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Newley Isolated Bacilli from the Saline Soil of the Halophytic Plant Halopeplis Perfoliata Community

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1. Abstract

Exploring bacterial existence in environments such as soils represents a major opportunity to understand their roles in such environments, which helps in utilizing these magnificent organisms in the different applications of human use. In the present study, samples were collected from the soils of Halopeplis perfoliata community at the coastal line of Jeddah, Saudi Arabia. The bacterial stains were isolated from the soil samples on nutrient agar with the addition of NaCl. Genomic DNA was processed to identify these bacteria by 16s rRNA approach. As a result of this study, four types of bacteria have been identified; Bacillus amyloliquefaciens, B. halotolerans, B. subtilis, and Ammoniphilus sp. These bacteria are significant beneficial for biotechnological applications.

2. Introduction

It is known that the salty soil located near the sea has sufficient surprises [1]. The bacterial diversity that exists in these extreme environmental conditions are different from any other environments [2]. The bacteria that tolerate high salinity, and stands high temperatures are beneficial to extend the growth of many halophytic plants, especially the symbiotic bacteria, which may be a key concerned in plant stress [3,4]. Bacillus species considered the most species isolated from salty soil because they can adapt and grow in a harsh environment [5]. As part of a more extensive study of the bacteria that live in the saline soil of halophytic plant H. perfoliata community at the shoreline of Jeddah, Saudi Arabia, we have identified some bacterial strains from soil samples by using 16s rRNA gene.

3. Samples Collection

Five soil samples were collected from soils H. perfoliata and Zygophyllum album which are located at the seacoast of seacoast of the Southern Corniche of Jeddah (21.2189793, 39.1748844), Saudi Arabia on the 9th of January in 2019.

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The sampling site was chosen because the area is characteristically hot, arid and sandy with a lower amount of rainfall, and the maximum temperature in summers rises to 52 °C. Despite all these characteristics, the halophytic plants grow significantly in this region.

4. Isolation of Bacteria

0.5 g of each five soil samples were taken and spread on nutrient agar which had contained 22 g of NaCl, and then the samples were transferred to incubation at 37°C for 24 hr. Individual colonies of bacteria that were grown on nutrient agar picked up and purified by re-streaking onto 50 ml liquid Luria broth (LB). 6 g of NaCl and 6 g of d-glucose were added to LB, and transferred to shaker incubator at 37°C for 24 hr.

5. Identification of the Isolated Bacteria

DNA was extracted using Wizard® Genomic DNA Purification Kit. PCR of the bacterial 16S rRNA gene fragments was amplified with the universal primer 16S- 27f: 5' AGATTTGATCMTGGCTCAG3'. 16S-519R: GWATTACCGCGGCKGCTG3'. The amplification was performed using an initial denaturation at 94 °C for 5 min, followed by 30 cycles of 94 °C for 30s denaturation, 57 °C for 40s annealing, 72 °C for 1.30s extension with a final elongation at 72 °C for 10 min [6]. Later on PCR amplicons were sequenced by Sanger method.

6. Nucleotide Sequence Accession Numbers

The complete sequences were identified by BLAST nucleotide searches in the NCBI website, and registered as follow: Bacillus amyloliquefaciens, B. halotolerans, B. subtilis and Ammoniphilus sp with accession numbers; MN860193, MN860192, MN860184 and MN880730 respectively.

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