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RESEARCH ARTICLE

BIODIVERSITY OF MARINE FUNGAL SPECIES FROM SEA SHORE SOIL SAMPLE.

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Abstract

In the present study investigated the mycodiversity of the three spots (Ramnad, Tuticorin, Memesal) Tamilnadu, India. The marine soil samples were taken by scrapping off the surface and sub-surface to a depth of 10 cm. Approximately 1.5 kg of soils were collected from each site and put in plastic bags and labeled based on the collection sites. Marine fungi are the potential and promising sources for biologically active secondary metabolic production. The microbiological analysis results were subjected to statistical and physico-chemical parameters of the marine soil should be analysed.

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Introduction:-

Fungi are osmotrophs and therefore feed by secreting enzymes into the environment to process nutrients externally before taking the resulting metabolites into the cell (Bartnick –Garcia *et al.*, 1987). Fungi are unicellular or filamentous organisms that are devoid the chlorophyll. The word fungi lower case and not in italics, is commonly use as a collective term for organisms traditionally studied by mycologist from all three kingdoms (Hawksworth, 1991). Sea shores are located at the junction between water and land which is defined by the sand, wave and tide, regimes (Schlacher *et al.*, 2008). Fungi are one of the important microbial components of soil. Soil is the region on the earth's crust where geology and biology meet, the land surface that provides a home to plant animal and microbial life (Pelczar *et al.*, 1993). Soil sample is the most effective and popular materials for especially isolating a number of micro organisms such as fungi. The saprophytic microfungi play an important role as decomposers of cellulose in the form of washed-up leaves, algae and animal products such as chitin, keratin and calcium carbonate. Marine fungi have proved to be rich source of new biologically natural products because of their particular living conditions, salinity, nutrition, higher pressure, temperature variation, competition with bacteria, viruses and other fungi, they may have developed specific secondary metabolic pathways compared with terrestrial fungi (Liberra and Lindequist, 1995).

Studying fungal diversity is vital if we are to shed light on terrestrial ecosystem functioning. Marine microbial communities (bacteria, fungi, algae, plankton and viruses) are considered important ecological components in marine environments due to their performance in biogeochemical process. Biodiversity refers to the variability of life on earth, all the living species of animals, plants and microorganisms. According to Hawksworth (2002), fungi are major component of biodiversity, essential for the survival of other organisms and are crucial in global ecological process. The study involves isolation and identification of fungal species from three different ecological soil types. Physical and chemical analysis of the soil was also studied. Marine fungi have been recognized as one of the last barely tapped resources for new biologically active secondary metabolites including antitumor, antibacterial,

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antifungal, antiviral and enzyme inhibitors compounds. Marine fungi have proved to be a rich source of new biologically natural products (Mansuma *et al.*, 2001). Surveys have been carried out in the marine habitats to study the diversity and distribution of prokaryotic and micro eukaryotic kingdoms.

Materials and Methods:-

Sample collection

Totally three locations belonging to sea shore soil environment was selected in three different district, Tamilnadu. Approximately 1.5kg of soils were collected from each site. All the soil samples were dried at room temperature. To avoid contamination, collected soil samples were stored in presterilized polythene bags and used for the isolation of fungi.

Isolation of fungi

The fungi were isolated by the serial dilution method (Warcup, 1950). One gram of the soil sample was diluted in 100ml distilled water. For isolation of total fungi, 10^3 to 10^5 dilutions were used with potato dextrose agar plates using a spread plate technique supplemented with chloromphenicol to inhibit the growth of bacteria. The plates were incubated at room temperature 28 c for 3-4 days.

Potato Dextrose Agar

Ingredients	gm/l
Potato infusion	200
Dextrose	20
Agar	20
Final ph	5.6

Identification of fungal isolates

Lactophenol cotton blue staining

Identification was based on colony morphology and spore characters. Spore morphology was studied by microscopic observation. A drop of lactophenol cotton blue stain was placed in the centre of clean glass slide. A fragment of the fungus colony was taken with a sterile needle and placed it on the drop of lactophenol cotton blue stain. Gently the colony was teased and applied a cover slip. It should not be pushed down or tapped as this may dislodge conidia from the conidiospores. Then it was observed under required magnification.

Soil Analysis

Physical analysis of the soil such as pH, Colour, Texture, Temperature etc. were studied. Chemical analysis of the soil such as available nitrogen, carbon etc. were also studied. The physico-Chemical characters were statistically correlated with soil fungal flora.

Presentation of data

Data analysis

Species diversity: The diversity of species was studied in terms of species richness and relative abundance of the species.

Species richness (s): it represents the total number of different species in a particular area (Harrison, 2004)

Relative dominance (d): It was measured by calculating the berger-parker dominance (Harrison, 2004).

$$d = n/N$$

Where n = no of individuals in a species

N = s = total no of individual

Presentation of data: In order to assess the dominance of individual species in each site percentage contribution was worked out as follows,

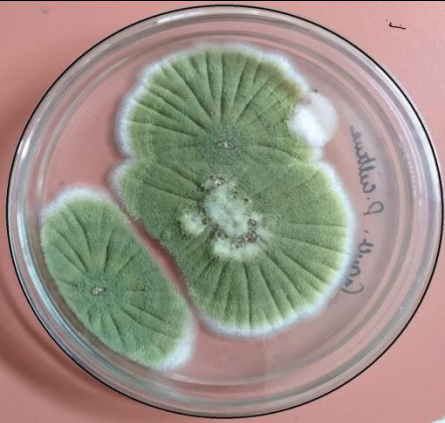


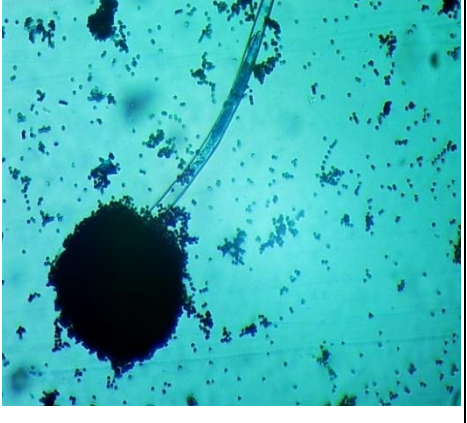

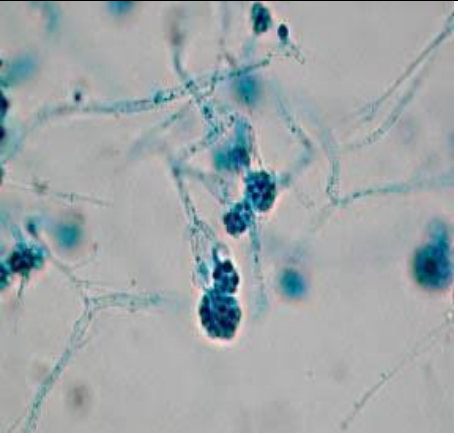
$$\% \text{ contribution} = \frac{\text{No. of colonies of fungus in a sample}}{\text{Total No. of all colonies of all the species in a sample}} \times 100$$

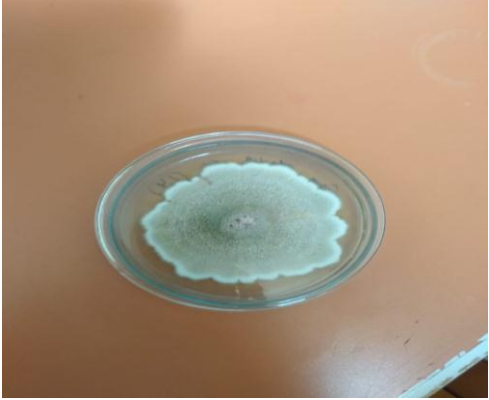
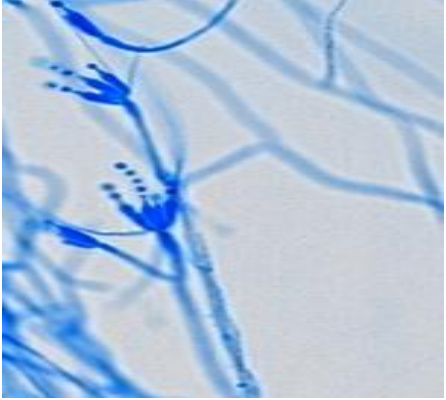




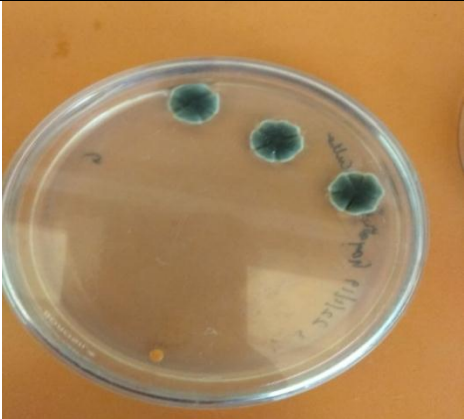

Result and Discussion:-

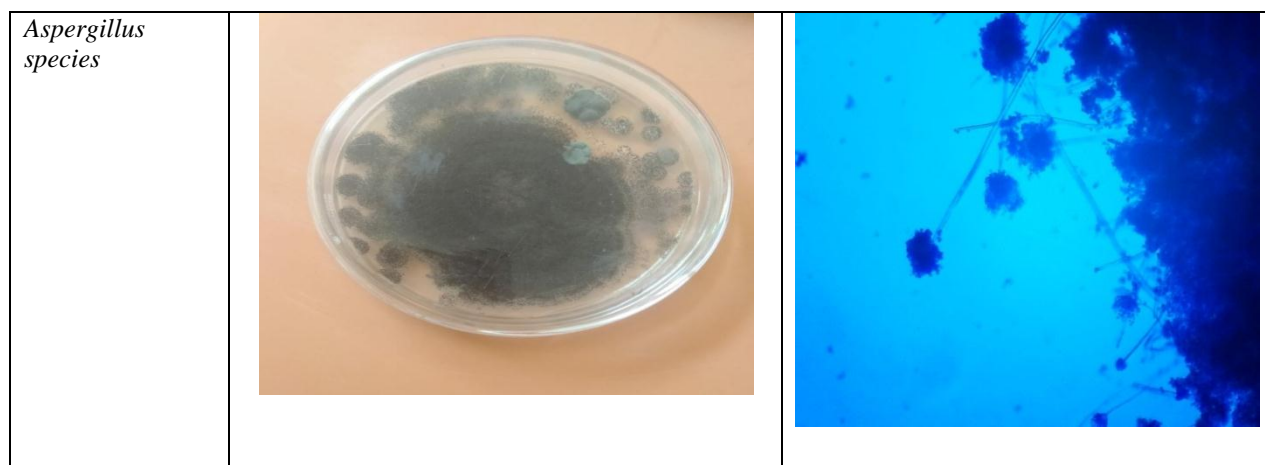
Isolation and Identification of fungi

Isolation of fungi from soil samples and the rapid screening by plating on potato dextrose agar plates. In the present study 8 isolates from marine soil were identified. The genus *Aspergillus* were dominant in all the three marine soil

samples. These fungal genera were identified according to their vegetative and reproductive characters following standard manuals and references. The fungi isolated from marine soil sample is shown as Picture 1.

<i>Aspergillus flavus</i>		
<i>Aspergillus niger</i>		
<i>Gliogladium species</i>		

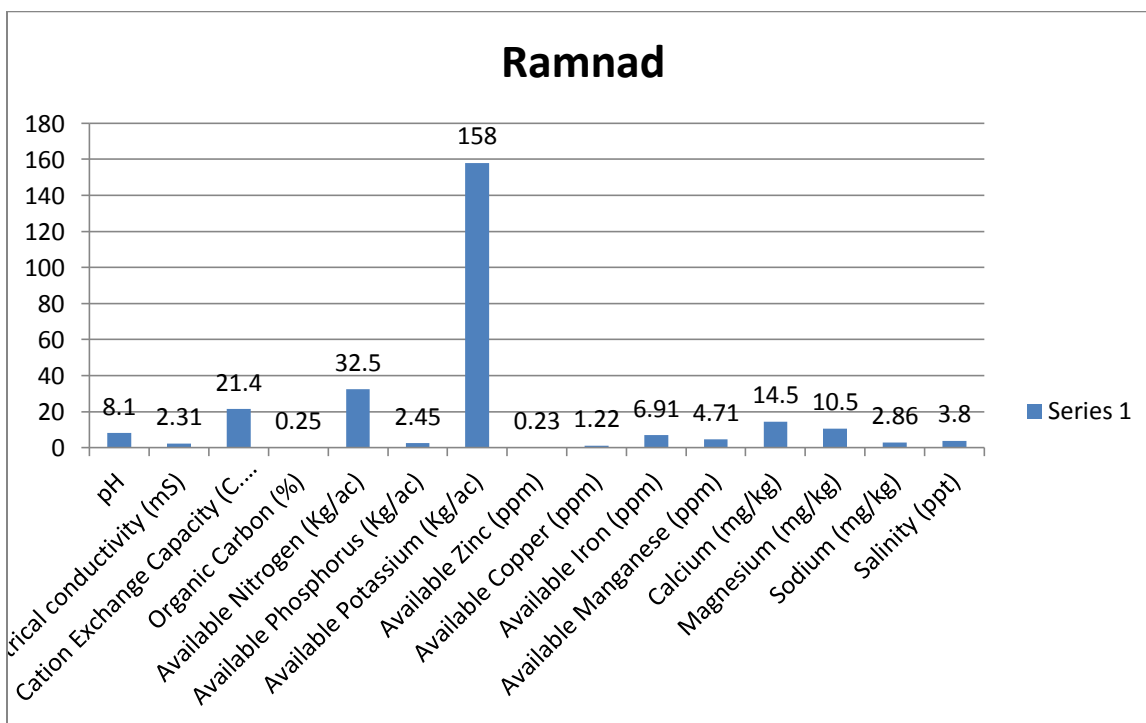
<i>Penicillium citrinum</i>		
<i>Penicillium species</i>		
<i>Chaetomium species</i>		
<i>Cladosporium species</i>		



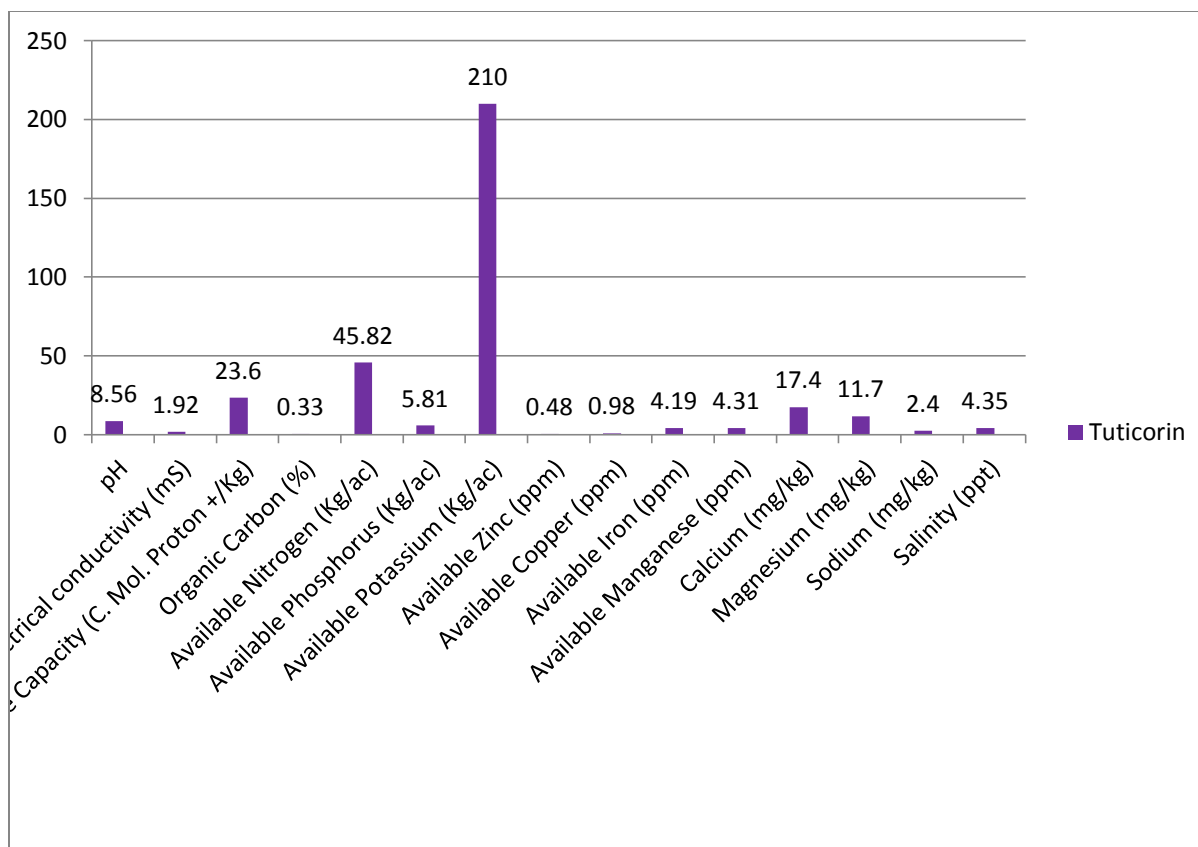
Picture 1:-It shows that the isolation and identification of fungi from marine soil sample.

Analysis of macro and micro nutrients

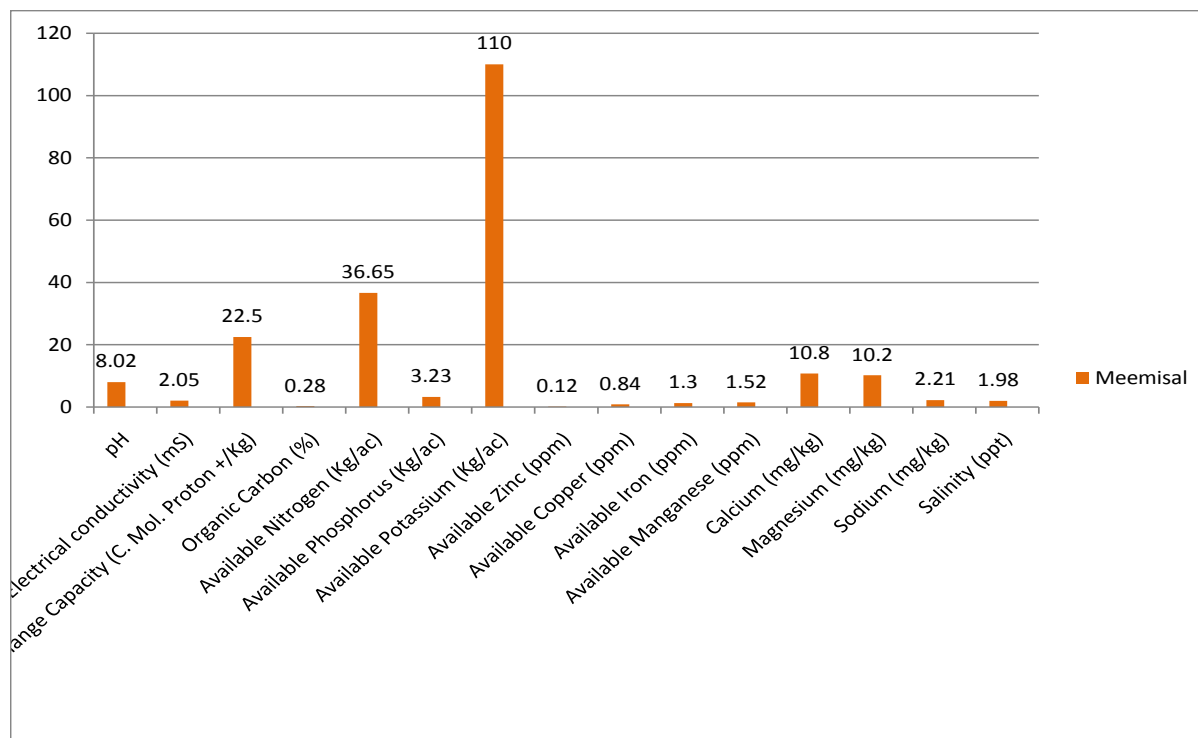
The primary macro nutrients like total N was determined by the Micro-Kjeldahl method (Jackson, 1973) and available P and K was analyzed by the Brays P1 method (Bray and Kurtz, 1945). Exchangeable bases (K, Ca, Na and Mg) and micro nutrients (Cu and Zn) of the soil were extracted with a Mehlich-3 solution (Mehlich, 1984). Organic carbon (C) was determined using Wet Walkely Black dichromate digestion method (Nelson and Sommers, 1982). Iron (Fe) was determined using the standard method described by Havlin and Sultanpour (1981). Organic matter (OM) was determined by the method of Walkely (1947).



Picture 2:-It represents the micro and macro nutrients should be analyzed in the ramnad district.



Picture 3:-It represents the micro and macro nutrients should be analyzed in the tuticorin district.



Picture 4:-It represents the micro and macro nutrients should be analyzed in mimisal (pudhukottai district)

Physico-chemical parameters

The marine sea shore soil has significant issue in the marine atmosphere s that influence the growth, replica and metabolic actins of microbes. In the present study the physico-chemical factor including soil texture, pH, electrical conductivity, cation exchange capacity, potassium etc... Parameters should be analyzed.

Table 1:-Physicochemical analysis of marine soil samples should be tabulated

S. No.	Parameters	Ramnad	Tuticorin	Meemisal
1	pH	8.10	8.56	8.02
2	Electrical conductivity (mS)	2.31	1.92	2.05
3	Cation Exchange Capacity (C. Mol. Proton +/Kg)	21.4	23.6	22.5
4	Organic Carbon (%)	0.25	0.33	0.28
5	Available Nitrogen (Kg/ac)	32.50	45.82	36.65
6	Available Phosphorus (Kg/ac)	2.45	5.81	3.23
7	Available Potassium (Kg/ac)	158	210	110
8	Available Zinc (ppm)	0.23	0.48	0.12
9	Available Copper (ppm)	1.22	0.98	0.84
10	Available Iron (ppm)	6.91	4.19	1.30
11	Available Manganese (ppm)	4.71	4.31	1.52
12	Calcium(mg/kg)	14.5	17.4	10.8
13	Magnesium(mg/kg)	10.5	11.7	10.2
14	Sodium(mg/kg)	2.86	2.40	2.21
15	Salinity (ppt)	3.80	4.35	1.98
16	Texture	Sandy	Sandy	Sandy loam
17	Total Number of Colony	18	13	12

Table 2:-Correlation between marine soil physicochemical parameter and total number of colony

Correlations																
	TN C	PH	EC	CE C	OC	AN	AP	AP O	AZ	AC	AI	AM	CA	MA	SO	S A
TN C	1															
PH	- 0.22 8	1														
EC	0.88 2	- 0.65 9	1													
CE C	- 0.77 8	0.78 9	- 0.98 2	1												
OC	- 0.68 0	0.86 9	- 0.94 5	0.99 0	1											
AN	- 0.62 6	0.90 2	- 0.91 9	0.97 7	0.99 7*	1										
AP	- 0.55 7	0.93 5	- 0.88 2	0.95 5	0.98 8	0.99 6	1									

AP O	0.13 3	0.93 5	- 0.34 9	0.52 0	0.63 7	0.69 0	0.74 9	1								
AZ	- 0.06 5	0.98 6	- 0.52 6	0.67 8	0.77 6	0.81 8	0.86 5	0.98 1	1							
AC	0.97 7	- 0.01 3	0.76 1	- 0.62 4	- 0.50 6	- 0.44 4	- 0.36 6	0.34 3	0.15 1	1						
AI	0.92 7	0.15 5	0.64 1	- 0.48 5	- 0.35 5	- 0.28 8	- 0.20 5	0.49 5	0.31 5	0.98 6	1					
A M	0.71 4	0.51 9	0.30 1	- 0.11 5	0.02 8	0.09 9	0.18 4	0.78 9	0.65 3	0.84 8	0.92 5	1				
CA	0.22 4	0.89 8	- 0.26 1	0.43 8	0.56 2	0.61 9	0.68 4	0.99 6	0.95 8	0.42 8	0.57 4	0.84 2	1			
M A	- 0.17 6	0.99 9*	- 0.61 9	0.75 6	0.84 2	0.87 8	0.91 6	0.95 2	0.99 4	0.03 9	0.20 6	0.56 3	0.92 0	1		
SO	0.99 1	- 0.09 8	0.81 3	- 0.68 8	- 0.57 7	- 0.51 8	- 0.44 3	0.26 2	0.06 7	0.99 6	0.96 8	0.80 0	0.35 0	- 0.04 5	1	
SA	0.44 1	0.77 4	- 0.03 3	0.22 2	0.35 9	0.42 4	0.50 0	0.94 8	0.86 7	0.62 3	0.74 5	0.94 3	0.97 4	0.80 6	0.55 5	1

Correlation is significant at the 0.05 level.

TNC - Total Number of Colony; PH - pH; EC - Electrical conductivity; CEC - Cation Exchange Capacity; OC - Organic Carbon; AN - Available Nitrogen; AP - Available Phosphorus; AP - Available Potassium; AZ - Available Zinc; AC - Available Copper; AI - Available Iron; AM - Available Manganese; CA - Calcium; MA - Magnesium; SO - Sodium; SA - Salinity.

Conclusion:-

The physico-chemical analysis of soil samples under evaluation showed variable concentrations of various boundaries. Irregular distribution of macronutrients and micronutrients were recorded. Thus the present study gives an idea about the fungal biodiversity in sea shores soil of three different districts. Tamilnadu. In all the three district surveyed, *Aspergillus* sp were common to all sites.

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