## ASSESSMENT OF FUNCTIONAL MICROBIAL DIVERSITY IN COASTAL AND DESERT SOIL ECOSYSTEMS OF GUJARAT

A Dissertation Thesis Submitted to Nirma University In Partial Fulfillment for the Degree of

> Master of Science In Microbiology

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## **CERTIFICATE**

This is to certify that the thesis entitled "Assessment Of Functional Microbial Diversity In Coastal And Desert Soil Ecosystems Of Gujarat" submitted to the Institute of Science, Nirma University, in partial fulfillment of the requirement for the award of the degree of M.Sc. in Microbiology, is a record of research work carried out by Pooja Rana (10MMB016), Bhruvi Shah (10MMB017) and Neelam Mahto (10MMB020) under the guidance of Dr. Nasreen S. Munshi. No part of the thesis has been submitted for any other degree or diploma.

Prof. Sarat Dalai

(Director)

Dr. Nasreen S. Munshi

(Dissertation Guide)

## **Declaration:**

The above dissertation was carried out jointly by Pooja Rana, Bhruvi Shah and Neelam Mahto under my guidance.

Dr. Nasreen S. Munshi

## 4. Summary

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In the present study functional microbial diversity was assessed for different coastal and desert soil ecosystem of Gujarat. Coast and sediment soil samples were collected from seven different coastal areas of Gujarat viz. Alang, Daman, Mandvi, Okha, Porbandar and Veraval while virgin and near shrub soil samples were collected from three different desert regions of Gujarat viz. Bhuj, Dhordo and Khavda. Nirma soil was taken as a control sample.

Soil samples were characterized for abiotic parameters like pH, temperature, electrical conductivity, moisture, water holding capacity, total organic carbon, Salinity and Chloride content. The pH was found nearly in alkaline range of 7 to 9. The coastal samples showed lower electrical conductivity than desert samples. Conductivity highly correlated with the salinity of the respective samples. Moisture content was found in the range of 3-25%. A significant negative correlation of moisture was found with microbial and enzyme activity. Salinity in coastal samples ranged between 0.1%- 0.6% NaCl while desert samples showed very high salt concentration up to 7.5% NaCl.

Microbial characterization was done for five major groups of organism i.e. General bacterial population, Nitrogen fixers and Nitrifiers, Functional groups of bacteria of other elemental cycles (Fe, P and S), Filamentous bacteria and Fungi including yeast. These organisms were enumerated by standard microbiological methods like spread plate; pour plate and Most Probable Number technique. All the bacterial populations were found in moderate to high number except gram negative bacteria, *Azospirillum*, Phosphate solubilizers, Mesophilic and Thermotolerant Actinomycetes, and moulds. Correlation coefficient analysis between abiotic factors and microbial count showed that pH and temperature affected microbial activity positively while EC, moisture and salinity affected negatively. As the soil in desert was highly compact and moisture content was high, which may be one of the reasons for low microbial population.

Functional diversity was studied by BIOLOG method using Ecoplate<sup>®</sup>. PCA analysis of 31 sole carbon source utilisation of 98 hours incubation showed that all coast and sediment samples were clustered together while desert samples were placed distantly. The total activity and Average Well Colour Development of the carbon source utilisation was obtained in the following decreasing order: BNS > NS > KNS > DPC > MC > PC > AC > DC > OC > VC. Bhuj near Shrub soil showed maximum utilisation of substrate. The C-source utilization by coastal soils was very poor as only two to three substrates were utilized, where as among the desert soils maximum 11 substrates out of 31 were utilised by Bhuj samples.

PCA analysis based on CLPP of all samples demonstrated grouping of all coast and sediment soils together while the desert soil seem to be different with respect to each other and the coastal soil.

Highest Shannon diversity value was obtained in soil samples of Bhuj and lowest was obtained in phordo samples on diversity calculation. All diversity indices showed a significant positive correlation with most of the microbial groups. Correlation between S and  $R_{margalef}$  was found to be significantly high (r = 0.786\*\*) and hence both the measures of richness provided similar results.

Enzyme assays were carried out for important enzymes of C, N, P cycle and dehydrogenase. Moderate to high enzyme activity was obtained in enzyme assays of cellulase, β-glucosidase, phosphomonoesterase and dehydrogenase. While in some samples protease, urease and L-asparaginase activity were not detected at all. β-glucosidase and L-asparaginase showed significant positive correlation with all diversity indices, most of the microbial groups and temperature while negative correlation with moisture and pH respectively. Alkaline phosphatase showed negative correlation with temperature. PCA analysis based on enzyme activity showed that samples were grouped together according to the regions.

Correlation between diversity indices, enzyme activities, abiotic parameters and microbial activities were compared by correlation co-efficient analysis by Pearsons' matrix method in SPSS (ver. 17.0) which helped us to understand the relation between community structure and functional diversity.

Diversity indices were found to be significantly correlated with enzyme activity and abiotic factors.

No significant correlation was found between enzyme activity and microbial functional groups as enzyme assays are not based on bacterial growth, the assay more closely reflects the bacterial community function *in situ* while the growth-based CLPP shows the functional diversity of the culturable part of the bacterial communities.

In general all the coastal regions were found to have low microbial diversity. Mandvi and Porbandar showed higher diversity as compared to other coastal samples. In desert regions Bhuj samples showed highest diversity while Khavda and Dhordo samples showed lower diversity which may be attributed to high salinity present.

Richness significantly correlated with functionally diverse groups of microorganisms which suggest that for high number of substrates to be utilized on Ecoplates<sup>®</sup>, functionally diverse microbial population is required.