in vitro* control of *Fusarium oxysporum* f. sp. *Lycopersici* with the FZB 24 strain of *B. amyloliquefaciens*. Similarly, Sundaramoorthy and Balabaskar (2013) reported the efficacy of *B. subtilis* isolates in inhibiting up to 40% the mycelial growth of

*F. oxysporum* f. sp. *lycopersici* *in vitro*, which is responsible for causing vascular wilt disease in tomato crops.

Species of *Bacillus* sp. are known to secrete peptides with antimicrobial action, such as phengicin, bacillomycin, and bacteriocin (Ramarathnam *et al.*, 2007; Xie *et al.*, 2009).

It was verified by Yi *et al.* (2024) that the ZK-9 strain of *B. Amyloliquefaciens* produces pectinase, protease, cellulase, and amylase, important enzymes involved in the ability of this bacterium to inhibit the growth of phytopathogenic fungi by disrupting the integrity and permeability of the cell membrane, disrupting metabolic pathways, and preventing pathogens from developing. The authors also observed that the bacterium showed high efficacy in field control, reducing ear fusarium by 71.76% and wheat crown rot by 82.14%, both caused by the fungus *Fusarium graminearum*.

Guleria *et al.* (2016) isolated an alkaline protease gene from the SP1 strain of *Bacillus amyloliquefaciens* and evaluated its antagonist activity *in vitro* against *F. oxysporum* using the agar diffusion technique. The authors observed that the enzyme was highly effective, reducing the growth of the pathogen by 35.07%, which highlights the role of the enzymes produced by *B. amyloliquefaciens* in its ability to control phytopathogenic fungi, such as *Fusarium* sp.

#### 4 CONCLUSION

The results demonstrate that the bacteria evaluated had a significant effect on the *in vitro* control of *Fusarium* sp., especially *Bacillus amyloliquefaciens*, which showed the highest efficiency of antagonism. *Bacillus subtilis* showed an intermediate effect, while *Priestia aryabhatai* had a lower performance. Thus, *B. amyloliquefaciens* stands out as a promising alternative for the biocontrol of the pathogen, with potential application in sustainable agricultural systems.

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