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Isolation of *Pseudomonas aeruginosa* from Quarry Sand

L Jeyanthi Rebecca*, R Kamalambigeswari and E Kowsalya

Department of Industrial Biotechnology, Bharath University, Chennai, India

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ABSTRACT

Quarries which are located close to surface water or ground water and the engineering people often facing the problems with drainage. Pseudomonas aeruginosa, can be used for biodegradation in liquid culture, but the roles and production in soils are unknown. Surface-active compounds for bacterial production like biosurfactants and bioemulsifiers which will increase the hydrocarbons of local pseudosolubility and that will improve in mass transfer to biodegrading bacteria. Several strains of potassium solubilizing bacteria, such as Acidothiobacillus ferroxidans, Bacillus mucilogenous, Pseudomonas, Burkholderia, B. edaphicus and B. circulans can be used as biofertilizer. Hence this study is focused on isolation and identification of Pseudomonas aeruginosa from quarry sand and it will be used further for more application purposes.

Keywords: *Pseudomonas aeruginosa*; Quarry sand; Biofertilizer; Biosurfactant; Pottasium solubilizing bacteria; Biodegradation

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INTRODUCTION

Some biosurfactants, are produced biologically from yeast or bacteria from various substrates including alkanes, oils, sugars, and wastes [1]. For e.g., Rhamnolipids produced by *Pseudomonas aeruginosa* from substrates including glycerol, pyruvate, citrate, fructose, succinate, olive oil, glucose and degradability and biocompatibility [2]. Rhamnolipids from *P. aeruginosa* UG2 were able to effectively remove ahydrocarbon mixture from a sandy loam soil, dependent on the type of hydrocarbon removed and the concentration of the surfactants used [2]. Phosphate Solubilizing Bacteria (PSB) is capable to convert insoluble phosphate in to soluble forms through the production of organic acids, chelates formation, exchange reaction [3]. Among bacteria strains capable to solubilize P are *Bacillus, Burkholderia, Enterobacter, Pseudomonas, Agrobacterium, Rhizobium, Micrococcus, and Flavobacterium* [4,5]. Potassium Solubilizing Bacteria (KSB) can dissolve K-minerals such as mica, illite and orthoclase in the soil through the production and excretion of organic acids or production of capsular polysaccharide [6,7]. Several strains of potassium solubilizing bacteria, such as *Pseudomonas, Burkholderia, Acidothiobacillus ferroxidans, Bacillus mucilogenosus, B. edaphicus, B. circulans* and *P. aenibacillus* sp. [7-9] can be used as biofertilizer.

MATERIALS AND METHODS

Quarry Sand Collection

Quarry sand was collected from the quarry industry, Nallambakkam, near Chennai, Tamilnadu. Sample was collected in a sterile plastic container and brought back to the laboratory for microbiological analysis and sample was stored at 4^{o}C until analysis.

Isolation of Bacteria

The microbes were isolated from quarry sand. 1 gm of sample was dissolved in 100 mL distilled water and serially diluted upto 10^{-6} using sterile distilled water and 1 ml of dilution was plated on to sterile nutrient agar plates and it was incubated at 37ºC for 48 hrs. After incubation well-isolated typical colonies were pure cultured. The pure culture was inoculated onto sterile nutrient agar slants, MacConkey agar plates and nutrient broth for further study. The culture was maintained on nutrient agar slants at 4ºC.

Identification of the Bacterial Isolates

Based on their cultural and morphological characteristics and biochemical reactions, the identification of isolate was performed using Bergey's Manual (1984).

RESULTS AND DISCUSSION

In this present study the *Pseudomonas aeruginosa* was isolated from the Quarry sand. Quarry sand was collected from the quarry industry, Nallambakkam, near Chennai Tamilnadu (Figure 1). Upon serial dilution and plating of quarry sand sample onto nutrient agar and MacConkey agar the bacteria was isolated. Quarry sand was collected from the quarry industry, Nallambakkam, near Chennai Tamilnadu.

Figure 1: Quarry Sand sample collected in Nallambakkam quarry industry

Isolation of *Pseudomonas aeruginosa* from Quarry Sand

Pseudomonas aeruginosa was isolated from the quarry sand. It was pure cultured and stored at 4ºC for further studies. The isolate was identified by the morphological, cultural and biochemical characteristics. The results were given in Tables 1 and 2.

Table 1: Morphological and cultural characteristics of bacteria from quarry sand

Bacterial isolate from quarry sand	Gram staining	Motility	Colony morphology on MacConkey agar
I	Gram negative rods	+	Clear = non-lactose fermenter (NLF)

Table 2: Biochemical characteristics of the bacterial isolates

Bacterial isolate from quarry sand	Growth on MacConkey agar	Growth in TSI	Indole test	Citrate test	Methyl red and Vogues Proskar	Identified bacteria
I	+\-	K\K no H_2S	+	-	-	*Pseudomonas aeruginosa*

Note- Reading on plates: +/- Growth and no fermentation; TSI Slant/Butt of the tube: K-Alkaline; H_2S- Hydrogen sulphide

Based on the morphological, cultural characteristics and biochemical reaction of the isolated bacteria was identified as *Pseudomonas aeruginosa*.

CONCLUSION

Biosurfactants, are biologically produced from bacteria from various substrates including wastes, oils, alkanes and sugars. Rhamnolipids from *P. aeruginosa* was able to removeeffectively a hydrocarbon mixture from a sandy loam soil and it was dependent on the type of hydrocarbon removed and the concentration of the surfactant used. *Pseudomonas aeruginosa* is Phosphate solubilizing bacteria (PSB) is capable to convert insoluble phosphate in to soluble forms through the production of organic acids, chelates formation, exchange reaction and can be used as a biofertilizer. Hence this study is focused on isolation and identification of *Pseudomonas aeruginosa* from quarry sand and it will be used further for more application purposes.

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