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# Diazotophic diversity as a functional diversity indices in semi arid soil of Patan (North Gujarat)

**Ratna Trivedi[1], S.A.Bhatt*[2]**

[1]Department of Microbiology, Shree Ramkrishna Insitute of Applied Sciences, M.T.B. College Campus, Athwalines, Surat - 395 001, Gujarat, (INDIA)

[2]Department of Life-Sciences, Hemchandracharya North Gujarat University, Patan - 384 265, (INDIA)

E-mail: drratnatrivedi@gmail.com; shreyas_bhatt@yahoo.co.in

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## ABSTRACT

Among all groups of microorganisms, diazotrophes play important role in N mineralization in an ecosystem. Species Diversity is often used as a measurement of functional diversity at substrate level utilization by specific enzyme activities like Urease, L-Asparaginase, L-Glutaminase, Amidase, etc. Mathematical indices that describe the species richness, species evenness and Shannon-Weaver Index (S.W.I.) is used for measurement of diazotrophic diversity indices. In present study S.W.I. is measured for diazotrophes and correlated with functional diversity for substrate level utilization by specific enzyme activities viz., Amidase and L-Glutaminase. Soil samples were collected at 0-10 and 20-30 cms depth from four different sites around Patan city during Monsoon and Post monsoon seasons during May 2001 to June 2002. 

## KEYWORDS

SWI;
Diazotrophes;
L-glutaminase;
L-asparaginase.

## INTRODUCTION

Several mathematical indices that describe the species richness and apportionment of species within the community, called *species diversity indices*, are used to describe the assemblage of populations within a community[5]. Species diversity indices have rarely been applied to microbial communities because of the technical difficulties in speciating large numbers of microorganisms with their use and requires[1]. Microbial ecologists often use numerical taxonomy to determine the microbial species (taxonomic units) present in a sample[2-4,7]. In numerical taxonomy, a large number of characteristics (often phenotypic) are determined for organisms isolated from a sample, and cluster analyses are performed to establish the similarities between organisms. Similar organisms are considered to belong the same species. Widely used measure of diversity is the Shannon-Weaver index[6], which is also known as the Shannon-Weiner index or simply the Shannon index. This general diversity index is sensitive to both species richness (relative species abundance) and species evenness. Diazotrophs are involved in nitrogen mineralization in soil by producing various enzyme viz., Amidase, Urease, L-Asparaginase, L-Glutaminase etc. Substrate level utilization is most important parameter to deter-

mine of functional diversity. Soil Amidase and L-Glutaminase activities are principally due to mainly nitrogen mineralizers present in the samples that give indirect measurement of species diversity.

## MATERIALS AND METHODS

### Study area and soil

The soil samples were collected from around Patan city of North Gujarat region. Nature of soil is mainly semi-arid to dry; which was characterized by lack of moisture and neutral to alkaline pH, which influences the activities of soil microorganisms.

### Sample collection

Soil samples were collected from four different sites at different depth 0-10 cm and 20-30 cm during June 2001 to May 2002 during monsoon and postmonsoon seasons.

### Soil analysis

Soil physical and chemical characteristics viz., soil texture, moisture, WHC, FC were measured while Chemical characters like pH by pH meter (Orion ), Organic Carbon (Walkey Black method) & Total Nitrogen (by Micro-Kjeldahl method) were measured.

### Microbiological analysis

Diazotrophs were isolated on Nitrogen Free Medium ($KH_2PO_4$-0.2 gm, $K_2HPO_4$-0.8 gm, $MgSO_4$-0.2 gm, $CaSO_4$-0.1 gm, $MgSO_4$-trace, $Na_2MoO_4$-trace, Yeast Extract-0.5 gm, Sucrose-20 gm, Agar-15 gm, D/W-1000 ml, pH-7.2) by enrichment techniques. Isolated diazotrophs were studied for their morphological, biochemical characters and tentatively identified to species level.

### Microbial diversity measurement

### Shannon-Weaver Index of diversity ($\overline{H}$)

$$\overline{H} = \frac{C}{N}(N \log N - \sum n_i \log n_i)$$

Where C=2.3, N= number of individuals, $n_i$= number of individuals in the $i^{th}$ species

### Species richness (d)

$$d = \frac{S-1}{\log N}$$

Where S= number of species, N= number of individuals

### Species evenness (e)

$$e = \frac{\overline{H}}{\log S}$$

Where $\overline{H}$ = Shannon-Weaver diversity index, S = number of species

## RESULTS

**TABLE 1 : Soil physical properties**

| Site | Gravel (%) | Sand (%) | Silt (%) | Clay (%) | % WHC | % FC | % Moisture |
|---|---|---|---|---|---|---|---|
| I | 3 | 27 | 38 | 32 | 18.17 | 22.8 | 1.71 |
| II | 1 | 30 | 52 | 17 | 29.16 | 14.66 | 5.55 |
| III | 2 | 38 | 54 | 6 | 22.95 | 7.88 | 4.97 |
| IV | 1 | 3 | 26 | 70 | 26.8 | 11.14 | 5.58 |

**TABLE 2 : Soil chemical properties**

| | Monsoon | Character | | |
|---|---|---|---|---|
| Sites | Depths | pH | Org C % | Total N % |
| I | 0-10 | 9.0 | 1.10 | 0.0021 |
| | 20-30 | 7.5 | 0.30 | 0.0067 |
| II | 0-10 | 7.4 | 1.00 | 0.0030 |
| | 20-30 | 8.3 | 0.74 | 0.0034 |
| III | 0-10 | 6.9 | 1.08 | 0.0014 |
| | 20-30 | 7.3 | 0.62 | 0.0029 |
| IV | 0-10 | 7.0 | 0.50 | 0.0025 |
| | 20-30 | 7.8 | 1.10 | 0.0019 |
| | Post Monsoon | | | |
| I | 0-10 | 7.5 | 1.09 | 0.0014 |
| | 20-30 | 8.5 | 1.86 | 0.0038 |
| II | 0-10 | 7.4 | 1.76 | 0.0016 |
| | 20-30 | 8.6 | 1.44 | 0.0022 |
| III | 0-10 | 6.7 | 0.60 | 0.0056 |
| | 20-30 | 7.4 | 1.42 | 0.0028 |
| IV | 0-10 | 6.8 | 1.34 | 0.0053 |
| | 20-30 | 7.6 | 0.10 | 0.0098 |

**TABLE 3 : Soil amidase activity (µg-$NH_3$ released /g air dried soil)**

| Site | Mon soon | | Po s t-Monsoon | |
|---|---|---|---|---|
| | 0-10cm | 20-30cm | 0-10cm | 20-30cm |
| I | 65 | 0 | 20 | 52.5 |
| II | 27.5 | 2.5 | 24 | 5 |
| III | 20 | 42.5 | 12.5 | 0 |
| IV | 62.5 | 27.5 | 5 | 2.5 |

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**TABLE 4 : Soil L-glutaminase activity (µg-$NH_3$ released /g air dried soil)**

| Site | Monsoon | | Post -Monsoon | |
|---|---|---|---|---|
| | 0-10cm | 20-30cm | 0-10cm | 20-30cm |
| I | 87.48 | 18.15 | 51.15 | 85.8 |
| II | 70.45 | 54.45 | 37.95 | 42.9 |
| III | 72.6 | 88.5 | 33 | 75.9 |
| IV | 56.1 | 69.3 | 75.9 | 72.6 |

**TABLE 5 : Soil diazotrophic diversity indices**

| Sites | Monsoon | Indices | | |
|---|---|---|---|---|
| | Depths | H | e | D |
| I | 0-10 | 0.780 | 0.084 | 3.740 |
| | 20-30 | 0.190 | 0.041 | 1.740 |
| II | 0-10 | 0.440 | 0.063 | 2.740 |
| | 20-30 | 0.440 | 0.063 | 2.740 |
| III | 0-10 | 0.430 | 0.062 | 2.740 |
| | 20-30 | 0.040 | 0.020 | 0.740 |
| IV | 0-10 | 3.130 | 0.169 | 7.740 |
| | 20-30 | 0.040 | 0.020 | 0.740 |
| **Post Monsoon** | | | | |
| I | 0-10 | 0.040 | 0.020 | 0.740 |
| | 20-30 | 0.430 | 0.062 | 2.740 |
| II | 0-10 | 0.430 | 0.062 | 2.740 |
| | 20-30 | 0.780 | 0.084 | 3.740 |
| III | 0-10 | 0.190 | 0.041 | 1.740 |
| | 20-30 | 1.220 | 0.106 | 4.740 |
| IV | 0-10 | 0.190 | 0.041 | 1.740 |
| | 20-30 | 0.040 | 0.020 | 0.740 |

## DISCUSSION

Species diversity indices relate the number of species and the relative importance of individual species. The two major components of species diversity are species richness, or variety, and evenness, or equaitability. Species richness can be expressed by simple ratio between total species and total numbers. Causation must be used in interpreting the Shannon-Weaver index because it is sensitive to sample size, especially with small samples. Theoretically, diversity should increase during seasonal changes, organic matter, nitrogen content and humidity. This increase was observed in all samples during present study, using enzyme activity at substrate level utilization. Functional diversity values were higher during post monsoon season with increase in soil depth and with both the enzyme activities. Shannon-Weaver index was lower at increase soil depth during monsoon seasons. This infers that high soil moisture may have negative impact on functional diversity values. Diversity measured with S.W.I. increased in post monsoon season during study period. Some pioneer communities of soil experiences scarcity of nutrients condition due to the fact that competition with plant communities as plants show high nutrient uptake during monsoon. All the communities of soils play equally important part in functional diversity measurement with substrate level utilization. Season and other microorganisms like protozoa are most active which feeds on other microbes restricting their population and activities.

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