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Research Article

EFFICIENCY OF ENTEROBACTER AEROGENUS BACTERIA IN PRODUCTION OF SIDEROPHORE, IAA AND PHOSPHATE SOLUBILIZATION

Devi Soundarya Sanapala[1] and Padal S.B[2]

[1]Department of Botany, Andhra University, Visakhapatnam, Andhra Pradesh, India
[2]Department of Botany, Andhra University, Visakhapatnam District, Andhra Pradesh, India

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ABSTRACT

Soil is the utmost important in agriculture practices as it is the source of the nutrients for all plants. But Agrochemicals pose serious threats like pollution of soil and cause sterility of soil. So eco-friendly ways can be adopted to complement the action of the agro-chemicals and to decrease their use. One of the best ecofriendly practices is using Plant Growth Promoting Bacteria (PGPB), which promote plant growth. In the present study, bacteria isolated and purified from virgin forest soil of sivalingapuram (Eastern Ghats region, Ananthagiri Mandal, Visakhapatnam, Andhra Pradesh, India).Based on the morphological characters, further bacteria was screened for biochemical and growth promoting properties such as Catalase, Amylase, Protease, Chitinase, Cellulase , IAA, Phosphate Solubilization and production of siderophore. Among the 36 Isolates (sivalingapuram), two isolates were found to possess high values of phosphate solubilizing and production of siderophore activity. It is concluded that high production of siderophore and phosphate solubilizing was performed by potential isolate SS5-15. Further it was characterized by molecular 16S r RNA gene sequencing and phylogenetic tree construction, the bacteria is identified as Enterobacter aerogenus.

INTRODUCTION

India is mainly an agricultural country and agriculture has been the backbone of the country's economy since time immemorial. With increasing world population and ever increasing demand for food, chemical fertilizers are used excessively in agricultural set ups to increase productivity and yield [1]. One such alternative is use of plant growth promoting rhizobacteria as bio fertilizers [2 and 3]. Microorganisms have a vital role in agriculture as they promote the exchange of plant nutrients and reduce application of chemical fertilizers as much as possible. Though beneficial plant microbe interactions in the rhizosphere can influence soil fertility [4]. Plant growth promoting rhizobacteria (PGPR) of many plant species and confer beneficial effects, such as increased plant growth and reduced susceptibility to diseases caused by plant pathogenic fungi, bacteria, viruses and nematodes [5]. And these plant growth promoting bacteria significantly affect plant growth by increasing nutrient uptake, producing biologically active phytohormones and suppressing pathogens by producing antibiotics, siderophores and fungal cell wall lysing enzymes[6,7,8,9 and10]. It is also reported that PGPR is capable of solubilizing both inorganic and organic phosphates in soil [11]. The use of PGPR as biofertilizers is one of the most promising tools to improve primary production with low inputs of chemical fertilizers, through any of the possible mechanisms such as biocontrol, nutrient mobilization, phytohormone production or nitrogen fixation [12].These bacteria improve the soil structure and minimize the toxic effects of heavy metals and other toxins like pesticides [13, 14, 15,16,17,18 and 19].

Phosphorus is the second important element after nitrogen as a mineral nutrient in terms of quantitative plant requirements. It plays an important role in virtually all major metabolic processes in plant including photosynthesis, energy transfer, signal transduction, macromolecular biosynthesis and respiration [20] and nitrogen fixation in legumes [21] .The Phosphorus solubilizing bacteria promotes the soil fertility by converting the insoluble phosphorus to soluble form to ease the plant absorption [22 ,23 and 24]. During the last two decades knowledge on phosphate solubilizing microorganisms increased significantly [25 and 26].

Siderophores are complexing agents that have a high affinity for iron and are produced by almost all microorganisms in response to iron deficiency. PGPB are known to release metal-

Corresponding author:* **Devi Soundarya Sanapala
Department of Botany, Andhra University, Visakhapatnam, Andhra Pradesh, India

chelating substances such as iron-chelating siderophores into the rhizosphere. These siderophore-producing PGPB then influence the uptake by plants of various metals, including Fe, Zn and Cu [27,28,29,30 and31].

In the present study we selected the five areas around Ananthagiri mandal, Visakhapatnam District. The aim of the present study is to identify and characterize the efficient PGPB from the soil samples and rhizospheres. The isolates were screened for plant growth promoting activities like phosphate solubilization, IAA and production of siderophores.

MATERIALS AND METHODS

Sample Collection

We collected totally 80 samples, surrounding soils as well as rhizospheres, at five different places of Ananthagiri mandal sample1-Borracaves, sample2-Tyda, sample3- Ananthagiri, sample4-kasipatnam,sample5-Sivalingapuram), Visakhapatnam and brought to the laboratory in sterile bags respectively. These sterile bags are kept in icebox, to maintain native conditions of soil samples until microbial analysis is completed. Now bacteria were isolated from respective samples and morphology was studied under the microscope (Hitachi U-2910). Then different screening properties (Amylase, Protease, Chitinase, Catalase, Cellulase, IAA, Phosphate Solubilization and production of Siderophore) were performed for primary screening to identify the potent strain. The screened potent strains were further studied and characterized.

Isolation and Purification of PGPB bacterial Strain from soil sample

The soil samples were separated and serially diluted up to 10^{-6} dilutions. Nutrient agar plates were aseptically prepared and 10^{-3} to 10^{-6} dilutions were transferred to each plate and incubated for 24 hours at 30^0c.Morphologically different colonies were picked and streaked on fresh agar plates to obtain pure cultures. The pure cultures of the isolates were sub cultured and stored at 4^0c for further investigation. Bacterial colonies were distinguished by morphological characters and Gram staining.

Identification and Characterization of PGPB bacteria strain

Characterization of isolated bacteria was performed based on conventional procedures such as, morphological studies, cultural properties, staining nature, biochemical assays and molecular sequencing. Morphological characters include colour, elevation and edge of the colony. Gram nature of each isolates was initially determined by using crystal violet and safranin staining. Among biochemical tests, amylase test, catalase test, gelatin hydrolysis, urease test, citrate utilization was performed based on conventional techniques for bacterial characterization (Followed by Bergey's Manual), molecular characterization was performed by 16S r RNA gene sequencing.

Screening Tests for Efficient PGPB

Screening for Amylase activity

Amylase activity is performed according to the method proposed by Bernfeld 1955 [32]. Nutrient agar medium is prepared with starch and poured in to sterile petri plates. Then the bacteria culture is inoculated and incubated at 35^0C for 48 hrs. After 48 hrs incubation Iodine solution is added on culture plate and observed the zone for starch hydrolysis.

Screening for protease activity

Protease activity is performed according to the Bharat *et al*., 2014 [33]. Nutrient agar medium is prepared with Gelatin and poured in to sterile Petri plates. Then the bacteria culture is inoculated and incubated at 35^0C for 48 hrs. After 48 hrs incubation saturated Ammonium solution is added on culture plate and observed the zone for gelatin hydrolysis.

Screening for Chitinase activity

Chitinase activity is performed according to the Sonia *et al*., 2003 [34]. Nutrient agar medium is prepared with Chitin and poured in to sterile Petri plates. Then the bacteria culture is inoculated and incubated at 35^0C for 48 hrs. After 48 hrs incubation Congo red solution is added on culture plate, after one min washed with saturated sodium chloride and observed the zone for chitinase activity.

Screening for Catalase activity

Catalase activity is determined according to the method of Katsuwon and Anderson 1989 [35]. A small amount of 24 hrs incubated bacterial colony is transferred to a surface of clean, dry glass slide using a loop and placed a drop of 3% H_2O_2 on bacterial culture and observed the bubble formation.

In Vitro Screening for Indole Acetic Acid (IAA) Production

IAA production is calculated based on the method proposed by Ehmann, 1977 [36]. Briefly, a mixture of components (peptone 500 mg, Beef extract 300 mg, Yeast extract 300 mg, Nacl 500 mg, Tryptophan 200 mg and Water 100 ml) were sterilized by autoclaving at 15 lb pressure for 15 min. Then bacterial culture is inoculated and incubated at 37^0C for 48 hrs. Later, the bacterial cells were separated by centrifugation at 3,000 rpm for 5 min. 1ml of the resulted supernatant was mixed with 4 ml of the Salkowski's reagent and the preparation was incubated for 30 minutes at room temperature under the dark, pink colour formation confirmed the presence of IAA in the supernatant.

Quantification of IAA

Quantification of IAA produced by the PGPB was done spectrophotometrically by reading the absorbance of the treated supernatant at 535 nm was read Hitachi U-2910 Spectrophotometer. The concentration of IAA produced by the isolate in the broth was quantified by comparing the standard graph made using standard IAA produced from Himedia.

In Vitro Screening for Phosphate solubilizing activity

This assay performed based on the methodology proposed by Jackson 1973. The isolates from inorganic sources was screened by using Pikovskaya agar [37] amended with insoluble tricalcium phosphate. Solubilization of the complex insoluble tricalcium phosphate was indicated by the zone of clearance around the bacterial colony.

Qualitative Estimation for Phosphate Solubilization

Qualitative estimation of P solubilization ability of different isolates was determined for their tri-calcium phosphate (TCP) solubilizing activity on Pikovskaya agar plates. Medium was poured in sterile petriplates and after solidification of plates the

isolates were spot inoculated on the centre of agar plate aseptically. Later, all the plates incubated at 2 28 ± 2°C for 5-days and observed the formation of clear zone to check the Phosphate solubilisation. The phosphate solubilisation index (SI) was calculated as the ratio of the total diameter (colony + halo zone) to the colony diameter (Premono *et al*., 1996; Sarker *et al*., 2014).

In Vitro Screening for Production of Siderophore activity

Determination of the type of Siderophore Iron Percholate Assay by using the protocol devised by Schwyn and Neilands [38]. The Siderophore production was demonstrated by formation of orange color around the bacterial colony in CAS - blue agar medium containing chrome azurol S dye and incubated at 28^0c for 2 weeks [39].

Molecular Identification of selected bacteria Using 16 s rRNA Gene sequencing

DNA Extraction

1. Bacterial Genomic DNA was isolated using the InstaGeneTM Matrix Genomic
2. DNA isolation kit - As per the kit instruction below procedure followed.
3. An isolated bacterial colony was picked and suspend in 1ml of sterile water in a
4. Microfuge tube.
5. Centrifuge it for 1 minute at 10,000-12,000 rpm to remove the supernatant.
6. Add 200 µl of Insta Gene matrix to the pellet and incubate at 56 °C for 15 minutes.
7. Vortex at high speed for 10 seconds and place the tube in a 100 °C in heat block or boiling water bath for 8 minutes.
8. Finally, vortex the content at high speed for 10 seconds and Spin at 10,000 - 12,000 rpm for 2 minutes.
9. In result, 20µl of the supernatant was used per 50 µl PCR reaction.

PCR protocol

Using below 16S rRNA Universal primers gene fragment was Amplified using MJ Research Peltier Thermal Cycler.

Primer details

Add 1µL of template DNA in 20 µL of PCR reaction solution. 27F/1492R Primers used for bacteria, and then PCR reaction performed with below conditions: Initial Denaturation 94℃ for 2 min and then 35 amplification Cycles at 94℃ for 45 sec, 55℃ for 60 sec and 72℃ for 60 sec.

Final Extension at 72℃ for 10 min. DNA fragments is amplified about 1,400bp in the case of bacteria. Include a positive control (*E. coli* genomic DNA) and a negative control in the PCR.

Purification of PCR product

Removed unincorporated PCR primers and dNTPs from PCR products by using Montage PCR Clean up kit (Millipore). The PCR product was sequenced using the 518F/800R primers. Sequencing reactions were performed using an ABI PRISM® BigDyeTM Terminator Cycle Sequencing Kits with AmpliTaq® DNA polymerase (FS enzyme) (Applied Biosystems).

Sequencing protocol

Single-pass sequencing was performed on each template using below 16s rRNA universal primers. The fluorescent-labeled fragments were purified from the unincorporated terminators with an ethanol precipitation protocol. The samples were resuspended in distilled water and subjected to electrophoresis in an ABI 3730xl sequencer (Applied Bio-Systems).

Sequence primer details

Blast

The 16s r RNA sequence was blast using NCBI blast similarity search tool. The phylogeny analysis of our sequence with the closely related sequence of blast results was performed followed by multiple sequence alignment.

1. The program MUSCLE 3.7 was used for multiple alignments of sequences [40]. The resulting aligned sequences were cured using the program Gblocks 0.91b.
2. This Gblocks eliminates poorly aligned positions and divergent regions (removes alignment noise) [41]. Finally, the program PhyML 3.0 aLRT was used for phylogeny analysis and HKY85 as Substitution model.
3. PhyML was shown to be at least as accurate as other existing phylogeny programs using simulated data, while being one order of magnitude faster. PhyML was shown to be at least as accurate as other existing phylogeny programs using simulated data, while being one order of magnitude faster. The program Tree Dyn 198.3 was used for tree rendering. [42].

RESULTS

Soil samples were collected from five different villages of Ananthagiri mandal, Visakhapatnam. To identify potential PGPB strain. Geographical location of soil sample shown in Figure 1. Among the 36 isolates from sivalingapuram, the most efficient one is selected based on morphological characters and primary screening, further the efficient isolate SS5-15, was screened for biochemical tests are shown in Table 1.

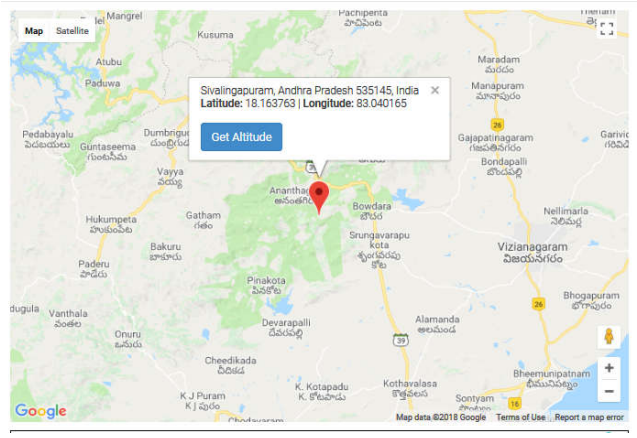

Figure 1 Geographical location of SS5-15 isolate

Total 36 isolates were screened for growth promoting activities such as Amylase, protease, chitinase, IAA, Phosphate solubilization, cellulose activity, HCN and production of siderophore were presented in Table 2.

Table 1 Biochemical tests of efficient isolate (SS5-15)

Name of test	Result
Microscopic structure (WET method)	Medium bacilli
Gram stain	- ve rod shape
Catalase	+ ve
Oxidase	- ve
Nitrate	+ ve
KOH	+ ve
Urase	-ve
Indole	- ve
Methyl red	- ve
Vogas proskauer	+ ve
Citrate	+ve
Gelatine	- ve
Casein	+ ve
Arginine	- ve
Lysine	+ ve
Ornithine	+ ve
Sucrose	+ ve
Lactose	+ ve
Manitol	+ ve
Salicin	- ve

in Table 5. High amount production of siderophore is by SS5-15, mentioned in Table 6.To confirm species level, the potent SS5-15 isolate was characterized by molecular 16S r RNA gene sequencing was shown in Figure 2: The phylogenic relationship is presented in Figure 3.

DISCUSSION

Plant Growth Promoting Rhizobacteria plays a major role in crop protection, growth promotion and in improvement of soil health. Previously reported that the PGPR inoculants supports the plant growth parameters like germination rate, seedling emergence, sustains to external stress factors and immunity towards diseases [43]. Therefore this study was carried out as an attempt to characterize the rhizosphere bacteria from Sivalingapuram region in Visakhapatnam, for their PGP activity. In vitro studies on Amylase, Protease, Chitinase, IAA, Phosphate Solubilization, production of siderophore, cellulose and HCN.

Table 2 Plant growth promoting properties of Rhizosperic isolates from sivalingapuram

Organism code	Amylase (Starch)	Protease (Gelatin)	Chitinase	IAA	Catalase	Phosphate Solublization	Iron	Cellulase	Oxidase	KOH	HCN
SS5- 1	–	+	-	+	-	-	-	-	+	+	-
SS5- 2	-	++	-	+	-	-	++	-	-	+	-
SS5- 3	-	+++	-	+	-	-	-	++	+	-	-
SS5 -4	-	-	-	+	-	-	+	-	-	-	-
SS5 -5	-	++	-	-	-	-	-	-	+	-	-
SS5- 6	-	+++	-	-	-	-	-	++	-	-	-
SS5- 7	-	+++	-	-	-	-	+	++	+	-	-
SS5- 8	-	-	-	-	-	-	-	-	+	-	-
SS5- 9 ca 1	-	-	-	++	-	+	+	-	-	+	-
SS5-10 ca2	-	+	-	+	-	-	++	-	-	+	-
SS5-11 ca3	+	++	-	-	-	-	+	-	+	-	-
SS5-12 ca4	++	+	-	-	-	-	-	-	+	-	-
SS5-13 k1	-	+	-	-	-	-	+	-	+	-	-
SS5-14 k2	-	++	-	-	-	-	-	-	+	+	-
SS5- 15 k3	++	+++	-	++	+	++	++	++	-	-	-
SS5 -16 k4	++	+++	-	-	-	-	+	+	+	-	-
SS5 -17 azo 1	-	-	-	+	-	-	-	-	-	-	-
SS5- 18 azo 2	-	-	-	+	-	-	-	-	-	-	-
R5 -1	-	-	-	-	-	-	-		-	-	-
R5 -2	-	-	-	-	-	-	-	++	+	-	-
R5 -3	+	-	-	-	-	-	-	-	+	-	-
R5 -4	+	-	-	-	-	-	-	-	+	+	-
R5 -5	-	-	-	-	-	-	-	++	+	-	-
R5 -6	-	-	-	-	-	-	-		+	-	-
R5 -7	-	-	-	+	-	-	-	-	-	-	-
R5 -8	-	-	-	-	-	-		++	-	-	-
R5 -9 ca1	-	-	-	+	-	-	-	-	+	+	-
R5 -10 ca2	+	-	-	-	-	-	-	-	-	-	-
R5 -11 ca 3	-	-	-	+	-	-	-	-	+	-	-
R5 -12 ca 4	+	-	-	+	-	-	-	-	-	-	-
R5 -13 ca 5	++	+++	-	-	-	-	-	-	-	-	-
R5 -14 k1	-	++	-	-	-	-	-	++	+	-	-
R5 -15 k2	+	+++	-	-	-	-	-	-	+	-	-
R5 -16 k3	+	++	-	-	-	-	-	++	+	+	-
R5 -17 k 4	-	-	-	-	-	-	-	-	+	+	-
R5 -18 k 5	+	+++	-	-	-	-	-	++	+	-	-

Plant growth promoting activities of SS5 -15 was showed in Table 3. Consolidated results showing the concentration of IAA (**µg**/ml) produced by the Rhizospere isolates under in vitro conditions was presented in Table 4.The potential SS5 -15 isolate produced high levels of Phosphate solubilization, shown promote the soil fertility and facilitate the plant growth promotion.

Table 3 Plant growth promoting activities of most efficient isolate (SS5 -15)

Organism Code	Amylase	Protease	IAA	Iron	Catalase	CMC	Oxidase	KOH	Phosphate Solubilizaton
SS5-15	++	+++	++	++	+	-	-	+	++

Amylase is an essential enzyme in degrading the starch and have huge applications in different industries of food, textile, fermentation and paper[44 and 45].In our present study, among 36 isolates were screened for amylase activity,11 isolates given positive results. The isolates SS5-11,SS5-12,SS5-15,SS5-16,RR5-3,RR5-4,RR5-12,RR5-13,RR5-15,RR5-16 and RR5-18.This series shown in Table 2. Proteases are commercially important enzymes of industrial productions and account for about 60% of sale around the world [46 and 47]. Among all the strains,18 isolates were shown best results. The isolates SS5-1,SS5-2,SS5-3,SS5-5,SS5-6,SS5-7,SS5-10,SS5-11,SS5-12,SS5-13,SS5-14,SS5-15,SS5-16,RR5-13,14,15,16 and 18.This series mentioned in Table 2. Chitinase plays a major role in many biological functions and widely distributed in plants, bacteria and fungi [48 and 49]. Cellulase are enzymes which able to break down cellulose. Reports suggest that most plant associated microorganism might have cellulose activity for adoption or establishment of a plant microbe interaction. Cellulase activities have seen in many Nitrogen fixing bacteria such as Bacillus spharricus, Bacillus circulans, Paenibacillus azotofixans and Azospirllum [50] . 11 isolates given promising results of cellulose activity. The isolates such as SS5-3,SS5-6,SS5-7,SS5-15,SS5-16,RR5-2,RR5-5,RR5-8,RR5-14,RR5-16,RR5-18.This series shown in Table 2.

Table 4 Consolidated results showing the concentration of IAA (**µg**/ml) produced by isolates under in vitro conditions.

S. No.	Isolate code	Mean Conc. ± SD (µg/ml)
1	SS5-1	13.43 ± **3.5**
2	SS5-2	15.67 ± 3.3
3	SS5-3	29±26.7
4	SS5-4	11.33 ± 6.7
5	SS5-9	43.33 ± 8.4
6	SS5-10	26.67 ± 12.5
7	SS5-15	75.61 ± 4.6
8	SS5-17	67 ± 19.6
9	SS5-18	30.17 ± 9.3
10	RR5-7	51.12 ± 9.6
11	RR5-9	45.3 ± 1.3
12	RR5-11	33.12 ± 4.7
13	RR5-12	29.6 ± 5.5

Table 5 Solubilization of Inorganic Phosphate in Pikovskaya medium by Bacterial isolates

S.NO	Isolate No	Result
1	SS5-9	+
2	SS5-15	++

Table 6 Siderophore production by Bacterial isolates in CAS- Blue Agar

S.NO	Isolate No	Result
1.	SS5-2	++
2.	SS5-4	+
3.	SS5-7	+
4.	SS5-9	+
5.	SS5-10	++
6.	SS5-11	+
7.	SS5-13	+
8.	SS5-15	++
9.	SS5-16	+

GCGGAGCTACACATGCAGTCGAGCGGTAACACAGAGAGCTTGCTCT
CGGGTGACGAGCGGCGGACGGGTGAGTAATGTCTGGGAAACTGCCT
GATGGAGGGGGATAACTACTGGAAACGGTAGCTAATACCGCATAAC
GTCGCAAGACCAAAGTGGGGGACCTTCGGGCCTCATGCCATCAGAT
GTGCCCAGATGGGATTAGCTAGTAGGTGGGGTAATGGCTCACCTAG
GCGACGATCCCTAGCTGGTCTGAGAGGATGACCAGCCACACTGGAA
CTGAGACACGGTCCAGACTCCTACGGGAGGCAGCAGTGGGGAATAT
TGCACAATGGGCGCAAGCCTGATGCAGCCATGCCGCGTGTATGAAG
AAGGCCTTCGGGTGTAAAGTACTTTCAGCGAGGAGGAAGGCGTTAA
GGTTAATAACCTTGGCGATTGACGTTACTCGCAGAAGAAGCACCGG
CTAACTCCGTGCCAGCAGCCGCGGTAATACGGAGGGTGCAAGCGTT
AATCGGAATTACTGGGCGTAAAGCGCACGCAGGCGGTCTGTCAAGT
CGGATGTGAAATCCCCGGGCTCAACCTGGGAACTGCATTCGAAACT
GGCAGGCTAGAGTCTTGTAGAGGGGGGTAGAATTCCAGGTGTAGCG
GTGAAATGCGTAGAGATCTGGAGGAATACCGGTGGCGAAGGCGGC
CCCCTGGACAAAGACTGACGCTCAGTGCGAAAGCGTGGGGAGCAA
ACAGGATTAGATACCCTGGTAGTCCACGCCGTAACGATGTCGACTT
GGAGGTTGTGCCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTTAAG
TCGACCGCCTGGGGAGTACGCCGCAAGGTAAAACTCAATGAATTGA
CGGGGGCCCGCACAAGCGGCTGGAGCATGTGGGTTTATTCGATGCT
ATCGCGAAGAACCTTACCTACTCTTGACATCCTGCTGGCACG

Figure 2 16S r RNA sequence of Enterobacter aerogenus

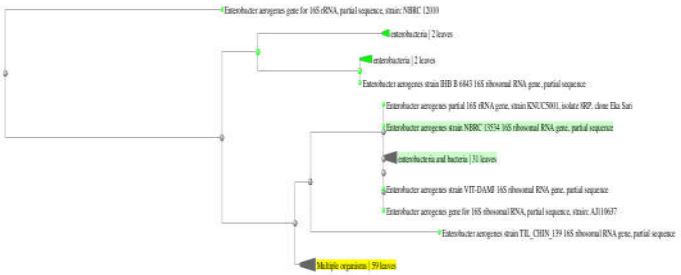

Figure 3 Phylogenic Tree of Enterobacter aerogenus

Phylogenetic relationship *Enterobacter aerogenus* on partial 16S rRNA gene sequences with 16S rDNA reference gene sequences available in NCBI. Reference sequences were determined after BLAST research. Based on the reference sequences after BLAST, the present isolate belongs to the phylum Proteobacteria, Class Gammaproteobacteria, order Enterobacteriales, family Enterobacteriaceae Genus Enterobacter and species aerogenus.

The rhizosphere bacteria appear to have a greater potential resource which colonize roots of plants and stimulate growth and yield directly and indirectly [51].Many rhizosphere bacteria have the capacity to synthesize IAA that has pronounced effect on plant growth and development [52 and 53]. Among 36 isolates, 13 isolates were shown positive results. In that SS5-15 is the best IAA producing strain.

Solubilization of complex phosphate is another important trait in plant growth promotion. A substantial number of microbial species exhibits phosphate solubilization capacity, these include bacteria, fungi, actinomycetes and even algae. In addition to Pseudomonas and Bacillus, other bacteria reported as P-solubilizers include Rhodococcus, Arthrobacter, Serratia, Chryseobacterium, Gordonia, Phyllobacterium, Delftia sp. [54 and 55], Azotobacter [56], Xanthomonas[57], Enterobacter, Pantoea and Klebsiella [58], Vibrio proteolyticus, Xanthobacter agilis [59]. In our present study, among the 36 isolates screened for phosphate solubilization,2 isolates were able to solubilize inorganic phosphate. The isolate SS5-15 showed best result of Phosphate solubilization.

Siderophores is associated with plant growth improvement either directly by carrying iron or indirectly limiting noxious organisms in the soil by sequestering iron available to them. The isolates SS5-2, SS5-4, SS5-7, SS5-15, SS5-16, RR5-7, RR5-9, RR5-11and RR5-12 were able to produce siderophores. Series has shown in Table 2.Thus, SS5-15 strain is consider as efficient isolate. In future this PAPB strain can helps on sustainable agriculture practice with minimal chemical inputs.

CONCLUSION

Based on the biochemical and plant growth promoting activities of different soil samples and rhizospheres, SS5-15 strain has shown remarkable results. After molecular characterization and sequencing analysis, SS5-15 was identified as Enterobacter aerogenus.

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