Research Article

Plant Growth Promoting of Endophytic Sporosarcina aquimarina SjAM16103 Isolated from the Pneumatophores of Avicennia marina L.

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Endophytic Sporosarcina aquimarina SjAM16103 was isolated from the inner tissues of pneumatophores of mangrove plant Avicennia marina along with Bacillus sp. and Enterobacter sp. Endophytic S. aquimarina SjAM16103 was Gram variable, and motile bacterium measured 0.6–0.9 μm wide by 1.7–2.0 μm long and light orange-brown coloured in 3-day cultures on tryptone broth at 26°C. Nucleotide sequence of this strain has been deposited in the GenBank under accession number GU930359. This endophytic bacterium produced 2.37 μMol/mL of indole acetic acid and siderophore as its metabolites. This strain could solubilize phosphate molecules and fixes atmospheric nitrogen. Endophytic S. aquimarina SjAM16103 was inoculated into four different plants under in vitro method to analyse its growth-promoting activity and role inside the host plants. The growth of endophytic S. aquimarina SjAM16103 inoculated explants were highly significant than the uninoculated control explants. Root hairs and early root development were observed in the endophytic S. aquimarina SjAM16103 inoculated explants.

1. Introduction

The genus Sporosarcina, which belongs to the family Bacillaceae, was created by Kluyver and van Neil [1] to accommodate bacteria that have spherical or oval-shaped cells, low DNA G+C content (40–42 mol%), and MK-7 as the major menaquinone. Sporosarcina species can be differentiated from other members of the Bacillaceae, by their coccoid or rod-shaped cells, motility, sporulation, and possession of MK-7 as the major menaquinone and A4α as the peptidoglycan variant. The genus Sporosarcina originally comprised two species, S. ureae and S. halophila [2, 3]. However, three species of the genus Bacillus, namely, Bacillus globisporus [4], B. psychrophilus [5], and B. pasteurii [6], which belonged to rRNA group 2 [7] and contain L-lysine in their cell wall, have recently been transferred to the genus Sporosarcina [8] as S. globispora, S. psychrophila, and S. pasteurii also identified a novel species, S. aquimarina isolated from seawater. However, there were no published reports on endophytic S. aquimarina isolated from the living cells. Therefore, the present study was aimed to investigate the role and effects of endophytic S. aquimarina isolated from the pneumatophores of A. marina.

Endophytic bacteria are living inside the plant tissue without eliciting symptoms of disease, common to a large number of plant species. Endophytic bacteria can promote the plant growth and yield and can act as a biocontrol agent [9]. In recent years, much attention has been paid to natural methods of crop growing in expectation of moving toward agriculturally and environmentally sustainable development [10]. Endophytic bacteria promote plant growth due to their abilities in nitrogen fixation [11], phytohormone production [12], solubilization of phosphorus [13], and disease control [14, 15].

Endophytic microbial inocula, primarily bacteria, are used as propagule priming agents, both as in vitro cocultures and transplanting [16]. It is an emerging trend in biotechnological approach aimed at reducing chemical...
input in plant production, while increasing plant fitness,
productivity, and resistance to diseases in the context of
sustainable horticulture.

In the present study, endophytic bacteria were isolated
from the surface sterilized pneumatophores of A. marina.
The isolates were identified phenotypically and genotyp-
ically. The isolate S. aquimarina was taken for further
investigation. This endophytic bacterium was inoculated
into four different plants, two fresh water plants (Bacopa
monnieri and Eupatorium triplinerve) and two mangrove
plants (Excoecaria agallocha and A. marina) to analyse its
growth-promoting efficacy and role as endophyte. These
plants were selected based on their needs in the society,
because they have high medicinal and economical values.
Medicinally, they are used for curing skin diseases, even
for leprosy [17], HIV [18], fungal diseases [19], mental disorders
[20, 21], and economically as fire wood, match boxes, paper
pulp [17], and used as fodder in India and in Australia.

2. Materials and Methods

2.1. Isolation of Strains and Growth Condition. Endophytic
bacterium was isolated from the inner tissues of healthy
pneumatophores. Parts of pneumatophores about 1 cm
diameter were sterilized with 70% ethanol and 0.1%
mercuric chloride [22]. Sterilized parts were excised with a
sterile scalpel blade. Slices (0.1 cm thickness) were placed on
nutrient agar plates and incubated at 26°C for 48 h. Bacterial
growth associated with the pneumatophore sections was
purified by repeated plating on nutrient agar, and cultures
were maintained as spore suspensions by freezing in 20% (V/V)
glycerol.

2.2. Phenotypic Characterization. The isolate was Gram-
stained and examined microscopically for its morphological
characteristics. Some set of physiological characteristics
include acid production from sugars (TSI), sodium citrate
utilization, urease production, starch, gelatin hydrolysis, and
voges-proskauer reaction was carried out using standard
protocols described by Gordon et al., [23]. Casein hydrolysis
was detected after 3 days of incubation on nutrient agar
medium containing precipitated tricalcium phosphate. The medium
was a modification of Pikovskaya medium [28]. Reduc-
tion of sulphur by the bacterium was tested qualitatively using
Burk’s N-free medium [28].

2.3. 16S rRNA Gene Sequence Analysis. Genomic DNA was
isolated from pure culture [24]. A large fragment (800–
1100 bp) of 16S rRNA was amplified by PCR using primers
5’-TGA GGA AGA TAA TGA CGG-3’ and 5’-CCT CTA TCC
TCT TTG CAA CC-3’. The 50 μL PCR reaction mixtures
contained 100 ng of DNA extract, 1 × Taq reaction buffer,
20 pmol of primers, 200 μM dNTPs, and 1.5 U of Taq DNA
polymerase (Promega). The thermocycling conditions con-
stituted of an initial denaturation at 94°C for 3 mins, 30 ampli-
fication cycles of 94°C for 1 min (denaturation), 57°C for 1
minute (annealing), 72°C for 2 mins (extension), and final
polymerization at 72°C for 4 mins. PCR product was purified
and sequenced. Searches in the GenBank/EMBL/DDBJ/PDB
data libraries were performed using BLAST (blastn) search
algorithm [25] in order to establish the identity of the isolate.

2.4. Determination of Cellular Fatty Acid Composition. Cel-

cular fatty acids composition of endophytic bacterium was
analyzed using the Sherlock system (MiDi Company, USA)
and according to the manufacturer’s instructions.

2.5. Plant Growth Promoting Activities

2.5.1. IAA Production. Indoleacetic acid (IAA) produced by
bacterium was assayed colorimetrically using FeCl3-
HClO4 [26]. The bacteria were grown in modified nutrient
broth M26 for 24 hours on a gyratory shaker (150 rpm) at
room temperature as seed culture. The medium contained
(in 1000 mL distilled water) 5 g NaCl, 10 g peptone, and
10 g of beef extract. After overnight incubation, 100 μL
of culture was inoculated to 10 mL minimal salt (MS)
medium amended with 5 mM L tryptophan [27] and grown
again for 48 hours on the shaker. The MS medium con-
tained (in 1000 mL distilled water) 1.36 g KH2PO4, 2.13 g
Na2HPO4, 0.2 g MgSO4·7H2O, and trace elements. The pH
of MS medium was adjusted to 7.0 before autoclaving. L-
Tryptophan solution was prepared as stock solution contain-
ing (in 100 mL distilled water) 10 g glucose, 1 g glucose, 1 g
L-Tryptophan, and 0.1 g yeast extract. The stock solution was
filtered through a sterile 0.2 μm membrane filter (Millipore).
1.5 mL bacterial broth culture was centrifuged at 12,000 rpm
for 5 mins. One millimeter of the supernatant was added to
2 mL FeCl3-HClO4 reagent. After 25 mins (after color
density reaches its maximum), the mixture was read in UV
spectrophotometer at 530 nm absorbance. The amount of
IAA produced per milliliter of culture was estimated using a
standard curve. The number of bacterial population in
the culture expressed in colony forming unit (CFU) was
estimated by the Miles and Misra drop plate method.

2.5.2. Phosphate Solubilization. Phosphate solubilization test
was conducted qualitatively by plating the bacterium in agar
containing precipitated tricalcium phosphate. The medium
was a modification of Pikovskaya medium [28].

2.5.3. Nitrogen Fixation. Fixation of atmospheric nitrogen
by the bacterium was tested qualitatively using Burk’s N-free
medium [28].

2.5.4. Sulphur Reduction. Reduction of sulphur by the bac-
terium was tested qualitatively by sulphate API agar [28].

2.5.5. Siderophore Production. Siderophore production was
tested qualitatively using chrome azurol S (CAS) agar as
described by Alexander and Zuberer [29]. The bacterial
culture was spread on the CAS agar plates with three repli-
cations. Orange halos around the colonies after overnight
incubation indicated siderophore production.
Table 1: Phenotypic characteristics of *Sporosarcina aquimarina* SjAM16103.

<table>
<thead>
<tr>
<th>Morphological characteristics</th>
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<tbody>
<tr>
<td>Shape</td>
<td>Bacilli (with flagellum)</td>
</tr>
<tr>
<td>Motility</td>
<td>Motile</td>
</tr>
<tr>
<td>Gram’s staining</td>
<td>Gram variable</td>
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<table>
<thead>
<tr>
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<tr>
<td>Temperature</td>
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<tr>
<td>Min 2–10°C</td>
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</tr>
<tr>
<td>Max 26–50°C</td>
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<tr>
<td>pH</td>
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<td>4.5</td>
<td>Negative</td>
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<td>9.0</td>
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<td>Salinity</td>
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<td>2%</td>
<td>Positive</td>
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<td>3%</td>
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<td>5%</td>
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<tr>
<td>7%</td>
<td>Positive</td>
</tr>
<tr>
<td>9%</td>
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Exoenzyme activities

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<thead>
<tr>
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<tbody>
<tr>
<td>Starch hydrolysis</td>
<td>Negative</td>
</tr>
<tr>
<td>Gelatin hydrolysis</td>
<td>Positive</td>
</tr>
<tr>
<td>Casein hydrolysis</td>
<td>Negative</td>
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Endoenzyme activities

<p>| | |</p>
<table>
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<tbody>
<tr>
<td>Catalase production</td>
<td>Positive</td>
</tr>
<tr>
<td>Urease production</td>
<td>Positive</td>
</tr>
<tr>
<td>Oxidase production</td>
<td>Positive</td>
</tr>
<tr>
<td>Voges-proskauer</td>
<td>Negative</td>
</tr>
</tbody>
</table>

2.6. Inoculation of Explants with Endophytic Bacterium. Nodal segments (length: 0.5 cm) of four different plants (*Bacopa monnieri*, *Eupatorium triplinerve*, *Excoecaria agallocha*, and *Avicennia marina*) were disinfected by sonicating in water for 20 min and dipping in 70% ethanol for 1 min, followed by 25 min of sodium hypochlorite (25%)/Tween 80 (0.01%) solution and rinsed three times with distillled and sterile water [30].

The sterile explants of *Bacopa monnieri* and *Eupatorium triplinerve* were cultured in a hormone-free Murashige-Skoog (MS) medium [31] with the addition of 200 μL of endophytic *Sporosarcina aquimarina* SjAM16103. The sterile explants of *Excoecaria agallocha* and *Avicennia marina* were cultured in a hormone-free X medium (M S Swaminathan Research Foundation, India) with the addition of 200 μL of endophytic *Sporosarcina aquimarina* SjAM16103. Then, these explants were cultured under a photoperiod of 16 h of light and 8 h of dark under an irradiance of 52 mmol m⁻² seg⁻¹. The explants without endophytic *Sporosarcina aquimarina* SjAM16103 were marked as control explants.

2.7. Statistical Analysis. The whole experiment was set up in the randomized design with 10 replicas. All the data collected from these experiments were subjected to an analysis of variance (ANOVA) using SPSS statistical tool. The significant level (*P > 0.05*) was evaluated between various growth parameters (shoot length, number of roots, and root length).

3. Results

3.1. Morphology. During this study, 13 bacterial strains were isolated from the pneumatophores of *A. marina* L. Among them, four strains (SjAM16101, SjAM16102, SjAM16103, and SjAM16104) were genotypically analysed as *Bacillus* sp., *Enterobacter* sp., *Sporosarcina aquimarina*, and *Bacillus cereus*, respectively. Strain SjAM16103 was Gram variable, and motile bacterium measured 0.6–0.9 μm wide by 1.7–2.0 μm long (Figure 1) and light orange-brown coloured in 3-day cultures on tryptone broth at 26°C.

3.2. Phenotypic Characteristics. Phenotypic characteristics of strain SjAM16103 are given in Table 1. The optimal growth temperature was 32°C. Strain SjAM16103 grew at 26–50°C but not at 2–10°C. The optimal pH for growth was 7.0, and growth was inhibited at pH values below 5.0. Strain SjAM16103 grew in the presence of 2–9% NaCl. Gelatin was hydrolysed and showed catalase, urease, and oxidase activities. Acid was produced in the triple sugar iron test. Fatty acids compositions of strain SjAM16103 are given in Table 3. Gas chromatographic methyl ester profiles of strain SjAM16103 are given in Figure 2.

3.3. 16S rRNA Gene Sequences. The 16S rRNA of strain SjAM16103 was directly sequenced following PCR amplification, and its partial nucleotide sequence was determined. The 16S rRNA sequence of strain SjAM16103 was 971 bp long (Table 2) and was identified as *Sporosarcina aquimarina* with highest similarity value of 98%. The nucleotide sequence of *S. aquimarina* SjAM16103 has been deposited in the GenBank/EMBL/DDBJ/PDB under accession number GU930359.
Table 2: Nucleotide sequence of *Sporosarcina aquimarina* SjAM16103.

<table>
<thead>
<tr>
<th>Nucleotide Sequence</th>
</tr>
</thead>
<tbody>
<tr>
<td>AAGTGGAGGGACGCCTCAGGAGGTAAAGCTACTACCTCTTTCTAAGCTCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGT GACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGGCCGAGGCTGGCACTCCGATGGTGTGACGGGCGGTGTGACAGCC</td>
</tr>
</tbody>
</table>
Figure 3: Inoculation of endophytic S. aquimarina SjAM16103 in B. monnieri (in vitro). (A) S. aquimarina SjAM16103 inoculated explants; (B) control explants.

Figure 4: Inoculation of endophytic S. aquimarina SjAM16103 in B. monnieri.
3.5.4. In *A. marina* Vierh. Shoot growth of *S. aquimarina* SjAM16103 inoculated explants was observed 5 days after incubation and measured 10 days after incubation. Whereas, in control explants, the shoot growth was observed 10 days after incubation and measured 15 days after incubation. Shoot length of *S. aquimarina* SjAM16103 inoculated explants was highly significant ($P > 0.05$) than the control explants (Figure 9). Root growths were observed 20 days after incubation in *S. aquimarina* SjAM16103 inoculated explants (Figure 10). Early leaves were observed in the *S. aquimarina* SjAM16103 inoculated explants. Root length of *S. aquimarina* SjAM16103 inoculated explants was significantly ($P > 0.05$) higher than the root length of control explants (Table 5).

### Table 3: Fatty acid composition of *Sporosarcina aquimarina* SjAM16103.

<table>
<thead>
<tr>
<th>Saturated fatty acids</th>
<th>Unsaturated fatty acids</th>
<th>Branched fatty acids</th>
<th>Summed feature</th>
</tr>
</thead>
<tbody>
<tr>
<td>$C_{10:0}$</td>
<td>$C_{11:0}$ 3OH</td>
<td>$C_{13:0}$ iso</td>
<td>3.15</td>
</tr>
<tr>
<td>$C_{12:0}$</td>
<td>$C_{12:0}$ 2OH</td>
<td>$C_{13:0}$ anteiso</td>
<td>0.14</td>
</tr>
<tr>
<td>$C_{14:0}$</td>
<td>$C_{12:0}$ 3OH</td>
<td>$C_{14:0}$ iso</td>
<td>0.41</td>
</tr>
<tr>
<td>$C_{15:0}$</td>
<td>$C_{15:0}$ 2OH</td>
<td>$C_{15:0}$ iso</td>
<td>0.19</td>
</tr>
<tr>
<td>$C_{16:0}$</td>
<td>$C_{15:0}$ 3OH</td>
<td>$C_{15:0}$ anteiso</td>
<td>2.58</td>
</tr>
<tr>
<td>$C_{17:0}$</td>
<td>$C_{15:1}$ w5</td>
<td>$C_{15:0}$ iso 3OH</td>
<td>0.19</td>
</tr>
<tr>
<td>$C_{18:0}$</td>
<td>$C_{15:1}$ w6</td>
<td>$C_{15:1}$ iso F</td>
<td>0.17</td>
</tr>
<tr>
<td>$C_{19:0}$</td>
<td>$C_{15:1}$ w8</td>
<td>$C_{15:1}$ anteiso A</td>
<td>0.08</td>
</tr>
<tr>
<td></td>
<td>$C_{16:0}$ 3OH</td>
<td>$C_{16:0}$ iso</td>
<td>2.15</td>
</tr>
<tr>
<td></td>
<td>$C_{16:0}$ N alcohol</td>
<td>$C_{16:0}$ iso 3OH</td>
<td>0.06</td>
</tr>
<tr>
<td></td>
<td>$C_{16:1}$ w5</td>
<td>$C_{16:1}$ iso H</td>
<td>0.25</td>
</tr>
<tr>
<td></td>
<td>$C_{16:1}$ w7 alcohol</td>
<td>$C_{17:0}$ iso</td>
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</tr>
<tr>
<td></td>
<td>$C_{16:1}$ w11</td>
<td>$C_{17:0}$ anteiso</td>
<td>0.13</td>
</tr>
<tr>
<td></td>
<td>$C_{17:0}$ 2OH</td>
<td>$C_{17:0}$ cyclo</td>
<td>0.13</td>
</tr>
<tr>
<td></td>
<td>$C_{17:0}$ 3OH</td>
<td>$C_{17:0}$ iso 3OH</td>
<td>0.13</td>
</tr>
<tr>
<td></td>
<td>$C_{17:1}$ w6</td>
<td>$C_{17:1}$ iso w10</td>
<td>0.58</td>
</tr>
<tr>
<td></td>
<td>$C_{17:1}$ w8</td>
<td>$C_{17:1}$ anteiso w9</td>
<td>0.62</td>
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<td></td>
<td>$C_{18:1}$ w9</td>
<td>$C_{18:1}$ iso w10</td>
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<tr>
<td></td>
<td>$C_{20:1}$ w7</td>
<td>$C_{20:1}$ cyclo w8</td>
<td>0.25</td>
</tr>
<tr>
<td></td>
<td>$C_{20:1}$ w9</td>
<td>$C_{20:1}$ cyclo w10</td>
<td>11.543</td>
</tr>
</tbody>
</table>

Unknown fatty acids
4. Discussion

Endophytic bacteria are well known in crop plants [9, 32] but largely have not been investigated in mangrove plants. In the present study, thirteen bacterial strains were isolated from the pneumatophores and four strains (SjAM16101, SjAM16102, SjAM16103, and SjAM16104) were selected for further studies based on their morphology and colonization. The strain SjAM16103 was identified as *Sporosarcina aquimarina* using 16s rRNA and was confirmed using biochemical tests and fatty acids profiling. The results obtained in this research are perfectly coincide with reports on endophytes from other hosts in which generally a large number of species can be isolated from a given host, but only very few species are present in a significant number [33]. *S. aquimarina* had been isolated from seawater in Korea [8]. However, this was the first report on *S. aquimarina* isolated from the inner tissue of the plant.

The strain SjAM16103, *S. aquimarina* could produce siderophore and IAA. Siderophores are iron chelating ligands which can be beneficial to plants by increasing the solubility of ferric iron (Fe III), which otherwise is unavailable for plant nutrition [34]. The production of IAA enhances root growth of the plants by stimulating plant cell elongation or cell division [35]. The colonization of pneumatophores by endophytic *S. aquimarina* SjAM16103 enhances growth of the entire plants.

The results indicated that reintroduction of naturally occurring endophytic bacteria into tissue culture can lead to improve plant growth and yield. The present study revealed that the endophytic *S. aquimarina* SjAM16103 isolated from the pneumatophores of *A. marina* was not host specific. Some endophytic genera, however, exhibit no host specificity and are invariably recovered from plants.
belonging to different groups and growing in different geographical locations [36, 37].

In the present study, another interesting observation was the growth of root hairs developed in the endophytic *S. aquimarina* SjAM16103 inoculated explants, root hairs could fix atmospheric nitrogen [38]. Endophytic N\(_2\)-fixing bacteria seem to constitute only a small proportion of total endophytic bacteria [39, 40], and increasing N\(_2\)-fixing populations in plants has been considered as a possibility to increase nitrogen fixation. This was the first report on *S. aquimarina* isolated from the living tissue and could produce IAA and fixes atmospheric nitrogen.

This research has been directed to find endophytic *S. aquimarina* SjAM16103 that could significantly increase the yields in four different plants (*B. monnieri*, *E. triplinerve*, *E. agallocha*, and *A. marina*) after their inoculation. Growth of *S. aquimarina* SjAM16103 inoculated explants of *B. monnieri*, *E. triplinerve*, *E. agallocha*, and *A. marina* was highly significant than their control explants. The development of leaves in *S. aquimarina* SjAM16103 inoculated explants was much earlier than control explants. Leaves were developed in *S. aquimarina* SjAM16103 inoculated explants of *A. marina* 5 days after incubation, whereas, in control explants, leaves were developed 13 days after incubation. Inoculant seems to be successful in this micropropagated plants, as there were few or no other microorganisms with which to compete. There could be enormous benefits to be gained through the inoculation of microorganisms into soilless mixes in which plants are transplanted at an early stage in their growth. The natural condition of plants seems to be in a close interaction with endophytes.

**Table 4: Plant growth-promoting activities of Sporosarcina aquimarina SjAM16103.**

<table>
<thead>
<tr>
<th>Growth promoting activities</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>IAA production</td>
<td>2.37 ((\mu\text{Mol/mL}))</td>
</tr>
<tr>
<td>P-solubilization</td>
<td>Positive</td>
</tr>
<tr>
<td>S-reduction</td>
<td>Negative</td>
</tr>
<tr>
<td>N-fixation</td>
<td>Positive</td>
</tr>
<tr>
<td>Siderophore production</td>
<td>Positive</td>
</tr>
</tbody>
</table>

IAA: Indole acetic acid production; P: phosphate solubilization; S: sulphur reduction.
Roots were developed earlier in the *S. aquimarina* SjAM16103 inoculated explants than the control explants. *S. aquimarina* SjAM16103 inoculated explants of *E. triplinerve* showed the growth of root hairs, whereas this characteristic feature was absent in the control explants of *E. triplinerve*. Early roots were observed in the *S. aquimarina* SjAM16103 inoculated explants of *E. agallocha*. Endophytic *S. aquimarina* SjAM16103 seems promising to increase crop yields, produced IAA and siderophore, fixed nitrogen, and solubilized phosphate. The distribution and biological activity of this endophytic bacterium deserve to be explored to make full use of their habitation inside the plants. The present study reveals that endophytic *S. aquimarina* SjAM16103 can be used as a biofertilizer, which can subsequently be used by the plant, thereby improving plant growth.

### 5. Conclusion

In the present study, mangrove was chosen because mangrove ecosystems are known for high productivity. At the same time, pneumatophores were chosen for this research due to its mechanisms (anaerobic respiration), which taken up gases directly from the atmosphere and various other nutrients, like iron, from the inhospitable soil. This report states that endophytic *S. aquimarina* SjAM16103 promotes the plant growth and produces plant growth promoting substances probably by means similar to plant growth-promoting rhizobacteria (PGPR). Plant growth promoting bacteria were environmentally friendly alternative to chemical fertilizers and pesticides, the use of which was regulated and sometimes forbidden, the market for bioinoculants is still expanding. Therefore, a better understanding of endophytic *S. aquimarina* SjAM16103 may help to elucidate its function and potential role more effectively in developing sustainable systems in agricultural field.

### Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Definition</th>
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</thead>
<tbody>
<tr>
<td>μm</td>
<td>Micrometer</td>
</tr>
<tr>
<td>μMol/mL</td>
<td>Micromoler per milliliter</td>
</tr>
<tr>
<td>mol %</td>
<td>Moler percentage</td>
</tr>
<tr>
<td>ng</td>
<td>Nanogram</td>
</tr>
<tr>
<td>pMol</td>
<td>Pico mole.</td>
</tr>
</tbody>
</table>

### Acknowledgments

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### References


