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Potential of *Streptomyces* sp strains isolated from fallow soils for the control of *Botrytis* sp in blueberries

Jorge Luis Leiva-Piedra [1]*, Yarman Roamin Seclen-Pozo [2], Ingrid Aracely Romero-Becerra [3]

[1] *Maestro en Protección de Cultivos, Universidad Tecnológica del Perú, Perú. (*Autor correspondiente: jleiva@utp.edu.pe*)
[2] *Estudiante de Ingeniería Industrial, Universidad Tecnológica del Perú, Perú*
[3] *Estudiante de Ingeniería Industrial, Universidad Tecnológica del Perú, Perú*

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ABSTRACT

Grey mould, caused by *Botrytis* sp., is one of the main phytosanitary constraints in blueberry production, due to its high incidence in the field and post-harvest and the increase in isolates resistant to conventional fungicides. In this context, the present study aimed to determine the potential of strains of the genus *Streptomyces* spp. isolated from barren soils for the biological control of *Botrytis* sp. in blueberries. Strains JCH7, JCH5 and JCH3 were isolated from arid soils in the district of Puerto Eten (Peru) and characterized morphologically, biochemically and genetically. Antagonistic activity was evaluated using dual confrontation and well diffusion assays using crude extracts, compared with a commercial synthetic fungicide. The results showed inhibitions greater than 58% in dual confrontation and up to 80.98% in well diffusion, with strain JCH3 standing out for its greater anti-fungal efficacy. Biochemical tests showed the production of key hydrolytic enzymes (cellulases, lipases and proteases), as well as chitinase activity in two of the strains. Genetic identification using the 16S rRNA gene confirmed JCH7 and JCH3 as *Streptomyces mutabilis*, while JCH5 showed high genetic divergence. Overall, the results confirm the high potential of *Streptomyces* spp. from fallow soils as biological control agents of *Botrytis* sp. in blueberries.

Keywords: Streptomyces spp.; biological control; Botrytis sp.; blueberry; antifungal metabolites.

Potencial das cepas de *Streptomyces* sp isoladas de solos em pousio para o controle de *Botrytis* sp em mirtilos

RESUMO (PT-BR)

O mofo cinza, causado por *Botrytis* sp., representa uma das principais limitações fitossanitárias na produção de mirtilo, devido à sua alta incidência no campo e pós-colheita e ao aumento de isolamentos resistentes a fungicidas convencionais. Nesse contexto, o presente estudo teve como objetivo determinar o potencial de cepas do gênero *Streptomyces* spp. isoladas de solos áridos para o controle biológico de *Botrytis* sp. em mirtilo. As cepas JCH7, JCH5 e JCH3 foram isoladas de solos áridos do distrito de Puerto Eten (Peru) e caracterizadas em nível morfológico, bioquímico e genético. A atividade antagonista foi avaliada por meio de ensaios de confronto duplo e difusão em poço utilizando extratos brutos, comparando-se com um fungicida sintético comercial. Os resultados mostraram inibições superiores a 58% em confronto duplo e até 80,98% em difusão em poço, destacando a cepa JCH3 por sua maior eficácia antifúngica. Os testes bioquímicos evidenciaram a produção de enzimas hidrolíticas importantes (celulases, lipases e proteases), bem como atividade quitinase em duas das cepas. A identificação genética através do gene 16S rRNA confirmou JCH7 e JCH3 como *Streptomyces mutabilis*, enquanto JCH5 apresentou alta divergência genética. Em conjunto, os resultados confirmam o alto potencial de *Streptomyces* spp. de solos baldios como agentes de controle biológico de *Botrytis* sp. em mirtilos.

Palavras-Chaves: *Streptomyces* spp.; controle biológico; *Botrytis* sp.; mirtilo; metabólitos antifúngicos.

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Potencial de las cepas de *Streptomyces* sp aisladas de suelos en barbecho para el control de *Botrytis* sp en arándanos

RESUMEN (SPANISH)

El moho gris, causado por *Botrytis* sp., representa una de las principales limitantes fitosanitarias en la producción de arándano, debido a su alta incidencia en campo y postcosecha y al incremento de aislamientos resistentes a fungicidas convencionales. En este contexto, el presente estudio tuvo como objetivo determinar el potencial de cepas del género *Streptomyces* spp. aisladas de suelos eriazos para el control biológico de *Botrytis* sp. en arándano. Las cepas JCH7, JCH5 y JCH3 fueron aisladas de suelos áridos del distrito de Puerto Eten (Perú) y caracterizadas a nivel morfológico, bioquímico y genético. La actividad antagonista se evaluó mediante ensayos de enfrentamiento dual y difusión en pozo utilizando extractos crudos, comparándose con un fungicida sintético comercial. Los resultados mostraron inhibiciones superiores al 58 % en confrontación dual y hasta 80,98 % en difusión en pozo, destacando la cepa JCH3 por su mayor eficacia antifúngica. Las pruebas bioquímicas evidenciaron la producción de enzimas hidrolíticas clave (celulasas, lipasas y proteasas), así como actividad quitinasa en dos de las cepas. La identificación genética mediante el gen 16S rRNA confirmó a JCH7 y JCH3 como *Streptomyces mutabilis*, mientras que JCH5 mostró alta divergencia genética. En conjunto, los resultados confirman el alto potencial de *Streptomyces* spp. de suelos eriazos como agentes de control biológico de *Botrytis* sp. en arándano.

Palabras clave: *Streptomyces* spp.; control biológico; *Botrytis* sp.; arándano; metabolitos antifúngicos.

1. Introduction

Grey mould, caused by *Botrytis cinerea*, is one of the main phytosanitary problems affecting blueberry production and quality worldwide, due to its ability to infect flowers, fruits and senescent tissues under conditions of high humidity and moderate temperatures (Roca et al., 2021). This pathogen compromises field yield and causes significant post-harvest losses, as it can develop even at low storage temperatures, making it difficult to control in long logistics chains (Chen et al., 2025). Several authors agree that the severity of grey mould has increased in recent years, associated with climatic variations, greater inoculum pressure and inadequate management practices, which have led to reports of losses that compromise the profitability of the berry industry (Almeida et al., 2023). Furthermore, the emergence of isolates resistant to multiple groups of fungicides exacerbates the situation, as it limits the effectiveness of conventional programmes and increases dependence on chemicals, creating additional problems related to safety and market restrictions (Plesken et al., 2021).

Likewise, blueberries (*Vaccinium* spp.) are recognised as a highly functional food due to their content of phenolic compounds (especially anthocyanins), vitamins, fibre and minerals (Stull et al., 2024). In addition, production has experienced rapid global expansion in terms of area and trade over the last decade, with a marked entry into demanding markets such as exports in 2024-25 to North America (174 tonnes), Europe (76 tonnes) and China (37 tonnes). In the production-commercial sphere, Peru has consolidated its position as the world's leading exporter of fresh blueberries, surpassing Chile and competing closely with Spain, Morocco and the United States (MIDAGRI, 2025). Likewise, blueberries have become one of the fastest growing crops over the last decade. According to the Food and Agriculture Organisation of the United Nations (FAO) and the International Blueberry Organisation (IBO), global blueberry production rose from approximately 1.2 million tonnes in 2020 to more than 1.8 million tonnes in 2024, representing growth of nearly 50% in less than five years. This increase is explained by growing demand in markets such as the United States, China, Europe, and the Middle East (International Blueberry Organisation, 2024; Jiao et al., 2021). On the other hand, genetic innovation and the use of advanced technology have enabled the introduction of improved varieties (Proarándanos, 2025). Among the blueberry varieties exported for the 2024/25 season, Ventura is reported to have the largest share of acreage, followed by Biloxi, Sekoya Pop and Rocío, among others, reflecting a transition towards materials with greater firmness, soluble solids and post-harvest suitability (Proarándanos - SENASA, 2024).

However, entities such as the International Society for Horticultural Science (ISHS) reported an increase in the incidence of fungal diseases associated with irregular climatic conditions, such as high humidity and moderate temperatures, between 2021 and 2025, which favour the proliferation of B. cinerea (Csorba et al., 2023). In response, the Fungicide Resistance Action Committee (FRAC) indicated in 2024 that B. cinerea already exhibits single and multiple resistance to several chemical groups, reducing the effectiveness of traditional control programmes (FRAC, 2024). This loss of effectiveness has a direct impact on the industry, generating additional costs, post-harvest losses and risks of rejection in markets with stricter maximum residue limits (Agriculture and Agri-Food Canada, 2023; Wang et al., 2021). In this context, B. cinerea ranks among the two most problematic and significant pathogens (Chen et al., 2025). Its ability to grow at low temperatures (even close to 5 °C) makes it a persistent risk during the storage and transport of export fruit (Zhang et al., 2021). When agronomic management is insufficient and logistics chains are lengthy, losses associated with grey mould can reach reductions ranging from 30% to 50% of the exportable volume.

The most common control has been based on fungicides with different modes of action (e.g., anilinopyrimidines, dicarboximides, hydroxyanilides, methyl benzimidazole carbamates, succinate dehydrogenase inhibitors [SDHI], and quinone inhibitors [QoI]) (Suthar et al., 2022). However, in recent years there has been an increase in the frequency of B. cinerea isolates with single and multiple resistance to these chemical groups, which seriously compromises the effectiveness of phytosanitary programmes. This loss of effectiveness not only limits control in the field and post-harvest, but also generates increasing regulatory pressure on producers, who must strictly comply with the maximum residue limits established by the most demanding international markets. These include the European Union, regulated by the European Food Safety Authority (EFSA), recognized for applying the most restrictive MRL standards; the United States, supervised by the Environmental Protection Agency (EPA) and the Department of Agriculture (USDA); and the United Kingdom, under the guidelines of the Food Standards Agency (FSA) (Sharma et al., 2022).

Similarly, strategic markets such as China, through the General Administration of Customs of the People's Republic of China (GACC), and Canada, regulated by the Canadian Food Inspection Agency (CFIA), maintain rigorous protocols that condition commercial access to fruit free of unauthorised molecules and with adequate post-harvest quality. In this scenario, marked by the advance of fungicide resistance and the intensification of international requirements, it is essential to incorporate more sustainable management strategies, including the use of biological control agents with antifungal capacity (Apaza, 2020). In recent years, species of the genus *Streptomyces* have established themselves as a biological alternative with high potential for the management of fungal diseases in high-value crops. These actinobacteria are notable for their ability to produce secondary metabolites with strong antifungal action, generate inhibitory volatile organic compounds, and secrete hydrolytic enzymes such as chitinases and glucanases that degrade the cell walls of pathogens such as *Botrytis cinerea*. In addition, they compete effectively for space and nutrients in the rhizosphere and can induce defensive responses in plants, strengthening their natural resistance. Thanks to this set of mechanisms, *Streptomyces* is emerging as a key agent for the development of modern and sustainable biofungicides, especially in agricultural systems where pressure to reduce chemical residues is increasing (Silva, 2020).

Scientific evidence has confirmed that *Streptomyces* spp. is an effective and sustainable alternative for controlling *Botrytis cinerea*, inhibiting mycelial growth and conidia germination in in vitro trials, as well as reducing the incidence and severity of post-harvest moulds when incorporated into integrated pre- and post-harvest management programmes (Chen et al., 2025; Yong et al., 2022). The development and validation of commercial formulations based on these actinobacteria such as *Streptomyces griseoviridis* K61 and *Streptomyces lydicus* WYEC 108 demonstrates their technological and regulatory viability as biocontrol agents, although their efficacy depends on factors such as environmental conditions, timing of application, and compatibility with post-harvest practices (Dow et al., 2023). Additional trials have shown that various Streptomyces strains have high antifungal capacity, with inhibitions reaching up to 88% against *Colletotrichum* *musae* and between 7.3% and 61.2% against *Botrytis cinerea*, associated with the production of secondary metabolites, bioactive compounds, and hydrolytic enzymes that directly affect the structure and development of the pathogen (Rios & Evangelista, 2022). Complementarily, Montesdeoca-Flores et al. (2023) corroborated these results by identifying that *Streptomyces netropsis* A52M significantly inhibited the growth and sporulation of B. cinerea both in vitro and in post-harvest fruits, reinforcing the potential of the *Streptomyces* genus as a key biotechnological tool for sustainable control strategies in crops of high commercial value, such as blueberries.

Given this scenario, marked by the increased incidence of grey mould, the loss of fungicide efficacy and the growing demands of international markets, there is an urgent need to identify safer, more sustainable management alternatives that are adapted to local conditions. This study seeks to determine the potential of *Streptomyces* sp strains isolated from fallow soils for the control of *Botrytis* sp in blueberries.

2. Material and Methods

2.1 Study site and sampling

Soil sampling was carried out on fallow land in the district of Puerto Eten, Lambayeque, Peru, characterized by arid conditions, sparse vegetation cover and low organic matter content. These environments exert high selective pressure on the soil microbiota, favoring the presence of actinobacteria with adaptive mechanisms and biosynthetic potential. The sampling site is shown in Figure 1.

Figure 1 - Soil sampling area for *Streptomyces* spp isolation
Figura 1 – Área de amostragem do solo para isolamento de *Streptomyces* spp

Source: Prepared by the authors
Fonte: Elaborado pelos autor

The soil in the study area was classified as sandy loam, with an electrical conductivity of 24.1 µS cm^{-1} and a slightly acidic pH (6.8). The organic matter content was 0.2%, with concentrations of phosphorus at 5.6 ppm, potassium at 275 ppm, and calcium carbonate at 0.5%. These characteristics are representative of low-fertility arid soils. The samples were collected at a depth of approximately 5 cm, obtaining around 1 kg of soil per sampling point, considering five points distributed throughout the study area.

2.2 Isolation of bacteria of the genus Streptomyces spp

For the isolation of soil bacteria, each 1 kg sample was placed on a tray, then 100 g of soil was taken from several random points. This soil was suspended in 900 ml of sterile distilled water, and serial dilutions were made from this solution up to 10-3. From the final dilution, 1 µl was taken and inoculated onto starch-casein agar (SCA) plates supplemented with nystatin (µg mL^{-1}), which were then incubated at 28 °C for seven days (Usha Nandhini et al., 2018).

2.3 Isolation of Botrytis spp. from blueberry fruit

The phytopathogenic fungi were isolated using the standardized protocol for infested plant tissue (blueberry fruit). The samples were placed in a humid chamber for a period of five days, then the tissues with the presence of the fungus were cut with a scalpel and immersed in 1% sodium hypochlorite, then rinsed three times with sterile distilled water. These tissues were placed on Petri dishes with potato dextrose agar and incubated at 25°C for seven days (Agrios, 2005).

2.4 Macroscopic and microscopic identification of bacteria of the genus Streptomyces spp.

The morphological characterization of the isolated strains included the evaluation of macroscopic and microscopic characteristics, considering the morphology of the colonies, pigmentation, aerial and substrate mycelium formation, and hyphal structure. (1) macroscopic characterization: The strains were cultured on oat agar for eight days (Bergey, 1993).

The colour, texture of the colonies, presence of aerial mycelium and production of diffusible pigments in the medium were evaluated. (2) Microscopic characterization: The strains were cultured on oat agar supplemented with 0.1% nystatin. Sterile coverslips were placed at a 45° angle on the agar and incubated at 25°C for 15 days. The coverslips were then observed under an optical microscope to evaluate hyphal septation and the arrangement of aerial and substrate mycelium (Goredema et al., 2020).

2.5 Dual confrontation test between Streptomyces spp. and Botrytis spp.

The antagonistic activity of the *Streptomyces* strains (JCH3, JCH5, and JCH7) was evaluated using the dual confrontation technique on potato dextrose agar plates. The bacterial suspensions were seeded at one end of the plate, while 5 mm diameter mycelial discs of *Botrytis* spp. were placed at the opposite end. The plates were incubated at 28 ± 2 °C for seven days. Plates with the fungus but without the bacteria were used as negative controls. The inhibition rate (IR) of radial growth was calculated using the following equation (1) (Pakdaman Sardrood, 2013).

$$\%\ de\ IR = \frac{Dc - Dt}{Dc}\ x\ 100 \tag{1}$$

Where: IR = inhibition rate (%), Dc = diameter of the negative control (mm), Dt = diameter of the treatment or positive control (mm).

2.6 Biochemical tests to determine enzyme activity

The production of hydrolytic enzymes associated with biological control, including chitinase, protease, lipase, and cellulase, was evaluated using specific media. The plates were incubated at 22 °C for 5 to 8 days, and enzyme activity was determined by the presence of hydrolysis halos around the colonies (Wei et al., 2017).

2.7 Production and extraction of secondary metabolites

The strains were reactivated in TSB broth and subsequently cultured in liquid ISP-1 medium under constant agitation (150 rpm) for seven days at 28 °C. The cultures were centrifuged to obtain cell-free supernatants, which were subjected to extraction with ethyl acetate (1:1, v/v). The organic phase was evaporated and the residue obtained was resuspended in 10% methanol for use in antifungal assays (Wei et al., 2017).

2.8 In vitro inhibition test between bioactive compounds and commercial chemically synthesized fungicide

The antifungal activity of the crude extracts was evaluated using the well diffusion method in PDA plates. Forty µL of each extract was placed in wells 0.2 cm in diameter, with a mycelial disc of the phytopathogen in the centre of the plate. All tests were performed in triplicate. Benzimidazole was used as a synthetic fungicide reference to compare the efficacy of the extracts (Kumar et al., 2025).

2.9 Genetic identification of bacteria of the genus Streptomyces

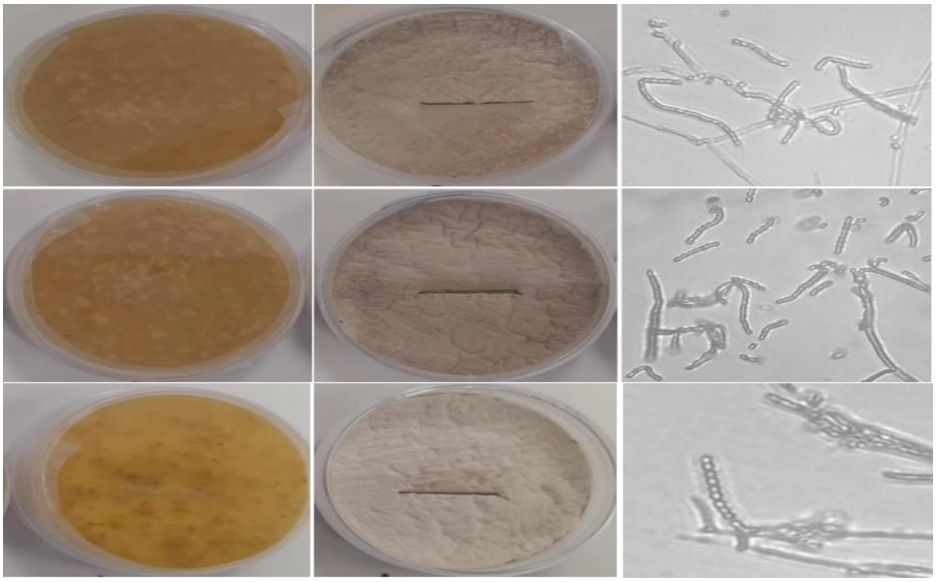
Genomic DNA from the strains was extracted using an optimized protocol for Streptomyces. The 16S rRNA gene was amplified by PCR using the universal primers 27F and 1492R. The amplified products were sequenced and the sequences obtained were analyzed using Ugene software and the EzBioCloud database to determine the taxonomic affiliation of the strains

3. Results and Discussion

3.1 Macroscopic and microscopic characterization

Figure 2 shows, in comparative arrangement, the macroscopic (colonies on plate) and microscopic (filamentous structures) characteristics of three bacterial isolates of the genus *Streptomyces*. At the macroscopic level, the colonies have a dry, opaque, and powdery texture, with rough and cracked surfaces, typical features of filamentous actinobacteria.

Figure 2 - Macroscopic and microscopic characteristics of *Streptomyces* strains isolated from soil.
Figura 2 – Características macroscópicas e microscópicas das cepas de *Streptomyces* isoladas do solo.



Source: Prepared by the authors
Fonte: Elaborado pelos autor

The colour varies between cream, beige, and yellowish tones, suggesting metabolic differences between strains, particularly in the production of secondary pigments and diffusible metabolites. The development of aerial mycelium is evident in mature colonies, with a cottony covering raised above the substrate, indicative of a complete development cycle. This pattern is characteristic of *Streptomyces*, whose growth involves progressive differentiation from substrate mycelium to aerial mycelium and sporulation. The absence of mucus and the brittle appearance of the colonies allow other non-filamentous bacterial genera to be ruled out.

At the microscopic level, fine, branched, septate hyphae are observed, with chains of spores arranged in a straight or slightly curved manner. The structures observed correspond to chains of arthrospores formed by segmentation of the aerial mycelium, a diagnostic feature of the *Streptomyces* genus. The uniformity of the hyphal diameter and the presence of well-defined septa confirm the actinobacterial nature of the isolates.

3.2 Dual confrontation

Table 1 shows the results of the dual confrontation test between three *Streptomyces* strains (JCH7, JCH5 and JCH3) against *Botrytis* spp.

Table 1 – Results of the inhibition test (dual confrontation) of *Streptomyces* spp. strains against *Botrytis* spp.
Tabela 1 – Resultados do teste de inibição (confronto duplo) das cepas de *Streptomyces* spp. contra *Botrytis* spp.

Phytopathogenic fungus	Strain	Measurement cm	% Inhibition	x̄ % inhibition
Botrytis spp	JCH7	1.7	60.47%	58.59%
		1.9	52.50%	
		1.6	62.79%	
	JCH5	2.0	53.49%	58.04%
		1.5	62.50%	
		1.8	58.14%	

JCH3	2.0	53.49%	58.76%
	1.6	60.00%	
	1.6	62.79%	
Control	4.3	0.00	0.00%
	4.0	0.00%	
	4.3	0.00%	

Source: Prepared by the authors
Fonte: Elaborado pelos autor

All the strains evaluated showed a marked antagonistic capacity, with average percentages of mycelial growth inhibition greater than 58%. The JCH3 strain recorded the highest average inhibition value (58.76%), followed by JCH7 (58.59%) and JCH5 (58.04%), showing homogeneous anti-fungal behaviour among the isolates. In contrast, the control treatment showed complete mycelial growth of *Botrytis* spp., with average diameters close to 4.3 cm and total absence of inhibition, confirming that the reduction in fungal growth observed in the treatments was due exclusively to the antagonistic action of the *Streptomyces* strains.

The analysis of variance (Table 2) indicated highly significant differences between treatments (F = 163.590; p-value = 0.000), demonstrating that the presence of *Streptomyces* spp. had a statistically significant effect on the inhibition of *Botrytis* spp. The coefficient of determination ($R^2 = 0.984$; adjusted $R^2 = 0.978$) indicates that more than 98% of the variability observed in pathogen inhibition is explained by the effect of the bacterial strains, reflecting the high robustness of the experimental model. Tukey's multiple comparison test ($p < 0.005$) presented in Figure 3 confirms that all *Streptomyces* strains form a statistically homogeneous group among themselves but clearly differentiated from the control. This result indicates that all, although there are no significant differences between the strains evaluated, they all have a consistent and reproducible antagonistic effect against *Botrytis* spp.

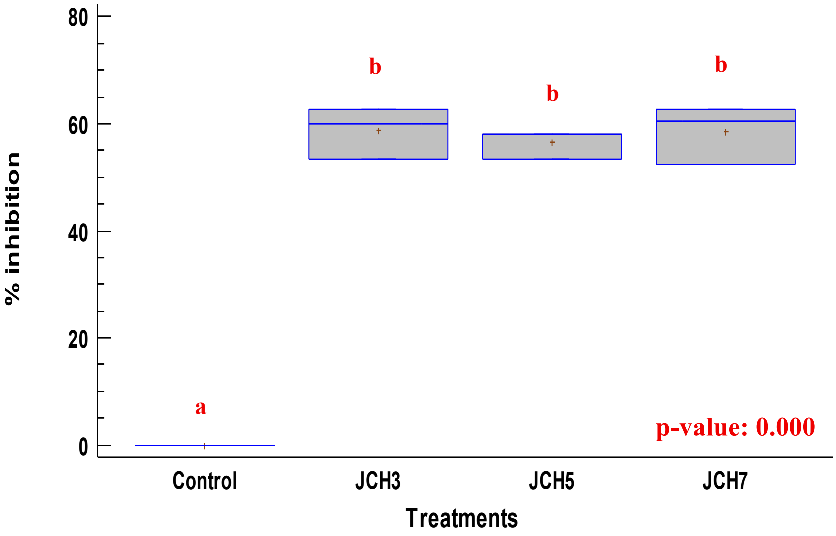
Table 2 – Analysis of variance to determine the effect of *Streptomyces* spp strains on the inhibition of *Botrytis* sp
Tabela 2 – Análise de variância para determinar o efeito das cepas de *Streptomyces* spp. na inibição de *Botrytis* sp.

	Sum of squares	gl	Quadratic mean	F-ratio	P-value
Between groups	7511.601	3	2503.867	163.590	0.000
Within groups	122.446	8	15.306		
Total	7634.047	11			

$R^2 = 0.984$ (adjusted $R^2 = 0.978$).

Source: Prepared by the authors
Fonte: Elaborado pelos autor

Figure 3 - Statistical analysis and Tukey's test ($p < 0.005$) to evaluate the % inhibition of *Streptomyces* spp. strains isolated from *Botrytis* spp. in the dual confrontation test.
Figura 3 – Análise estatística e teste de Tukey ($p < 0,005$) para avaliar a % de inibição das cepas de *Streptomyces* spp. isoladas de *Botrytis* spp. no teste de confronto duplo.



Source: Prepared by the authors
Fonte: Elaborado pelos autor

The variance components show that the largest proportion of total variability is concentrated between treatments (7511.601) compared to intra-treatment variation (122.446), indicating a high consistency of replicates within each group and, at the same time, a marked separation of the overall effect attributed to the strains evaluated. Likewise, the ratio between the mean square between groups (2503.867) and within groups (15.306) results in a high F (163.590), confirming that the magnitude of the biological effect associated with Streptomyces spp. on the inhibition of Botrytis sp. is dominant over the experimental variation. In explanatory terms, the values of $R^2 = 0.984$ and adjusted $R^2 = 0.978$ support that the model describes practically the entire observed response, suggesting that the differences in % inhibition are mainly due to the treatment applied and not to random noise. Overall, this variance structure supports the conclusion that the trial has high power to detect real differences between the control and the actinobacteria treatments, reinforcing the robustness of the statistical inference.

The subgroups of the Tukey test show a clear separation between the control (letter 'a') and the *Streptomyces* spp. strains (letter 'b'), confirming that the presence of these actinobacteria significantly increases inhibition compared to treatment without antagonist. The assignment of the same subgroup to all strains indicates that, under the test conditions, their responses are statistically equivalent to each other; therefore, the result suggests a consistent antagonistic effect shared by the evaluated strains, rather than a marked differential performance between isolates. Additionally, the p-value = 0.000 reported in the figure confirms that the overall difference between groups is highly significant, consistent with the ANOVA, and supports that the observed pattern is not attributable to chance. This statistical homogeneity between strains can be interpreted as evidence of a reproducible biocontrol potential, useful for supporting their selection as candidates, subsequently prioritising non-statistical criteria (e.g., stability, sporulation, ease of formulation, or performance under uncontrolled conditions).

3.3 Biochemical potential of Streptomyces isolated from barren soil

Table 3 shows the biochemical profile of the bioactive compounds produced by *Streptomyces* spp. strains JCH7, JCH5 and JCH3, evaluated by enzymatic activity tests for cellulase, chitinase, lipase and protease.

Table 3 – Results of biochemical tests performed with bioactive compounds extracted from isolated strains of *Streptomyces* spp.
Tabela 3 – Resultados dos testes bioquímicos realizados com compostos bioativos extraídos de cepas isoladas de *Streptomyces* spp.

Strain	Replicate	Cellulase	Chitinase	Lipase	Protease
JCH7	1	+	+	+	+
	2	+	+	+	+
	3	+	+	+	+
JCH5	1	+	−	+	+
	2	+	−	+	+
	3	+	−	+	+
JCH3	1	+	+	+	+
	2	+	+	+	+
	3	+	+	+	+

Source: Prepared by the authors
Fonte: Elaborado pelos autor

The results show that the three strains exhibited positive activity for cellulase, lipase and protease in all replicates analyzed, indicating consistent and reproducible expression of these hydrolytic enzymes. In contrast, chitinase activity showed a differential pattern among the strains. Strains JCH7 and JCH3 showed positive activity in all the replicas, while strain JCH5 was negative for this enzyme. This variability suggests metabolic and genetic differences between the isolates, possibly associated with the presence or absence of specific gene clusters involved in chitin degradation. The consistency of the results between replicates indicates metabolic stability in the production of hydrolytic enzymes, reinforcing the reliability of the data obtained and the biotechnological potential of the strains evaluated.

3.4 Inhibition test – diffusion in well

Table 4 presents the results of the well diffusion assay, which evaluated the antifungal activity of bioactive compounds extracted from Streptomyces spp. strains JCH7, JCH5, and JCH3 against Botrytis spp.

Table 4 – Results of the inhibition test (well diffusion) of *Streptomyces* spp. strains against *Botrytis* spp.
Tabela 4 – Resultados do teste de inibição (difusão em poço) das cepas de *Streptomyces* spp. contra *Botrytis* spp.

Phytopathogenic fungus	Strain	Measurement cm	% Inhibition	X % inhibition
Botrytis spp	JCH7	1.6	77.14%	75.59%
		1.7	75.00%	
		1.7	74.63%	
	JCH5	2.1	70.00%	72.23%

	1.9	72.06%	
	1.7	74.63%	
JCH3	1.3	81.43%	80.98%
	1.4	79.41%	
	1.2	82.09%	
Iprodione	0.0	100.00%	100.00%
	0.0	100.00%	
	0.0	100.00%	
Control	7.0	0.00%	0.00%
	6.8	0.00%	
	6.7	0.00%	

Source: Prepared by the authors
Fonte: Elaborado pelos autor

The results show significant inhibition of the pathogen's mycelial growth in all treatments evaluated, with average inhibition percentages ranging from 72.23% to 80.98%. The JCH3 strain showed the highest average inhibitory effect (80.98%), followed by JCH7 (75.59%) and JCH5 (72.23%). These values reflect the high antifungal efficacy of the crude extracts, particularly in the case of JCH3, whose performance was like that observed in the treatment with iprodione, used as a reference fungicide (100% inhibition). In contrast, the control treatment showed complete mycelial growth of Botrytis spp., with no detectable inhibition, confirming that the antifungal effect observed is due exclusively to the action of the bioactive compounds.

The analysis of variance (Table 5) showed highly significant differences between treatments ($F = 2400.03$; $p = 0.000$), indicating that the inhibition of Botrytis spp. growth is strongly influenced by the type of extract applied. The high coefficient of determination ($R^2 = 0.950$; adjusted $R^2 = 0.947$) suggests that 95% of the variability in inhibition percentages is explained by the effect of the treatments, demonstrating the high statistical robustness of the experimental model. Tukey's multiple comparison test ($p < 0.005$), shown in Figure 4, confirmed the existence of statistically differentiated groups. Iprodione formed an independent group with total inhibition, while Streptomyces extracts were grouped according to their antifungal potency, with JCH3 standing out as the most effective biological treatment, clearly differentiated from the negative control.

Table 5 – Analysis of variance to determine the effect of *Streptomyces* spp strains on the inhibition of *Botrytis* sp, diffusion in well.

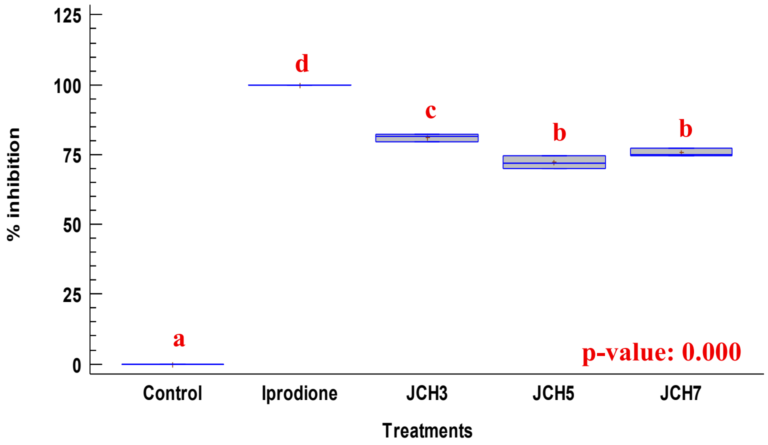
Tabela 5 – Análise de variância para determinar o efeito das cepas de *Streptomyce*s spp. na inibição de *Botrytis* sp., difusão em poço.

Source	Sum of Squares	Gl	Mean Square	F-ratio	P-Value
Between groups	17600.4	4	4400.09	2400.03	0.0000
Intra-group	18.3335	10	1.83335		
Total (Corr.)	17618.7	14			

$R^2 = 0.950$ (adjusted $R^2 = 0.947$).

Source: Prepared by the authors
Fonte: Elaborado pelos autor

Figure 4 - Statistical analysis and Tukey's test ($p < 0.005$) to evaluate the % inhibition of bioactive compounds extracted from isolated strains of *Streptomyces* spp. against *Botrytis* spp. in the well diffusion assay.
Figura 4 – Análise estatística e teste de Tukey ($p < 0,005$) para avaliar a % de inibição dos compostos bioativos extraídos das cepas isoladas de *Streptomyces* spp. contra *Botrytis* spp. no ensaio de difusão em poço



Source: Prepared by the authors
Fonte: Elaborado pelos autor

The variance partition confirms that the response (% inhibition) is dominated by the effect of the treatment: the sum of squares between groups (17,600.4) greatly exceeds the intra-group (18.3335), showing that the differences observed are mainly due to the type of extract applied and not to experimental dispersion. This pattern is reflected in a very high F (2400.03) with p = 0.000, indicating a clear statistical separation between treatments. Furthermore, the values of $R^2 = 0.950$ and adjusted $R^2 = 0.947$ support that the model explains approximately 95% of the variability in the % inhibition, confirming a highly consistent data structure and an adequate design capacity to discriminate real effects. Taken together, these results support the conclusion that the well diffusion assay is highly statistically robust, with low residual variation, which strengthens the validity of the biological effect attributed to the extracts of Streptomyces spp. compared to the control.

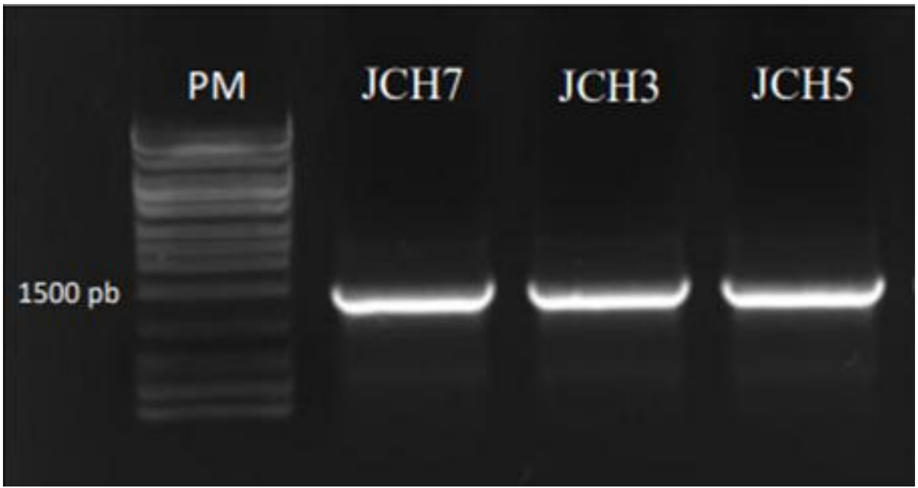
The subgroups of the Tukey test ($p < 0.005$) show the existence of multiple statistically differentiated groups, confirming that there are not only overall differences between treatments, but also a gradient of antifungal efficacy. In particular, the fungicide Iprodione is classified as an independent group with total inhibition, marking the upper limit of performance in the trial and serving as a positive reference. In contrast, the negative control is clearly separated from the rest, indicating an absence (or minimal presence) of inhibitory effect. Among the biological treatments, Streptomyces extracts are distributed into groups according to their potency, and the grouping pattern supports that some strains exhibit significantly higher activity than others under the same experimental conditions. This statistical differentiation suggests that biocontrol performance is not homogeneous among strains and justifies prioritising extracts with greater inhibition (highlighting the superior performance of the most effective biological treatment).

3.5 Genetic identification

Figure 5 shows the result of the amplification of the 16S rRNA gene of strains JCH7, JCH3 and JCH5 by PCR, showing single, intense bands close to 1,400 bp in 1% agarose gel. This result confirms the adequate quality of the extracted genomic DNA, the specificity of the universal primers used, and the correct amplification of the 16S rRNA region, widely used for bacterial taxonomic identification.

Table 6 summarises the results of the comparative analysis of the sequences obtained against the EzBioCloud database.

Figure 5 - Result of the amplification of the 16S rRNA region in 1% agarose gel electrophoresis.
Figura 5 - Resultado da amplificação da região 16S rRNA em eletroforese em gel de agarose a 1%



Source: Prepared by the authors
Fonte: Elaborado pelos autor

Table 6 - Molecular identification results of the sequences obtained by comparison with the EzBioCloud database
Tabela 6 – Resultados da identificação molecular das sequências obtidas por comparação com o banco de dados EzBioCloud

Strain	bp No.	Closest homologue	Identity (%)/Completeness	Variation ratio
JCH7	1398	*Streptomyces mutabilis*	98.39/99.60	20/1367
JCH3	1403	*Streptomyces mutabilis*	98.08/100	22/1365
JCH5	875	*Streptomyces* spp	66.60/100	278/840

Source: Prepared by the authors
Fonte: Elaborado pelos autor

Strains JCH7 and JCH3 showed high percentages of identity with *Streptomyces mutabilis* (98.39% and 98.08%, respectively), with completeness values above 99%. These results fall within the accepted threshold for species-level assignment using the 16S rRNA gene, allowing both strains to be identified as *S. mutabilis* or phylogenetically very close lineages.

In contrast, strain JCH5 showed significantly lower identity (66.60%) with *Streptomyces* spp. sequences, despite having 100% completeness. The high number of nucleotide variations observed suggests that this strain cannot be assigned with certainty to a known species, indicating a possible affiliation to a highly divergent or potentially undescribed lineage within the *Streptomyces* genus.

4. Discussion

4.1 Macroscopic and microscopic characterization

Figure 2 shows a comparative overview of the macroscopic and microscopic characteristics of the three bacterial isolates of the genus Streptomyces. At the macroscopic level, the colonies were observed to be dry, opaque and powdery, with a rough and cracked surface, without the presence of mucus; in addition, colour variation was observed between cream, beige and yellowish tones, and the development of aerial mycelium was observed in mature colonies. At the microscopic level, thin, branched, septate hyphae were observed, with chains of spores arranged in a straight or slightly curved manner; these structures correspond to chains of arthrospores formed by segmentation of the aerial mycelium, a feature compatible with the genus *Streptomyces*.

The macroscopic characteristics observed in the isolates are broadly consistent with those described for species of the genus *Streptomyces* isolated from arid and oligotrophic soils. The dry, powdery texture, together with the formation of abundant aerial mycelium, has been associated with mechanisms of adaptation to water stress and radiation, common in environments with low nutrient availability (Zakalyukina et al., 2024);(Tangjitjaroenkun et al., 2025). These adaptations favour aerial spore dispersal and the ecological persistence of the genus. The colour variability observed among colonies suggests physiological differentiation between strains, probably linked to the differential activation of biosynthetic pathways of secondary metabolites. Recent studies have shown that pigmentation in *Streptomyces* is often correlated with the production of antibiotics, phenolic compounds, and molecules with antifungal activity(Luo et al., 2025);(Hoskisson et al., 2024). In this context, colour differences could subsequently explain variations in antagonistic capacity against phytopathogens.

From a microscopic point of view, the presence of septate hyphae and spore chains confirms the taxonomic identity of the genus and reflects an active physiological state. The structural differentiation of the aerial mycelium is directly associated with the activation of gene clusters responsible for the synthesis of antibiotics and hydrolytic enzymes, such as chitinases and proteases, which are key in the biological control of phytopathogenic fungi (Manteca et al., 2019);(El-Akshar et al., 2025);(Rioseras et al., 2014) (Rioseras et al., 2014).

The morphology observed coincides with reports of *Streptomyces mutabilis* and related species, which exhibit well-branched hyphae and smooth or slightly curved spore chains, associated with high biosynthetic potential(Yan et al., 2025). This aspect is particularly relevant, as it has been shown that strains with a high degree of mycelial differentiation are more efficient in the production of broad-spectrum antifungal metabolites(Nazari et al., 2023; Solórzano et al., 2025). Taken together, the combination of macroscopic and microscopic features observed in the figure supports the correct assignment of the isolates to the genus *Streptomyces* and suggests a high aptitude to produce secondary metabolites with potential application in biological control. These structural characteristics, typical of actinobacteria adapted to extreme environments, reinforce the value of arid soils as reservoirs of strains of high biotechnological interest.

4.2 Dual confrontation

The results obtained show that the *Streptomyces* spp. strains evaluated have high biological control potential against *Botrytis* spp., a necrotrophic phytopathogen responsible for severe economic losses in numerous crops of agricultural importance. The inhibition levels observed (≈58–59%) are consistent with recent reports describing reductions in *Botrytis* spp. mycelial growth of between 50% and 70% in in vitro trials using antagonistic actinobacteria (Louviot et al., 2024; Solórzano et al., 2025). The absence of significant differences between strains suggests that the mechanism of antagonism is shared and probably multifactorial. Recent studies have shown that *Streptomyces* spp. exert their antifungal effect through the simultaneous production of polyene antibiotics, siderophores, hydrolytic enzymes (chitinases, proteases, and glucanases), and volatile organic compounds, which act synergistically on the cell wall and plasma membrane of *Botrytis* spp (Cuervo et al., 2023) (Nazari et al., 2023; Torres-Rodriguez et al., 2022)

The high value of the F ratio statistic and the high coefficient of determination reinforce the experimental consistency of the trial and confirm that the variability observed is mainly due to the antagonistic interaction between bacteria and fungi. Similar results have been reported for strains of *Streptomyces lydicus*, *S. griseoviridis* and *S. mutabilis*, which have shown comparable efficacy in controlling *Botrytis cinerea* through dual confrontation (Hassan et al., 2025;(Montesdeoca-Flores et al., 2023);(Olanrewaju & Babalola, 2019) . From an ecological perspective, the observed efficacy can be attributed to the adaptive capacity of *Streptomyces* from edaphic environments with high microbial competition. In these ecosystems, selective pressure favours the evolution of strains with highly active secondary metabolism, capable of inhibiting aggressive fungal competitors such as *Botrytis* spp(Luo et al., 2025) ;(Shepherdson et al., 2023) (Abbasi et al., 2021)

Although the inhibition percentages obtained are lower than those achieved by synthetic fungicides under controlled conditions, several studies indicate that these levels are sufficient to significantly reduce the incidence of *Botrytis* in agricultural systems when integrated into integrated pest management (IPM) strategies. In addition, the use of *Streptomyces* spp. offers key environmental advantages, such as lower risk of resistance, absence of toxic residues, and compatibility with sustainable agricultural practices(Solórzano et al., 2025; Steinberg & Gurr, 2020; Vurukonda et al., 2018) . Overall, the results of the dual confrontation confirm that strains JCH3, JCH5, and JCH7 are promising candidates for the development of antifungal bioformulations for the control of *Botrytis* spp., supporting their potential application in agricultural biocontrol programmes.

4.3 Biochemical potential of Streptomyces isolated from barren soils

Biochemical assays revealed a consistent enzymatic profile in the three strains evaluated (*Streptomyces* spp. JCH7, JCH5, and JCH3), with positive cellulase, lipase, and protease activity in all replicates (3/3) for each strain. In contrast, chitinase activity showed a differential pattern, being positive in JCH7 and JCH3 (3/3 replicates) and negative in JCH5 (3/3 replicates). Taken together, these results demonstrate intra-strain stability (no variation between replicates) and specific functional differences associated mainly with chitinolytic capacity, which was restricted to two of the three isolates. The production of hydrolytic enzymes by *Streptomyces* spp. is one of the most important mechanisms associated with their biological control capacity against phytopathogenic fungi. In this study, the simultaneous detection of cellulases, lipases and proteases in all the strains evaluated confirms the high metabolic versatility of isolates from barren soils, environments characterized by low nutrient availability and high microbial competition.

The cellulase activity observed in the three strains suggests an important role in the degradation of structural polysaccharides present in plant debris and tissues colonized by phytopathogens, facilitating access to nutrients and weakening the structure of the fungal host. Recent studies have shown that cellulases produced by *Streptomyces* indirectly contribute to pathogen inhibition by altering the edaphic microenvironment and limiting fungal colonization(Yang et al., 2026) Nunes et al., 2025(Dow et al., 2023; Nunes et al., 2025). Lipase activity, consistently detected in all strains, is associated with the degradation of membrane lipids and hydrophobic compounds, causing alterations in the cellular integrity of fungi. It has been reported that lipases from *Streptomyces* spp. act synergistically with other antifungal metabolites, increasing membrane permeability and enhancing the overall inhibitory effect(Alam et al., 2022; Alzahrani et al., 2025; Junior et al., 2025) .

Likewise, the presence of protease activity in all strains evaluated indicates a direct mechanism of degradation of structural proteins and essential enzymes of the pathogen. Extracellular proteases produced by *Streptomyces* spp. have been widely associated with fungal mycelium lysis and radial growth inhibition in in vitro assays(El-Akshar et al., 2025; Homero et al., 2021) ;(Kumar et al., 2020). Chitinase activity, which is only positive in strains JCH7 and JCH3, is of particular importance, given that chitin is one of the main components of the cell wall of many phytopathogenic fungi. The ability to hydrolyze this polymer gives these strains an additional competitive advantage over *Botrytis*, as demonstrated in recent studies where chitinolytic *Streptomyces* strains achieved reductions of more than 70% in the mycelial growth of agricultural pathogens(Anis Mufida et al., 2024) (Lv et al., 2021; Nazari et al., 2023) (Umar et al., 2021)

The absence of chitinase activity in strain JCH5 suggests that its antifungal effect is based on alternative mechanisms, such as the production of polyene antibiotics, siderophores, or volatile organic compounds. This functional diversity is common in *Streptomyces* and reflects a complementary metabolic strategy, in which different strains use different biosynthetic pathways to achieve a similar antagonistic effect(Luo et al., 2025) (Cuervo et al., 2023). From an ecological perspective, the ability to produce multiple hydrolytic enzymes reflects an adaptation to barren soils, where the availability of complex substrates requires high degradative efficiency. This trait has been identified as a key characteristic of *Streptomyces* isolated from extreme environments, which often exhibit high potential for biotechnological and agricultural applications(Zakalyukina et al., 2024) (Nuguse & Kejela, 2024). The biochemical results obtained confirm that strains JCH7, JCH5 and JCH3 possess a broad enzymatic arsenal associated with the biological control of phytopathogenic fungi. The combination of cellulases, lipases and proteases in all strains, together with chitinase activity in two of them, suggests a multifactorial antagonism mechanism, supporting their potential use in the development of sustainable agricultural bio-inputs.

4.4 Inhibition test – diffusion in well

The results obtained in the well diffusion test confirm that the bioactive compounds produced by *Streptomyces* spp. strains have high antifungal potential against *Botrytis* spp. The inhibition levels above 70% observed in this study are consistent with recent research reporting similar efficacies for crude *Streptomyces* extracts against *Botrytis cinerea* in in vitro and in(Montesdeoca-Flores et al., 2023; Rammali et al., 2024; Xiao et al., 2021) (Rammali et al., 2024).

The greater efficacy observed in strain JCH3 suggests a higher concentration or diversity of antifungal metabolites in its extract, possibly associated with polyene antibiotics, macrolides, or phenolic compounds. Recent studies have shown that species such as *Streptomyces mutabilis* and *S. lydicus* produce metabolites capable of altering the permeability of the fungal membrane, inducing oxidative stress and disrupting the cell wall of *Botrytis* spp(Luo et al., 2025) (Hassan et al., 2025).

The fact that the crude extracts do not achieve the 100% inhibition observed with iprodione does not diminish their biotechnological relevance. On the contrary, several authors highlight that inhibition levels between 70% and 85% are considered highly promising for biological control agents, especially when evaluated in vitro and without optimised formulation(Mani et al., 2024) (Devi et al., 2023) (Nazari et al., 2023) (Solórzano et al., 2025). In addition, compounds of biological origin have significant environmental advantages, such as lower residual toxicity and lower risk of resistance development.

The high F-statistic value and high coefficient of determination indicate that experimental variability was minimal and that the results are highly reproducible. This statistical consistency supports the hypothesis that secondary metabolites produced by *Streptomyces* spp. are primarily responsible for the observed inhibition, rather than random factors in the trial(Devi et al., 2023) (Steinberg & Gurr, 2020). From an enzymatic point of view, the antifungal activity observed can be attributed to the combined action of hydrolytic enzymes (chitinases, proteases, and lipases) and secondary metabolites previously detected in these strains. The synergy between these compounds has been widely documented as a key mechanism for the effective inhibition of necrotrophic fungi such as *Botrytis* spp., whose chitin- and protein-rich cell wall is particularly susceptible to this type of biochemical attack(El-Akshar et al., 2025) (Cuervo et al., 2023).

Likewise, the edaphic origin of the strains (from barren soils) could explain their high biosynthetic capacity. Environments with high selective pressure tend to favour the evolution of microorganisms with intensified secondary metabolism, capable of producing bioactive compounds with a broad antifungal spectrum (Mayimele & Gurusamy, 2026) (Norte et al., 2025). The results of the well diffusion assay demonstrate that the bioactive compounds extracted from the JCH7, JCH5 and, especially, JCH3 strains of *Streptomyces* spp. are promising biological alternatives for the control of *Botrytis* spp. These findings support their potential application in integrated disease management programmes and justify further studies aimed at identifying specific metabolites, optimizing formulations and validating them under in vivo conditions.

4.5 Genetic identification

The successful amplification of the 16S rRNA gene in the three strains confirms the validity of the molecular approach used and supports its use as a primary phylogenetic marker for the identification of actinobacteria. The 16S rRNA gene remains a fundamental tool in bacterial taxonomy due to its combination of conserved and hypervariable regions, which allow genera and species to be differentiated with high reliability(Agnolucci et al., 2019; Saravana Kumar et al., 2025).

The identification of strains JCH7 and JCH3 as *Streptomyces mutabilis* is particularly relevant from a biotechnological point of view. This species has been reported to produce secondary metabolites with antifungal, antibacterial and plant growth-promoting activity, including polyene antibiotics, siderophores and hydrolytic enzymes(Tangjitjaroenkun et al., 2025) (Hassan et al., 2025). Recent studies indicate that strains of *S. mutabilis* have a high antagonistic capacity against phytopathogenic fungi such as *Botrytis*, *Fusarium*, and *Cercospora*, which is consistent with the antifungal results observed in this study(Manathunga et al., 2024) (Nazari et al., 2023) (Solórzano et al., 2025).

The identity percentages obtained (⩾ 98%) fall within the accepted range for the delimitation of bacterial species using 16S rRNA, especially when accompanied by high completeness values. However, several authors point out that values close to the threshold (98 – 98.7%) may reflect cryptic species complexes within *Streptomyces*, suggesting the possible need for complementary genomic analyses, such as ANI or whole genome sequencing, for definitive taxonomic resolution(Parab et al., 2022; Seshadri et al., 2022; Zakalyukina et al., 2024) .

The case of strain JCH5 is particularly significant. The low percentage of identity observed (66.60%), together with a high number of nucleotide variations, indicates substantial genetic divergence from the sequences deposited in public databases. This result suggests that JCH5 could correspond to a phylogenetically distant lineage within the genus *Streptomyces* or even to a species not yet described. Recent research has shown that arid and barren soils are reservoirs of actinobacteria with high genetic diversity and unique biosynthetic clusters, often absent in strains from conventional agricultural environments(Luo et al., 2025) (Goodfellow et al., 2018).

The high genetic divergence observed in JCH5 could be associated with evolutionary adaptations to extreme conditions, such as low nutrient availability, water stress, and high radiation, factors that promote the selection of highly specialised genomes. Such adaptations have been linked to the production of novel secondary metabolites with potential applications in biocontrol and agricultural biotechnology (Mayimele & Gurusamy, 2026; Dhaini et al., 2025; Shepherdson et al., 2023). The results of genetic identification confirm that the fallow soils in the study area harbour both known species of high biotechnological interest, such as *Streptomyces mutabilis*, and highly divergent lineages with potential for the discovery of new species. This genetic diversity reinforces the value of these environments as strategic sources for the prospecting of microorganisms with applications in the biological control of phytopathogens and sustainable agriculture.

Overall, the identification of Streptomyces isolates with consistent functional capabilities supports their potential as bio-inputs for reducing the environmental burden associated with chemical disease control, favoring more sustainable management strategies that are compatible with soil microbiota conservation (Nazari et al., 2023) (Wend et al., 2024) From an implementation perspective, the available evidence indicates that the transition to production scale depends mainly on stability and performance in the field, in addition to technological variables such as formulation, shelf life, and quality control, aspects that often limit the commercialization of microbial biocontrol agents (Chaudhary et al., 2024) (Verma et al., 2024). Therefore, the applied projection of these isolates is strengthened by integrating them into validation routes that include trials under real conditions, environmental safety assessment, and the development of formulations that improve persistence and efficacy, as a basis for their eventual transfer and commercial use (Wend et al., 2024) (Verma et al., 2024)

5. Conclusion

This study allowed for a comprehensive determination of the potential of strains of the genus *Streptomyces* spp. isolated from fallow soils as biological control agents against *Botrytis* sp., a pathogen of high phytosanitary relevance in blueberry cultivation. The results obtained show that fallow soils are a valuable source of actinobacteria with high antagonistic capacity and metabolic diversity.

The strains evaluated (JCH7, JCH5, and JCH3) demonstrated significant inhibition of *Botrytis* sp. mycelial growth in both dual confrontation assays and well diffusion tests, achieving inhibition percentages greater than 58% and up to 80.98% using crude extracts, confirming the production of antifungal metabolites with relevant biological activity. Strain JCH3 stood out for presenting the highest levels of inhibition, approaching the effect observed with the synthetic reference fungicide, highlighting its high potential as a biological alternative. The biochemical profile of the bioactive compounds showed consistent production of key hydrolytic enzymes —cellulases, lipases, and proteases— in all strains, as well as chitinase activity in JCH7 and JCH3, suggesting a multifactorial antagonism mechanism based on the structural degradation of the pathogen's cell wall and membrane, complemented by the action of secondary metabolites.

Genetic identification through amplification and analysis of the 16S rRNA gene confirmed that strains JCH7 and JCH3 belong to *Streptomyces mutabilis*, a species recognized for its biosynthetic capacity and application in biocontrol, while strain JCH5 showed high genetic divergence, suggesting the possible presence of an undescribed lineage, which reinforces the value of fallow soils as reservoirs of microbial diversity with biotechnological potential. The findings demonstrate that the isolated *Streptomyces* spp. strains have high potential for the biological control of *Botrytis* sp. in blueberries, making them promising candidates for the development of sustainable agricultural bio-inputs. These results support the need to continue studies aimed at characterizing specific metabolites, optimizing formulations and validating them in vivo, with a view to incorporating them into integrated disease management programmes and reducing the use of synthetic fungicides in blueberry production systems.

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