Isolation and characterization of halotolerant bacteria from soils in the municipality of Remedios, Cuba

Aislamiento y caracterización de bacterias halotolerantes procedentes de suelos del municipio Remedios, Cuba

Isolamento e caracterização de bactérias halotolerantes provenientes de solos do município Remedios, Cuba

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Crop production

Abstract

The isolation of halotolerant bacteria represents a strategy to mitigate soil salinization and promote crop adaptation. This study aimed to identify bacteria from saline soils in Remedios, Cuba, and evaluate their potential as plant growth-promoting bacteria (PGPB). Three bacterial strains tolerant to 200 mM NaCl were selected and identified as *Proteus* sp. RM 4.2, *Lysinibacillus* sp. RM 6.2, and *Tenebrionicola* sp. RM 4.1. The three strains showed the ability to fix nitrogen and produce siderophores and lytic enzymes. In tomato plant inoculation assays, *Proteus* sp. RM 4.2 significantly promoted plant growth in the evaluated cultivars, standing out as the strain with the greatest growth-promoting effect. Overall, the results indicated that the evaluated strains exhibited functional traits associated with plant growth promotion and represented a viable alternative for developing bioinoculants in saline soils.

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Resumen

El aislamiento de bacterias halotolerantes representa una estrategia para mitigar la salinización del suelo y favorecer la adaptación de los cultivos. Este estudio tuvo como objetivo identificar bacterias de suelos salinos en Remedios, Cuba, y evaluar su potencial como promotoras de crecimiento vegetal (PGPB). Se seleccionaron tres cepas bacterianas tolerantes a 200 mM de NaCl, identificadas como *Proteus* sp. RM 4.2, *Lysinibacillus* sp. RM 6.2 y *Tenebrionicola* sp. RM 4.1. Las tres cepas presentaron capacidad de fijar nitrógeno, producir sideróforos y enzimas líticas. En ensayos de inoculación en plantas de tomate, *Proteus* sp. RM 4.2 promovió significativamente el crecimiento vegetal en los cultivares evaluados, destacándose como la cepa con mayor efecto promotor de crecimiento. En conjunto, los resultados indicaron que las cepas evaluadas presentaron rasgos funcionales asociados a la promoción del crecimiento vegetal y representan una alternativa viable para el desarrollo de bioinoculantes en suelos salinos.

Palabras clave: taxonomía bacteriana, caracterización funcional, tolerancia, salinidad.

Resumo

O isolamento de bactérias halotolerantes representa uma estratégia para mitigar a salinização do solo e favorecer a adaptação das culturas. Este estudo teve como objetivo identificar bactérias de solos salinos em Remedios, Cuba, e avaliar seu potencial como promotoras de crescimento vegetal (PGPB). Três cepas bacterianas tolerantes a 200 mM de NaCl foram selecionadas e identificadas como *Proteus* sp. RM 4.2, *Lysinibacillus* sp. RM 6.2 e *Tenebrionicola* sp. RM 4.1. As três cepas apresentaram capacidade de fixar nitrogênio e de produzir sideróforos e enzimas líticas. Em ensaios de inoculação em plantas de tomate, *Proteus* sp. RM 4.2 promoveu significativamente o crescimento vegetal nos cultivares avaliados, destacando-se como a cepa com maior efeito promotor. Em conjunto, os resultados indicam que as cepas avaliadas apresentam características funcionais associadas à promoção do crescimento vegetal e representam uma alternativa viável para o desenvolvimento de bioinoculantes em solos salinos.

Palavras chave: taxonomia bacteriana, caracterização funcional, tolerância à salinidade

Introduction

The diversity of soil microorganisms worldwide is threatened by anthropogenic changes, such as land-use intensification, deforestation, and extreme weather events. These factors alter the structure and function of microbial communities, directly affecting soil ecological processes and their stability (Alzate-Zuluaga *et al.*, 2020). It is estimated that soils are home to approximately a quarter of the planet's biodiversity, including plant growth-promoting microorganisms (PGPB), which are a valuable tool to promote sustainable and environmentally friendly agriculture (Rosabal *et al.*, 2021).

Within this diversity, microorganisms that inhabit environments with high concentrations of salts stand out, particularly halotolerant bacteria, which are capable of favoring the adaptation of plants to conditions of salt stress (Guerra-Camacho *et al.*, 2024). In Cuba, more than one million hectares are affected by salinity, of which approximately 14 % corresponds to cultivated areas (García-Reyes *et al.*, 2021). Specifically, in the municipality of Remedios, in the northeast of the Villa Clara province, there are coastal settlements that are highly vulnerable to sea level rise and anthropogenic influence (Pineda-Ruiz *et al*., 2023). This phenomenon increases soil salinity and poses a threat to agricultural productivity. Despite this, information on the diversity and functionality of halotolerant bacteria in these soils is limited, which highlights the need for local studies to evaluate their potential as plant growth promoters.

In this context, the study of microorganisms in saline soils should include rhizobacteria (Velasco-Jiménez *et al.*, 2020). The evaluation of their phylogenetic and functional diversity in agroecosystems under salinity conditions allows the identification of strains capable of mitigating environmental impacts, favoring the recovery of degraded soils, and improving crop nutrition (Ahluwalia *et al.*, 2021). The municipality of Remedios, in Cuba, is characterized by the presence of vertisol soils affected by salinity (Hernández-Jiménez *et al*., 2023), which makes it a strategic site for the search for microorganisms with potential as PGPB. Therefore, the present study aimed to isolate, identify, and characterize halotolerant bacteria from soils of coastal settlements in the municipality of Remedios, in order to explore their potential use as plant growth-promoting bacterial inoculants for crops exposed to high salt concentrations.

Materials and methods

Between 2024 and 2025, slightly salinized soil samples (2.7-3.5 dS.m^{-1}) (Hernández-Jiménez *et al*., 2023) were taken from 18 farms distributed in nine coastal settlements in the municipality of Remedios, Villa Clara province, Cuba, which corresponded to independent agricultural agroecosystems and with a non-uniform sampling area. Five soil subsamples were collected from each farm at a depth of 20 cm using an auger, following a W or zigzag pattern (Villalba-Martínez and Encina-Rojas, 2024); these were then homogenized to obtain 1 kg of composite sample per farm. The composite samples were processed using the serial dilution method for the isolation of bacteria in nutrient agar culture medium (Rodríguez-Martínez and Zhurbenko, 2018). From these, the morphotypes present in the 1×10^{-5} dilution were determined; a representative strain of each morphotype was purified and assigned a code with the abbreviation RM, in reference to the municipality of Remedios. To evaluate salinity tolerance, the strains were inoculated in the same medium supplemented with different NaCl concentrations (0, 50, 150, and 200 mM) and incubated at 30 °C for 24 h, selecting only those capable of growing at the maximum concentration evaluated (200 mM).

Identification and characterization of bacterial isolates

For the identification and characterization of the selected bacteria, morphological methods, biochemical tests, and genetic analyses were combined. Colonies were described based on macroscopic characteristics (shape, color, margin, elevation, and surface), and cell shape was determined microscopically, by Gram stain in fresh 24-h cultures. Additionally, DNase and coagulase tests were performed to rule out pathogenicity and ensure the safety of the isolates (Madigan *et al*., 2019). The identification of the strains was carried out by phylogenetic analysis of the 16S rRNA gene. Macrogen Inc. (Seoul, Republic of Korea) performed amplification and sequencing using the 27f/1493r and 785F/907R universal primers, respectively. The sequences obtained were edited using BioEdit v.7.0.9.0 (Hall, 1999)

and compared with the GenBank library through BLAST searches to form a database of type strain sequences. The phylogenetic tree was built with MEGA v.7 with 1,000 bootstrap replicates (Kumar *et al*., 2016); the selection of the nucleotide substitution model was carried out with jModelTest 2 (Darriba *et al*., 2012), and the identity percentages were determined with MatGAT v.2.01 (Campanella *et al*., 2003). The sequences were deposited in GenBank under the accession numbers OR902474, OR902479, and PP051518.

Characterization of bacteria as plant growth promoters

The ability of the isolates to promote plant growth was evaluated in specific culture media; the bacteria were inoculated by stabbing and incubated at 28 °C for 24 - 48 h. In the media where the strains showed activity, evidenced by the formation of halos, the diameters of the colonies (TC) and of the halos (TH) in three directions were measured, and the activity index (AI) was calculated using the formula: IA = TH / TC.

Biological nitrogen fixation was evaluated qualitatively according to Elbeltagy *et al*. (2001), using nitrogen-free Winogradsky medium; the isolates with growth were reinoculated for four consecutive passages to rule out residual nitrogen carryover. The solubilization of phosphates and potassium was evaluated in NBRIP and Alexandrov-BPB media, respectively, supplemented with 0.001 % bromophenol blue (Nautiyal, 1999; Sugumaran and Janarthanam, 2007; Zelaya-Molina *et al*., 2025). The production of indolic compounds was determined in TSA with 1 % tryptophan using the Salkowski reagent (Gordon and Weber, 1951), and that of siderophores in CAS (Louedn *et al*., 2011; Nair *et al*., 2007). Enzyme activity was assessed by testing for peroxidases, amylases, proteases, lipases, esterases, cellulases, and peptidases in specific media (Ramos-Garza *et al*., 2016; Al-Mohaini *et al*., 2022).

Effect of strains on tomato cultivars (*Solanum lycopersicum* L.)

The inoculum of each strain was prepared in nutrient broth and multiplied in an orbital shaker (EDMUD BUHLER, Germany) at 120 rpm and 30 °C, for 24 h, until a final concentration of 1×10^8 CFU.mL^{-1} was reached, determined according to the McFarland scale (1907). Subsequently, a mixture of inoculum with distilled water was applied to the soil in a proportion of 1:10 (v/v). The tests were carried out in greenhouses under semi-controlled conditions (temperature and humidity); tomato cultivars T-60 and FL-5 (determined growth and fresh consumption) from INIFAT were used. Two seeds per pot were

sown with salinized Vertisol soil (8 dS.m^{-1}) (Hernández-Jiménez *et al*., 2023). It was irrigated with drinking water (150 mL) to avoid salt leaching and was verified by registering a final conductivity similar to the initial value. Four treatments were established with 25 plants and three replicates, and the trial was repeated after 30 days. At 60 days after sowing, the following parameters were evaluated: height, root length, stem diameter, number of leaves, and dry mass.

Statistical analysis

A completely randomized design was used in the laboratory, and a randomized block design was employed in the plant trial. The data were subjected to normality (Kolmogorov-Smirnov) and homogeneity (Cochran C, Hartley, and Bartlett) tests, one-way analysis of variance (ANOVA), and Duncan's test (α = 0.05), using STATGRAPHICS Plus version 5.0 (Statistical Graphics Corporation, 1994).

Results and discussion

From the soil collection, 15 bacterial morphotypes were obtained, with densities ranging from 2×10^6 to 1.5×10^7 CFU.g^{-1} of soil; one representative isolate of each morphotype was isolated and purified for subsequent characterization. After evaluating the growth of the 15 strains at three NaCl concentrations, only three grew at 200 mM; this evidenced their salinity tolerance and supported their selection for subsequent evaluations.

Morphological characteristics of the strains

In the selected isolates, bacillary forms predominated, forming white colonies with entire margins and a dry consistency on nutrient agar medium (Table 1).

The DNase and coagulase tests were negative, which is a favorable criterion for subsequent evaluation in plant growth promotion trials.

Taxonomic identification of isolates

Phylogenetic analysis based on the sequence of the 16S rRNA gene allowed the identification of bacterial isolates (Figure 1).

The RM 4.2 strain was grouped in the genus *Proteus*, with a 100 % identity with the type strain sequence of *P. mirabilis*, and identity percentages of 97.3-99.1 % (Table 2) with the type strain sequences of the closest species, according to the analysis with the MatGAT program (Campanella *et al*., 2003).

Table 1. Morphological characteristics of saline soil strains from coastal settlements in the municipality of Remedios in Villa Clara province, Cuba.

Strains	Cell morphology	Gram stain	Colonie color	Morphology of the colonies	Image
RM 4.1	Wide bacilli	Gram -	White	Irregular with entire margins, convex, dry, and with a sunken center containing granules	
RM 4.2	Wide bacilli	Gram -	White	Circular with irregular margins, convex and dry consistency with granules	
RM 6.2	Short bacilli	Gram +	White	Circular with entire margins, convex and dry consistency with granules	

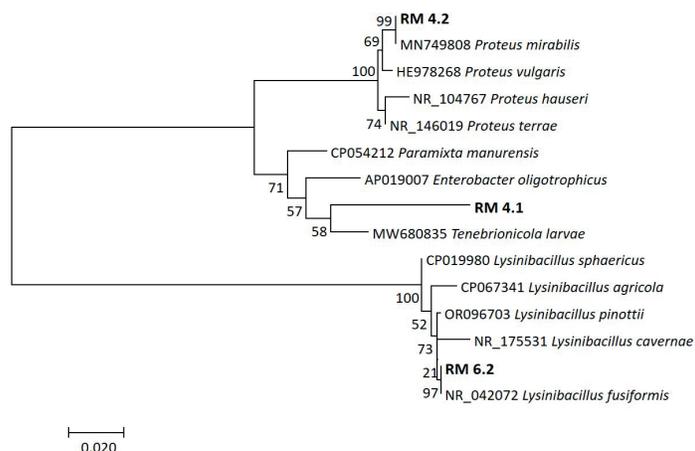RM= code in reference to the municipality of Remedios.

Figure 1. Phylogenetic tree based on the 16S rRNA gene sequences, built using the MEGA 7 program, the maximum likelihood method, and the K2P nucleotide substitution model. Branch values correspond to bootstrap percentages obtained from 1,000 replicates.

Table 2. Identity of the 16S rRNA gene sequences of the bacterial strains obtained from saline soils of Remedios, Cuba, with respect to the type strain sequences used in the phylogenetic analysis.

Strain	Type strain sequence	Identity (%)
Tenebrionicola sp. RM 4.1	MW680835 *Tenebrionicola larvae*	93.9
	AP019007 *Enterobacter oligotrophicus*	93.5
	CP054212 *Paramixta manurensis*	93.1
Proteus sp. RM 4.2	MN749808 *Proteus mirabilis*	100.0
	HE978268 *Proteus vulgaris*	99.1
	NR_104767 *Proteus hauseri*	98.5
	NR_146019 *Proteus terrae*	97.3
Lysinibacillus sp. RM 6.2	NR_042072 *Lysinibacillus fusiformis*	99.9
	OR096703 *Lysinibacillus pinotti*	99.2
	NR_175531 *Lysinibacillus cavernae*	98.4
	CP067341 *Lysinibacillus agricola*	98.4

The identity percentages were determined using the MatGAT version 2.01 program. RM= code in reference to the municipality of Remedios.

The strain RM 4.1 was grouped within the genus *Tenebrionicola*, showing proximity to *T. larvae*, the only species described of the genus, with 93.9 % identity relative to its type strain sequence. Likewise, it presented an identity range of 93.1-93.5 % with the type strain sequence, corresponding to species of the phylogenetically closest genera, according to the analysis with MatGAT (Campanella *et al*., 2003). On the other hand, the RM 6.2 strain was located within the genus *Lysinibacillus*, with greater affinity to *L. fusiformis*, with a 99.9 % identity with the type strain sequence of this species and a range of 98.4-99.2 % with the type strain sequences of the closest species, according to the analysis with MatGAT (Campanella *et al*., 2003) (Table 2).

It should be noted that the identified species are not frequent in studies of plant growth-promoting bacteria, especially *P. mirabilis*, *L. fusiformis*, and particularly *Tenebrionicola* sp., whose presence has been little documented in this type of environment.

Characterization of plant growth-promoting strains

All three strains showed growth in nitrogen-free Winogradsky medium, suggesting their ability to fix atmospheric nitrogen. The RM 4.1 strain presented significant differences with respect to the others (Table 3).

Table 3. Plant growth promotion characteristics (PGP) of bacteria from saline soils in Remedios, Cuba.

Tests	Bacterial strains			
	RM 4.1	RM 4.2	RM 6.2	ESx
Biological nitrogen fixation (diameter of colonies in cm)				
Winodrasky medium	1.86 a	1 c	1.56 b	0.0158
Nutrient solubilization index				
Phosphate (Calcium)	1.52 a	1.44 b	0	0.0050
Phosphate (Aluminum)	0	0	2.00 a	0.0171
Potassium	0	0	0	
Iron and siderophore production index				
Iron	1.25 c	1.30 b	3.18 a	0.0043
Production of indolic compounds				
Indolic compounds	+	-	+	

Equal letters do not differ significantly between rows (Duncan's Multiple Range Test, α = 0.05, n = 10). ESx= standard error. RM= code in reference to the municipality of Remedios.

Regarding phosphate solubilization capacity, differences were observed among the three strains. RM 4.1 and RM 4.2 solubilized calcium phosphate, with a predominance of RM 4.1, while only RM 6.2 solubilized aluminum phosphate (Table 3). None of the strains showed the ability to solubilize potassium. Similarly, all of them produced siderophores, with RM 6.2 standing out with significant differences compared to the others. In the production of indolic compounds, RM 4.1 and RM 6.2 were exclusively positive (Table 3).

In relation to enzyme production, the results showed a remarkable versatility among the strains evaluated (Table 4). The RM 4.2 strain distinguished itself from the rest by producing five lytic enzymes, highlighting its potential compared to the other two. Regarding amylase activity, RM 4.1 and RM 6.2 stood out, the latter being the one with the highest capacity. Only RM 4.2 showed protease and esterase activity, while both RM 4.1 and RM 4.2 produced lipases, with a predominance of MR 4.2. All three strains showed cellulase activity, with RM 6.2 standing out. Regarding peroxidase, all strains were positive.

Table 4. Production of enzymes by bacteria isolated from saline soils in Remedios, Cuba.

Enzymes	Bacterial strains			
	RM 4.1	RM 4.2	RM 6.2	ESx
Peroxidases	+	+	+	
Enzyme activity indices				
Amylases	1.25 b	0	3.55 a	0.0153
Proteases	0	1.44	0	
Lipases	1.67 b	2.43 a	0	0.0041
Esterases	0	2.95	0	
Cellulases	1.35 c	1.47 b	3.02 a	0.0035
Peptidases	0	0	0	

Equal letters do not differ significantly between rows (Duncan's Multiple Range Test, α = 0.05, n = 10). ESx= standard error. RM= code in reference to the municipality of Remedios.

Similar studies on microorganisms from saline soils have revealed how they activate tolerance and resistance mechanisms that allow them to mitigate these adverse effects through plant growth promotion, not only in the presence of salts but also of potentially toxic metals (Ahluwalia *et al.*, 2021). These manifestations described in the literature support the functionality and adaptations observed in the new strains, which reveal that they can not only survive in saline environments, but can also have positive effects in agricultural environments affected by this type of stress.

In general terms, plant growth-promoting bacteria (PGPB) have a set of attributes that support plant development, including atmospheric nitrogen fixation through nitrogenase production (Masood *et al.*, 2020), the stimulation of antioxidant enzymes such as oxidoreductase, peroxidase, ascorbate peroxidase, superoxide dismutase, catalase, glutathione reductase and adenosine triphosphatase (Liu *et al.*, 2021), as well as the synthesis of 1-aminocyclopropane-1-carboxylate (ACC) deaminase that contributes to reducing ethylene levels in plant tissues.

In addition, these bacteria reinforce the activity of soil enzymes, such as alkaline and acid phosphatases, urease, and β-glucosidase (Kumar *et al.*, 2021), and produce siderophores that facilitate plant access to iron (Jalal *et al.*, 2023). In this context, the strains obtained in the present study show characteristics that position them as promising candidates for PGPB, with emphasis on saline environments.

Studies on strains of *Proteus mirabilis* describe them as human pathogens (Mekibib *et al.*, 2015). However, some authors have reported that they are also PGPB, with the ability to solubilize phosphate and potassium (Saheed and Ikhajiagbe, 2024), in addition to producing indole-3-acetic acid (Sushma *et al.*, 2021) and siderophores (Amaresan *et al.*, 2021). These strains show tolerance to different stress conditions, such as salinity (Akintokun *et al.*, 2019), reduce proline accumulation against zinc toxicity (Suganya *et al.*, 2020), and activate antioxidant enzymes such as catalase, peroxidase, and superoxide dismutase in the presence of other metals (Sushma *et al.*, 2021).

In the case of *Lysinibacillus fusiformis*, it is known as a plant growth-promoting species (Ahsan and Shimizu, 2021) due to the production of phytohormones such as indole-3-acetic acid and abscisic acid, involved in the elongation and regulation of plant growth. It also has the ability to solubilize phosphates through the production of organic acids (fumaric, lactic, malic, citric, succinic, propionic, and acetic) and inorganic phosphorus (Passera *et al.*, 2021). In addition, its potential to synthesize antimicrobial compounds as siderophores has been described (Passera *et al.*, 2021) and enzymes with antifungal action against pathogens such as *Aspergillus niger*, *Botrytis cinerea*, *Phomopsis viticola,* and *Rhizoctonia solani* (Passera *et al.*, 2021). It also produces urease (Jibrin *et al.*, 2020) and participates in bioremediation processes for heavy metals such as cadmium and copper (Mathivanan *et al.*, 2016).

As for the genus *Tenebrionicola*, *T. larvae* is the first species described within this genus and was isolated from larvae of *Tenebrio molitor* L. (Dong *et al.*, 2022); however, there are no reports of strains of this species or possible strains related to this genus as plant growth promoters, since studies have been based mainly on their taxonomic value, rather than in their possible uses or specific biotechnological applications. The evidence found in the present study opens new lines of research that indicate that this bacterium could be applied and be of benefit to sustainable agriculture.

Together, the three strains analyzed in this study fix atmospheric nitrogen, two solubilize calcium phosphate and aluminum phosphates; all promote the production of siderophores and, in two cases, produce indolic compounds. Similarly, all three strains generate lytic enzymes, reinforcing their potential as plant growth-promoting bacteria.

Inoculation of the strains in tomato cultivars (*Solanum lycopersicum* L.)

When evaluating the effect of the application of the three strains on tomato cultivars T-60 and FL-5, established in a salinized Vertisol soil, the strain *Proteus* sp. RM 4.2 stood out, showing higher values in all growth and development indicators, with significant differences compared to the rest of the treatments in both cultivars (Table 5). In contrast to the studies of Islam *et al.* (2016) and Suganya *et al.* (2020), these results demonstrate that the strain *Proteus* sp. RM 4.2 was also effective in significantly stimulating both tomato cultivars, which is consistent with several studies describing the role of *Proteus* sp. as PGPB.

Table 5. Effect of bacterial strains isolated from saline soils in the municipality of Remedios, Cuba, on the growth and development of tomato cultivars T-60 and FL-5, in a salinized Vertisol soil during two trials.

Treatments	Plant height (cm)	Root length (cm)	Stem diameter (cm)	Number of leaves (u)	Dry plant mass (g)
Cultivar T-60					
Tenebrionicola sp. RM 4.1	6.49 b	1.38 b	0.07 b	3.81	0.26 b
Proteus sp. RM 4.2	7.31 a	2.02 a	0.13 a	4.11	0.46 a
Lysinibacillus sp. RM 6.2	6.56 b	1.40 b	0.09 b	3.80	0.31 b
Control	6.35 c	1.01 c	0.03 c	3.42	0.12 c
ESx	0.0408	0.0484	0.0066	0.3755 n.s	0.0383
Cultivar FL-5					
Tenebrionicola sp. RM 4.1	6.71 b	1.18 c	0.06 b	4.19	0.25 b
Proteus sp. RM 4.2	7.56 a	1.92 a	0.16 a	4.40	0.42 a
Lysinibacillus sp. RM 6.2	6.82 b	1.25 b	0.07 b	4.21	0.29 b
Control	5.72 c	0.86 d	0.03 c	4.09	0.10 c
ESx	0.0261	0.0281	0.0074	0.2378 n.s	0.0309

Equal letters in the same column do not differ significantly from each other, according to Duncan's Multiple Range test α = 0.05 n=25. ns: not significant. ESx= Standard error. RM= code in reference to the municipality of Remedios.

P. mirabilis has been reported to stimulate the growth of corn (*Zea mays* L.) in saline soils (Akintokun *et al.*, 2019) and improve nutrient uptake under high concentrations of Zn, which has a favorable impact on grain yield and quality (Suganya *et al.*, 2020). Likewise, its ability to increase soil nutrients and chelate iron in rice cultivars (*Oryza sativa* L.) has been described (Saheed and Ikhajiagbe, 2024).

The results of the present study confirm the capacity of the strain *Proteus* sp. RM 4.2 to promote plant growth, as it has the ability to fix nitrogen, solubilize phosphate, as well as produce siderophores, indolic compounds, and hydrolytic enzymes (peroxidases, proteases, lipases, esterases, and cellulases). The combination of these functional traits was associated with the highest promoting effects observed in the bioassays in tomato plants (*S. lycopersicum*), compared to the other strains evaluated (Table 5), which is in accordance with the

attributes previously indicated for this species (Amaresan *et al*., 2021; Saheed and Ikhajiagbe, 2024; Suganya *et al*., 2020; Sushma *et al*., 2021). However, its agricultural use requires additional studies to rule out phytopathological risks.

In the case of *L. fusiformis*, several studies have documented its ability to improve the uptake of N, P, and K, as well as the yield in corn and the development of seedlings of different crops under greenhouse conditions (Passera *et al*., 2021). Similarly, positive effects on tomato yield and nutritional quality have been reported (Jyolsna *et al*., 2021). The results obtained in this study confirm that the strain *Lysinibacillus* sp. RM 6.2 presents functional traits comparable to those previously described, and its inoculation promotes plant growth under salt stress conditions (Table 5).

For its part, the strain *Tenebrionicola* sp. RM 4.1 showed positive effects on plant growth promotion in the bioassays conducted, which are relevant given that there are no previous reports on its potential as a plant growth-promoting bacterium. This study constitutes the first record of this genus in Cuban saline soils and its first functional evaluation as a PGPB, expanding knowledge about the microbial diversity associated with saline agroecosystems. Together, these results provide relevant information for the selection of native bacteria with potential application in crop management in salinized soils in the municipality of Remedios, in Cuba.

Conclusions

The three strains isolated from saline soils in the municipality of Remedios showed functional attributes associated with plant growth promotion. Among them, the strain *Proteus* sp. RM 4.2, related to *P. mirabilis*, stood out significantly in stimulating the growth of tomato cultivars. These results indicated that the studied strains have potential for the development of bioinoculants aimed at improving crop growth in saline soils. However, additional studies under field conditions are required, as well as biosecurity evaluations, especially in the case of *Proteus* sp. RM 4.2, to rule out phytopathological risks and ensure safe use in agriculture.

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