

Research Article

Isolation, screening, and characterization of plant growth enhancing endophytic bacteria from halophytic *Heliotropium curassavicum* L. collected from salt stress areas of Srikakulam, Andhra Pradesh

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Abstract

Farmers use excessive chemical fertilizers to boost crop productivity to meet growing agricultural demands. However, this practice is costly and environmentally hazardous. Sustainable increase in crop yield can be achieved through alternatives like microbial-based fertilizers. In the quest to identify plant growth-promoting endophytic bacteria, the present study was carried out and selected unexplored halophytic plant *Heliotropium curassavicum* L. Thirteen endophytic bacterial strains were isolated from both aerial and root portions of *H. curassavicum*. These isolates were tested for salt tolerance, enzyme production, and synthesis of growth-promoting secondary metabolites, like Indole-3-acetic acid (IAA) and phosphate solubilization. Most of the isolates belonged to the Bacillus family, exhibiting varying Gram staining and biochemical reactions. The majority are Gram-positive bacteria, non-motile, spore formers, and exist in two cells or chains. All isolates could tolerate up to 10% NaCl concentration and a temperature of 42°C. Based on phenotypic, bio-chemical characteristics, isolate HCR3 showed promising properties in synthesizing IAA and phosphate solubilization abilities. The isolate HCR3 grew well upto 10% NaCl concentration and also 42°C temperature. Based on molecular characterization by using 16S rRNA gene-based analysis HCR3 isolate was identified and belonged to the Genus *Pseudomonas* with the highest similarity index with *Pseudomonas khazarica* sp. HCR3 showed IAA production of 37µg ml⁻¹, had a phosphate solubilization ability of 3.5 ppm, and recorded protease activity on gelatin medium. The findings highlight the potential of HCR3 and other strains from halophytic *H. curassavicum* L. to enhance plant growth through secondary bioactive metabolites, offering eco-friendly solutions for sustainable agriculture.

Keywords: Endophytes, Heliotrope, Indole-3-acetic acid, Plant growth promoting bacteria, *Pseudomonas khazarica*

INTRODUCTION

As the world population increases, it is insufficient to meet the rising population's food demand. So, the farmers heavily depend on the utilization of chemical fertilizers. These chemical fertilizers had many side effects, like leaching out and polluting water basins, decreasing soil fertility, and destroying useful microorganisms and friendly insects. Treatment of endophytic bacteria with plant growth-enhancing characters is a promising option for increasing crop growth and crop yield and reducing chemical fertilizers (Bhattacharyya *et al.*, 2012). Endophytic bacteria are ecofriendly and safe to

environment (Batra *et al.*, 2018).

Endophytic biology is a promising discipline for the practical application of useful microbiota to assist crop production and control plant diseases (Maiyappan *et al.*, 2020). Usually, plants have been considered single organisms, but plants are associated with various types of microorganisms in nature. The interaction between the plants and symbiotic organisms is likely to have more importance for the health of the plant and disease resistance. Plant growth includes the synergistic activity of numerous diverse life forms in a highly multifaceted environment. The area of the growing plant is a composite of microbial activity, consisting of soil micro-

biota, plant surface microbiota, atmospheric microbiota and inter and intra-cellular colonizing microbes or endophytes (Batra *et al.*, 2018).

Endophytes gaining momentum in biotechnological and agricultural industries due to their capability to produce various secondary active metabolites that can act as antimicrobial, antitumor and immunosuppressive agents (Gouda *et al.*, 2016; Yadav., 2018). A great number of bioactive substances synthesized by endophytic bacteria are economically valuable for humans. Endophytic bacteria also play an important role in nutrient recycling, biodegradation and bioremediation (Nair, 2014).

Endophytes bacteria are a chemical reservoir for novel substances like anticancer, antioxidant, antiviral, antiparasitic, immunomodulatory, insecticidal etc., which can be employed. Bacteria are the most prevalent group of organisms found in entire plant microflora compared to the other four major groups: Actinomycetes, Algae, Fungi and Protozoa. The vast diversity of endophytic bacteria harbors different plant species. The predominant genera are *Enterobacter*, *Bacillus*, *Burkholderia*, *Azospirillum* and *Pseudomonas* (Lodewyckx *et al.*, 2002; Weyens *et al.*, 2009).

Medicinal plants are a valuable source of many pharmacological effective compounds with wide applications in food, medicine and cosmetic fields. Although medicinal plants harbor a rich diversity of microbiota which had the potential to produce plant growth promoting substances (Phurailatpam L. *et al.*, 2022).

Bioprospecting of halophytes has been rarely undertaken, which has numerous benefits in the fields of agriculture and pharmacy. *Heliotropium curassavicum* is a halophyte that can withstand numerous abiotic features like droughts, salinity and elevated heat (Poithirajet *et al.*, 2021). *H. curassavicum* L. is a seashore salt heliotrope belonging to the Borage family (Boraginaceae). The plant is occasionally employed as a supply of food and medicine. Leaves of *H. curassavicum* can be used raw in salads or cooked as potherb, dried leaves can be used as a tea and ashes of the plant are utilized as a salt substitute (Tropical Plants Database) tropical.theferns.info/viewtropical.php?id=Heliotropium+curassavicum). It makes an ideal plant to investigate its potential for endophytic bacteria with unique characteristics like plant growth promotion, phosphate solubilization and synthesizing extracellular enzymes. Hence, the present investigation was undertaken to explore this unexplored halophytic species to screen endophytic bacteria.

MATERIALS AND METHODS

Study area and sample collection

This present investigation was conducted during 2020-21 and 2021-22 at Botany Department, Andhra Univer-

sity, Visakhapatnam, Andhra Pradesh. Plant tissue samples were collected from aerial and root parts of *H. curassavicum* L. located at holophytic zone of Khaspanaupada of Srikakulam district within a latitude of 18°34'25.6"N and longitude of 84°18'50.4"E (18.573986, 84.313925) (Fig. 2). The plant tissue samples were sealed in sterile plastic bag and transported to laboratory for further studies.

Surface sterilization and sterility check

Plant tissue samples were washed with saline water, dried and soaked in ethanol (70%) and kept for one minute. This samples was washed thrice in saline water and soaked in 1% sodium hypochlorite for about five minutes. The entire procedure was repeated twice. Sterility check was also carried out, as outlined by Gyaneshwar *et al.* (2001).

Segregation of endophytic organisms from leaves and roots of halophytic *Heliotropium curassavicum* L.

Subsequent to surface sterilization 1g of each tissue sample was macerated into paste with 0.9% Saline water and subjected to the serial dilution method. The highest diluted solution (4th and 5th dilution) were spread (100µl) on Nutrient Agar plates enriched with 3% NaCl and incubated for 48 hours. After incubation, distinct colonies were purified by the quaternary streak method on Nutrient agar supplemented with 3% NaCl and properly labelled with the codes assigned. Individual pure cultures obtained were treated to further studies (Gyaneshwar *et al.*, 2001).

Preservation of isolated endophytes

All the endophytic bacteria were grown on nutrient agar and kept for overnight incubation. Young cultures at mid-log phase was taken in 20% (w/v) glycerol and preserved in deep freeze at -20°C. The feasibility of the organisms was checked periodically by retrieving the culture on nutrient agar every two to three months.

Phenotypic description of the endophytic bacterial isolates

Pure cultures were placed on nutrient agar enriched with 3% NaCl and the cell and colony morphology were recorded as per the outline provided by Hawksworth *et al.* (1983). Pure cultures at logarithm growth stage were microscopically observed as per the method outlined in Aneja (2006). Gram staining was done as stated in Hucker's method, 1923.

Effect of salt concentration on growth

All the isolated colonies were grown on nutrient agar with varied salt concentrations like 3%, 6%, 8% and 10% Sodium chloride. Observations on growth were recorded every 24 and 48 hours of incubation.

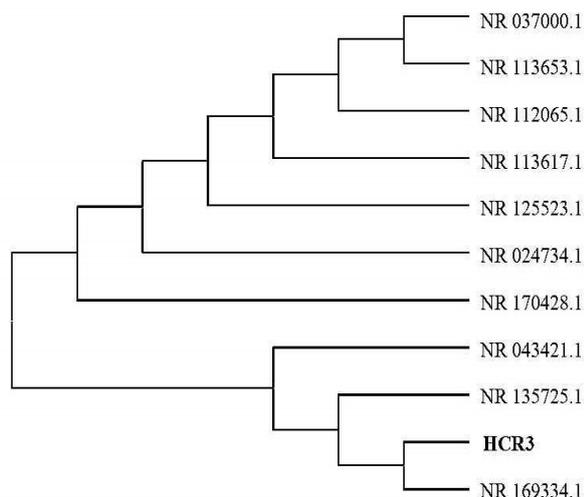


Fig. 1. Phylogenetic Tree of 16S ribosomal RNA sequence of HCR3 strain compared with those Maximum similar entries of type strains from NCBI nucleotide database. The tree was constructed by Maximum likelihood method and Tamura-Nei mode

Biochemical description of endophytic bacterial strains

The young cultures were subjected to the KOH string, catalase, and oxidase tests following the methods described by Aneja (2006). The IMViC test was conducted according to the procedure outlined by Seeley (1981).

Aminoacid decarboxylation test

The amino acid decarboxylation test was employed to

determine the capability of the organisms to produce the carboxylase enzyme, which split the carboxyl group of amino acids into amine and carbon dioxide. Arginine, Lysine, and Ornithine were the commonly used amino acids for this test. The test was performed by inoculating the bacteria culture into tubes containing decarboxylase medium, adding 0.5% respective amino acids and kept for incubation. Bacteria that produced the decarboxylase enzyme acted on the amino acids and produced alkaline products, increasing pH. Bromocresol Purple detected the increased pH. A yellow color indicated a negative reaction, while purple coloration of the media indicated a positive test.

Properties of isolated endophytic organisms

The isolated endophytic organisms was tested for their different properties, including the production of enzymes like protease and amylase, as well as their plant growth-enhancing characteristics such as phosphate solubilizing ability and the synthesis of the phytohormone IAA (Indole 3-acetic acid).

Protease and amylase production

Protease and amylase production was screened using gelatin and starch hydrolysis assays. The organisms were grown 1% starch and 1% gelatin nutrient agar plates to assess their ability to produce amylase and protease enzymes, respectively. After incubating for 48 hours, the starch plates were flooded with diluted iodine solution, and gelatin plates were saturated with ammonium sulphate reagent (approximately 5g per 10ml). The formation of halo zones was observed and recorded.

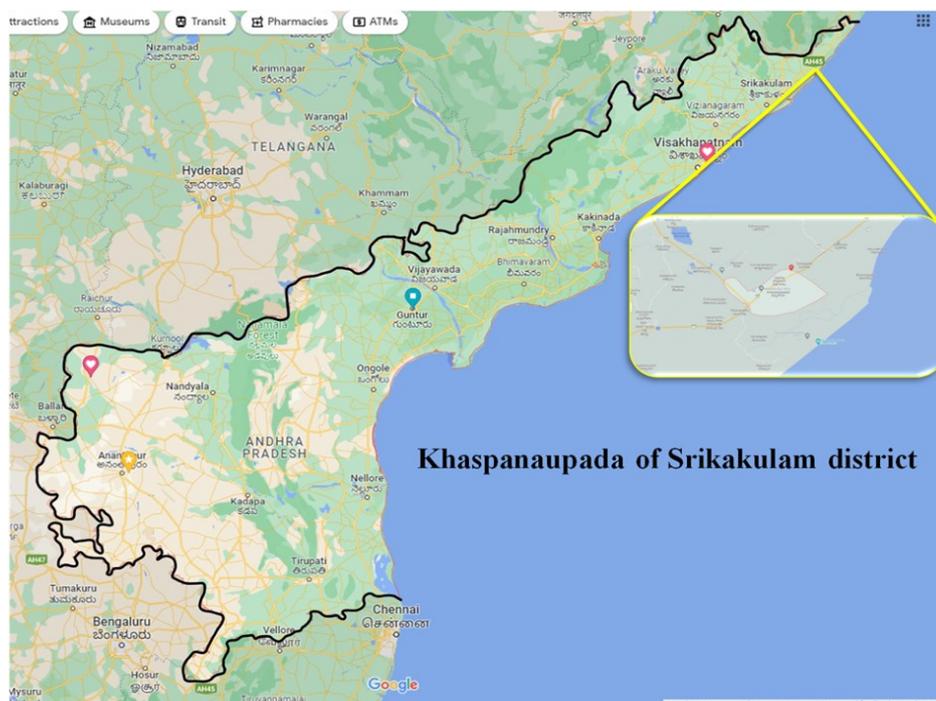


Fig. 2. Map showing the area under study, highlighted in insert area (Khaspanapadu of Srikakulam district)

Table 1. Naming of isolated endophytic bacterial cultures

Plant	Parts of the plant	Isolates*
<i>Heliotropium</i>	Aerial parts (6 isolates)	HCA1, HCA2, HCA3, HCA4, HCA5 & HCA6
<i>curassavicum L.</i>	Root (7 isolates)	HCR1, HCR2, HCR3, HCR4, HCR5, HCR6 & HCR7

*H-*Heliotropium*, C-*curassavicum*, A-Aerial and R-Roots

Production of plant growth promoting characters

IAA production

All the organisms were screened for IAA production using colorimetric assay with Salkowski reagent (1 ml of 0.5M Ferric chloride in 4.9 ml of 35% per chloric acid). Young cultures were inoculated in nutrient broth and incubated for 48 hours. After incubation, the cultures were centrifuged at 2,000 rpm for 20 min. Then, 1.5 ml of the clear solution was mixed with 1.5 ml of Salkowski's reagent. Cultures positive for IAA production exhibited a range of colour from orange to dark purple. The colour development was measured using a spectrophotometer at 536nm.

Phosphate solubilizing ability

All the organisms were screened for their phosphate solubilization ability using a colorimetric assay with Pikovskaya broth. The cultures were grown in Pikovskaya broth for 48 hours, followed by centrifugation at 2,000 rpm for 20 minutes. After centrifugation, 50µl of clear solution was mixed with 3.95 ml of distilled water to make up to 4 ml. To 4 ml of sample 750 µl of phosphate reagent and incubated for 5 min. development of blue colour with different intensities was measured using Spectrophotometer at 680 nm.

Molecular identification using 16S ribosomal

RNA gene method

Nucleic acid was extracted from the bacterial sample, and its purity was accessed using 1.0% agarose gel electrophoresis. The 16S ribosomal RNA gene sequence fragments were amplified using 16SrRNA-Forward and 16SrRNA-Reverse primers. The PCR product, a 1500 base pairs template, was purified. A consensus genome of the 16S ribosomal RNA gene was generated by aligning the forward and reverse sequence data using Aligner Software. The gene sequencing of the 16S rRNA was performed by conducting a BLAST search with the 'nr' database of NCBI GenBank. The top ten gene sequences with the highest identity scores were selected and aligned using the Multiple Alignment software tool Clustal W. A phylogenetic tree and Distance matrix was constructed using MEGA10 (Kumar *et al.*, 2018).

RESULTS

Detailing of isolated endophytic bacteria from

Heliotropium curassavicum L.

Isolated endophytic bacteria from *H.curassavicum L.*

were obtained by surface sterilizing both aerial and root samples. The samples were then cut into pieces and serially diluted. Bacterial cultures were grown on nutrient agar Petri plates from the highest dilutions. Individual colonies were extracted and streaked on nutrient agar plates using the quaternary streak method to obtain pure cultures. Colonies with distinct morphological characteristics were selected for further analysis. A total of 13 isolates were obtained from both aerial and root samples and labelled accordingly.

General morphology

Phenotypic, biochemical, and molecular characterization were performed on these isolates. In terms of general morphology, the majority of the tested isolates were Gram-positive (85%), except for HCA6 and HCR7, which were Gram-negative. The cell morphology varied among strains, with most isolates appearing as rod-shaped cells in diplobacilli and chain formations. The length of the cells ranged from small rods to medium and long rods. All tested isolates were able to grow at temperatures up to 42°C. Approximately 54% of the strains were motile, while the remaining 46% were non-motile.

Growth at various salt concentrations

The growth of the isolates at various salt concentrations were evaluated. Most of the isolates (77%) grew at 0% NaCl concentration, except for HCR7. All tested isolates exhibited tolerance to salinity up to 8% and 10% NaCl concentrations, except for HCR5 and HCR7, which were able to grow up to 6% NaCl concentration.

Biochemical features of isolated endophytic bacterial isolates

Biochemical tests were conducted on all the isolates to determine their characteristics. The results showed that all the tested endophytic strains exhibited a positive reaction for the catalase when 3% H₂O₂ was added. The isolated cultures were also subjected to other biochemical tests, including the String test, oxidase, IMViC test, and amino acid decarboxylase test. The recorded observations for these tests are detailed in Table 4.

Protease production by Gelatin hydrolysis test

Protease production was assessed using the gelatin hydrolysis test. All isolates were plated on gelatin medium, and after 48 hours of incubation, the plates were saturated with ammonium sulfate for 30 minutes. The presence of a clear zone indicated the hydrolysis of gelatin varied among strains, ranging from 0.9 cm

Table 2. Property study of tested isolates from *Heliotropium curassavicum* L.

Plant	Part	Code	Plant Growth Hormone		Enzyme production	
			IAA ($\mu\text{g/ml}$)	PSB (ppm)	Amylase production	Protease production
<i>Heliotropium curassavicum</i> L.	Aerial parts	HCA1	-	3.0	-	+ (3.5)
		HCA2	-	1.2	+ (1.5)*	+ (3.0)
		HCA3	-	2.3	+ (3.5)	-
		HCA4	-	2.2	-	+ (4.0)
		HCA5	-	3.0	+ (2.5)	+ (2.0)
		HCA6	-	1.8	+ (1.3)	-
	Root	HCR1	-	<0.5	+ (2.0)	-
		HCR2	-	<0.5	+ (2.2)	-
		HCR3	37.0	3.5	-	-
		HCR4	<10	3.3	-	+ (3.4)
		HCR5	<10	3.7	-	+ (4.0)
		HCR6	-	4.0	+ (1.5)	+ (2.5)
		HCR7	20.0	3.7	+ (0.9)	+ (2.5)

*Figures in the parenthesis indicate the diameter of the clear zone in centimetres

(HCR7) to 3.5 cm (HCA3).

Amylase production by Starch hydrolysis test

Two days old inoculated plates with tested organisms were subject to starch test by flooding with diluted iodine solution. The starch present in the agar plates reacts with the iodine solution, forming blue colour in the plates. The positive test indicates the formation of a clear zone around the inoculated cultures. Starch hydrolysis by tested isolates showed varied results ranging from 2.0 cm (HCA5) to 4.0 cm (HCA4 and HCR5) in diameter.

Synthesis of plant growth enhancing hormone IAA (Indole-3-acetic acid)

The plant growth-enhancing hormone IAA (Indole-3-acetic acid) synthesis was examined by colorimetric analysis using Salkowski's reagent. The isolated cultures were inoculated in nutrient broth and incubated for two days. After incubation, the cultures were centrifuged and the clear solution was collected. To this, 1.5 ml of Salkowski's reagent was added. Colour and intensity changes were observed and measured using a UV Spectrophotometer at 536nm. All the tested isolates showed varied results in IAA production. The highest IAA production was recorded with HCA3 (37.0 $\mu\text{g/ml}$) followed by HCR3 (20.0 $\mu\text{g/ml}$) and HCR7 (20.0 $\mu\text{g/ml}$) as detailed in Table 2.

Phosphate solubilizing ability

Phosphate solubilizing ability was accessed by culturing the isolates in 4 ml of 3% Pikovskaya's broth and incubating them for 2 days. After the incubation period, 750 μl of phosphate reagent was added to 4 ml of the sample, and the absorbance was measured using a UV Spectrophotometer at a wavelength of 680 nm. All the tested isolates exhibited phosphate solubilizing ability. The highest phosphate solubilizing ability was recorded

with HCR6 (4.0 ppm), followed by HCR3 and HCR5 (3.7 ppm) and the lowest was recorded with HCR1 and HCR2 (<0.5 ppm) (Table 2).

Classification of endophytic strains by using 16S ribosomal RNA molecular method

Because of its ability to produce higher amount of IAA when evaluated with other isolated strains, the HCR3 strain was selected for further studies.

The entire DNA was obtained from the cultures of HCR3 (NCBI accession No. NR_169334.1) and its concentration was estimated using a 1% agarose gel to obtain single high molecular weight DNA band. The 16S ribosomal RNA gene fragments were amplified using 16S ribosomal RNA-Forward and 16S ribosomal RNA-Reverse primers using BDT version 3.1 Cycle sequencing kit on ABI 3730 X1 Genetic Analyzer. BLAST analysis of the 16S ribosomal RNA gene sequence of the HCR3 strain against the NCBI database revealed a higher relationship with *Pseudomonas khazarica*. The 16S ribosomal RNA sequence identity between the endophytic isolate HCR3 and *P.khazarica* was 100 %. The phylogenetic tree, constructed using the neighbour-joining algorithm, also confirmed that the HCR3 isolate and *P. khazarica* formed a distinct group separate from other related species.

DISCUSSION

The current research work is first to investigate the interactions between endophytes and the halophytic plant *H. curassavicum* L. Limited research findings are available on this subject, particularly regarding the interactions between endophyte bacteria and the host plant. However, more research has been conducted on the interaction between fungi and *H.curassavicum* L. Therefore, the present discussion focuses on endophytic bacteria, fungi, and the host plant

Table 3. Cultural and biochemical characters of tested isolates from *Heliotropium curassavicum* L.

	HCA1	HCA2	HCA3	HCA4	HCA5	HCA6	HCR1	HCR2	HCR3	HCR4	HCR5	HCR6	HCR7
Microscopic observation	Rods occur in two cells	Small rods	Small rods	Small rods occur in two cells	Large rods occur in chains of 2-4	Small rods occur in two	Small rods	Large rods	Small rods	Small rods occur in two cells	Small rods	Large rods occur in chains	Pleomorphism
Gram's staining	+	+	+	+	+	+	+	+	-	+	+	+	-
Endospore	+	+	+	+	+	+	+	+	-	+	+	+	-
Shape /position	Oval/centre of the cell	Oval/centre of the cell	Oval/centre of the cell	Oval/Free cell	Oval/centre of the cell	Oval/centre of the cell	Oval/Free cells	Oval/centre of the cell	-	Oval/centre of the cell	Oval/centre of the cell	Oval/centre of the cell	-
Motility	-	+	-	+	-	-	++	-	++	++	-	-	++
KOH	-	-	-	-	-	-	-	-	+	-	-	-	+
Catalase	+	+	+	+	+	+	+	+	+	+	+	+	+
Oxidase	-	-	-	-	-	-	-	-	++	-	-	-	+
Growth at 42°C	+	+	+	+	+	+	+	+	+	+	+	+	+
Starch hydrolysis	+	+(1.5)*	+(3.5)	-	+(2.5)	+(1.3)	+(2.0)	+(2.2)	-	-	-	+(1.5)	+(0.9)
Gelatin hydrolysis	+(3.5)	+(3.0)	-	+(4.0)	+(2.0)	-	-	-	-	+(3.4)	+(4.0)	+(2.5)	+(2.5)
Growth on 0% NaCl	++	++	++	++	++	++	++	++	-	++	++	++	-
Growth on 3% NaCl	++	++	++	++	++	++	++	++	++	++	++	++	++
Growth on 6% NaCl	+	+	+	+	-	+	+	+	+	+	+	+	+
Growth on 8% NaCl	+	+	+	+	-	+	+	+	+	+	+	+	-
Growth on 10% NaCl	++	++	++	++	++	-	-	+	+	+	++	++	-
Hemolysis	No colour change	No colour change	Green	Green	No colour change	Green	No colour change	Green	No colour change	No colour change	No colour change	No colour change	No colour change
Bacillus selective media (HiMedia)	-	-	Green	Green	-	Green	-	-	-	-	-	-	-
Growth on TCBS	-	-	+Red colour colony	-	-	-	-	-	-	-	-	-	Green
Growth on Congo Red	-	-	+Red colour colony	+Red colour colony	-	+ colourless	+ colourless	+ colourless	+ colourless	+ colourless	+ colourless	+ colourless	+ colourless
Indole test	-	-	+	-	-	-	-	-	+	-	-	-	+
Methyl Red test	+	-	+	+	+	-	-	+	-	+	+	+	-
Voges Proskauer test	-	-	-	-	-	-	-	-	-	-	-	-	-
Citrate test	-	-	-	-	-	-	-	-	-	-	-	-	-
Lysine decarboxylase	-	-	-	-	-	+	+	+	-	-	-	-	+
Arginine decarboxylase	-	-	-	-	++	++	++	+	+	+	++	+	+
Ornithine decarboxylase	-	-	-	+	+	+	+	-	+	+	+	-	+
Preliminary identification	Bacillus sp.	Bacillus sp.	Bacillus sp.	Bacillus sp.	Bacillus sp.	Bacillus sp.	Bacillus sp.	Bacillus sp.	<i>Pseudo-</i> <i>monas</i> <i>khazarica</i>	Bacillus sp.	Bacillus sp.	Bacillus sp.	<i>Vibrio</i> sp

*+ indicates normal growth, ++ indicates luxuriant growth and '-' indicates No growth; Results obtained are the Mean of three replication; *Figures in parenthesis indicates the halo zones measured in cm around the colony and the mean of three replications.

H. curassavicum L.

In the present, salt tolerance, and plant growth promoting bacteria belonging to the genera *Bacillus*, *Pseudomonas* and *Vibrio* were identified from *H. curassavicum* L. This finding is consistent with a study conducted by Alishahi *et al.* (2020), which also reported the presence of the *Microbacterium* and *Pseudomonas* genera in high abundance in samples from halophytic plants.

Tian and Zhang (2017) have also reported the widespread distribution of *Microbacterium* and *Pseudomonas* genera in various halophytic plants. Moreover, previous reports have highlighted the plant growth-promoting abilities of species belonging to the *Mycobacterium* and *Pseudomonas* genera, including the production of IAA, phosphate solubilization and ACC (1-aminocyclopropane-1-carboxylate) deaminase activity (Alishahiet *al.*, 2013; Madhaiyanet *al.*, 2010).

Approximately 85% of the isolated strains from *H. curassavicum* L. belonged to the *Bacillus* family. The findings is consistent with study conducted Gaoet *al.* (2021), where *Bacillus* species accounted for 38.5% of the total number of endophytic bacterial strains isolated from different halophytic plant species. The abundance of *Bacillus* species can be attributed to their ability to form spores, which allows them to withstand harsh environmental condition.

The study's results revealed that most of the isolates (85%) from *H. curassavicum* L. could tolerate a salinity level of 10% NaCl. These findings are in line with the study conducted by Sanjay *et al.*, (2014), where 85% of the endophytic bacteria strains obtained from halophytic plant species showed growth at 7.5 per cent NaCl, and 75% were able to tolerate up to 10 per cent salt concentration.

Similar, ALKahtani *et al.* (2020) reported that endophytic bacterial cultures isolated from medicinal plants *Achillea fragrantissima* (Forssk) and *Fagoniamollis* Delile exhibited the ability to solubilize phosphate with varying halo zone sizes. Additionally, all the isolates in the current study showed the production of Indole-3-acetic acid (IAA), which is consistent with the findings of Hassan *et al.* (2017), who reported diverse capabilities for phosphate solubilization, antimicrobial activity, and IAA synthesis among isolated endophytes.

Pseudomonas khazarica, a novel species first identified by Tarhriz *et al.* (2020) is known for its ability to degrade polycyclic aromatic hydrocarbon. In another study conducted by Phurailatpam *et al.* (2022) isolated 67 bacterial endophytes collected from the ethnomedicinal plant *Piper longum* L. Their results showed that inoculation of *P. khazarica* S34 was recorded to promote seed germination and seed growth in wheat and tomato through the production of indole-3-acetic acid. These findings aligned with the results of the present study.

Conclusion

This study is the first to identify and report the abundance of salt-tolerant, plant growth promoting bacteria of the genera *Bacillus*, *Pseudomonas*, and *Vibrio* isolated from *H. curassavicum* L. These bacterial strains have demonstrated potential for enhancing plant growth through various mechanisms such as IAA production, phosphate solubilization, and the production of various enzymes. The findings contribute to our understanding of the plant-microbe interactions in halophytic ecosystems and highlight the potential applications of these bacteria in promoting plant growth under saline conditions. Further research in this area is warranted to explore the full potential of these endophytes and their mechanisms of action for sustainable agriculture and environmental remediation.

Conflict of interest

The authors declare that they have no conflict of interest.

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