ABSTRACT

Forty-one aerotolerant subsurface bacterial colonies were isolated from terrestrial subsurface environment at various depths (5 feet to 50 feet at 5 feet intervals). Among those, 32 morphologically distinct bacterial colonies were selected for further gram staining reaction, various sugar utilization patterns and scanning electron microscopic studies. Morphologically different, gigantic structure with capsular sheath, footprint like appearance, and neither gram positive and gram negative bacteria were isolated from 40 – 50 feet depth. Most of the subsurface bacteria have utilized only 03 different types of sugars out of 24 different variety of sugars.

INTRODUCTION

Microorganisms are the only life forms that can inhabit most deep subsurface environments. The term deep terrestrial subsurface in reference to bacteriological studies is an emerging frontier for study of biodiversity, physiology, adaptation of bacteria. These unexplored terrestrial subsurface bacteria have begun to be used for innovative approaches for environmental management. A diverse microbial community has been found across geological profile, including very different physiological groups. It has been suspected that microorganisms living in the deep terrestrial subsurface are responsible for a number of important geochemical phenomena. It is necessary to understand the biochemical mechanisms of microbial metabolisms and the ecological interaction between various groups of microorganisms.

MATERIALS AND METHODS

Sampling site - Soil samples were acquired from Kattur, Tiruchirappalli, Tamil Nadu state, India.

Physico-chemical properties - Various physico-chemical properties were analysed.

Isolation of bacterial strains - Soil samples were blended with 0.1% sodium pyrophosphate (Na2HPO4.10H2O; pH-7) and serially diluted in a buffered salts solution (NaCl, KCl, KH2PO4, CaCl2.2H2O, MgCl2.6H2O, pH-7), plated on PTYG agar medium (peptone, tryptone, yeast extract & glucose) and incubated aerobically at 37°C for 3–4 days.

Gram staining studies - 32 well grown bacterial cultures were selected for gram staining reaction.

Electron microscopy studies - 06 bacterial isolates were used for scanning electron microscopic studies.

Utilization of various carbon sources - In total, 24 different sugars were used in this study.

RESULTS

The results of physical and chemical properties analysis have given in the (Fig.1, 2, 3 & 5).

Subsurface bacterial diversity - Forty-one aerotolerant subsurface bacterial colonies were isolated. Among those, 32 morphologically distinct bacterial colonies were selected for further studies. The bacterial culture photos (NTN01 to NTN07, NTN10 to NTN14, NTN16 to NTN18 and NTN20 to NTN36) are given in the Fig. (6, 7 & 8).

Gram’s staining reaction - Seven bacterial colonies were neither gram positive nor gram negative, no distinct morphological shape, gigantic structure with capsular sheath and footprint like appearance were identified. Others were gram-positive rods, gram-negative rods, gram-positive cocci and gram -ve cocci, bacilli respectively (Fig. 9 -16).

The sizes of those bacteria were identified from 4.5μm to 2.2μm (Fig.17 & 18).

Various sugar utilization tests - Most of the bacteria have utilized only 03 different types of sugars like maltose, galactose and glycerol or lactose, trehalose and mannose. Some bacteria were utilized nearly 14 different varieties of sugars. However, very few bacteria utilized the L-arabinose, α-methyl-D-glucurate and inulin. None of the isolates were utilized sodium gluconate, salicin, glucosamine and dulcitol (Fig.12, 13 & 19).

CONCLUSION

Morphologically different, gigantic structure, and neither gram +ve and –ve bacteria were isolated from 40 – 50 feet depth. In the present study, a modest attempt was made on the establishment of subsurface bacterial germ plasm in the Department of Microbiology, Bharathidasan University. Much more laboratories would be involved to explore the biodiversity of the subsurface bacteria and subsequently establish germ plasm of these least studied potential microbes.

References: