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Full Length Research Article

An Epitome of Mycodiversity and their Genomics with Emphasis on Enzyme Screening of Coastal Soil Samples of Dhanushkodi

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Abstract

The fungal diversity and its genomic analysis with reference to enzyme screening assay. Totally 44 mycodiversity from four different seasons such as postmonsoon, summer, premonsoon and monsoon were carried out. Maximum physicochemical characters were recorded in monsoon and minimum at postmonsoon. Mycofloral populations were high in premonsoon. Correlation of physicochemical factors with microbial population of soil samples were significantly resulted. Analysis of enzymes like amylase, protease and cellulase from forty-four fungal colonies were screened. Among the forty-four fungi screened, only three fungi has showed maximum zone of clearance in amylase than the other two enzymes respectively. The gene sequencing were performed for the fungal organisms such as *Aspergillus niger*, *A. terreus* and *A. flavus*. Above the fungal colonies were confirmed by the 18s rDNA gene sequencing and submitted to the NCBI. Hence, the three fungi like *A. niger*, *A. terreus* and *A. flavus* was suitable candidature for industrial enzyme production.

***Key words*:** Mycodiversity, Soil parameters, Amylase, Protease, Cellulase, Molecular studies

Biological approaches using microorganisms are gaining importance as an eco-friendly and cost-effective substitute to mitigate the pollution load [1]. A significant fraction of the environmental microorganisms remains unknown or unexploited due to the limitations associated with their cultivation in the laboratory through classical techniques [2]. The land area is the place where the land meets the sea. Marine fungi represent ecologically important group among benthic organisms acting as key intermediates of energy flow from detritis to higher tropic level in marine ecosystems [3]. The ecological importance of filamentous fungi in marine system that represent a diverse range of saprobes, pathogens and symbionts forming an integral part of coastal and deep sea [4]. Many of these fungi had been proven to be rich source of structurally novel and biologically active secondary metabolites which are emerging as a significant new chemical resource for drug discovery [5]. Diversity and distribution of different organism in the marine environments and influenced by the physico chemical properties of both water and the sediments. Point calimer includes many diverse habitats such as sandy and muddy shores and mangroves which have various physico chemical features. Soil samples were collected from the stations. viz, old light house, new light house, chola light house and Muniappan lake. A total of 59 species belonged to 20 genera ,6 species belonged to 4 genera were phycomycetes, 5 species belonged Ascomycetes and 47 species belonged to 13 genera were of Deuteromycetes. The diversity of soil fungi in coastal ecosystem has been studied by many workers notably [6-7]. Microorganisms are considered important suppliers of various bioproducts with applications in several industrial areas, such as enzymes. In fact, in the last decade, we have seen a significant increase in the demand for enzymes [8]. However, there are no reports available on soil fungi in Dhanushkodi coastal area. The present investigation was deliberated to study the diversity of soil fungi in Dhanushkodi coastal ecosystem.

MATERIALS AND METHODS

Dhanushkodi coastal area located in the south eastern part of Tamil Nadu, India. It lies between 9755°N and 65° 67°N latitudes and 57.6°E and 67.5°E longitudes.

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Collection of soil samples

The soil samples were collected seasonally like post monsoon, summer, pre monsoon and monsoon. The soil site at depth of 10 cm by using metal spatula and sterilized every time with 70% alcohol. At each station 5 to 7 samples were collected randomly and were pooled together. The samples were kept in sterilized polythene bags sealed and transported to the laboratory.

Isolation of soil fungi

The soil mycoflora were isolated [9]. The soil sample weighed 1g was diluted in 10 ml of 50% seawater (30ppt): distilled water. One ml of the diluted sample (10^{-2} and 10^{-3}) was poured and spreaded on the petriplates containing sterilized PDA medium (potato – 250 gm, dextrose –20 gm, agar agar - 15gm, distilled water500ml + marine water 500 ml, pH 7.7) supplemented with one percent streptomycin sulphate solution for prevention of bacterial growth. The inoculated plates were incubated in a dust free room at the temperature (24±2 °C) for 5-7 days. The colonies growing on PDA plates with different morphology were counted separately. The fungal cultures were then transferred, subcultured and the pure cultures were maintained on PDA medium.

Fungal morphology were studied macroscopically observing colour texture and microscopically by staining with lactophenol cotton blue were observed under Nikon phase contrast microscope (Nikon, Japan) for the Conidiophores and arrangement of spores [10].

Identification

The identification of fungal taxa followed as the standard Manual of soil fungi such as A Manual of *Penicillia* [11], A, Manual of soil fungi [10], Manual of *AspergilIi* [12], Hyphomycetes [13], Dematiaceous Hyphomycetes [14].

Presentation of data

Percentage of contribution and percentage of frequency of fungal isolates were calculated by using the following formula:

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

$$\text{Percent frequency} = \frac{\text{Number of all samples in which a particular fungus occurred}}{\text{Total number of samples examined}} \times 100$$

Based on the frequency occurrences, the fungi were grouped as rare (0-25% frequency), occasional (26-50 frequency), frequent (51-75% frequency) and common (76-100% frequency) species.

Physico-chemical analysis of soil

The physico-chemical parameters of collected soil samples were analyzed by standard methods. The analysis of physico- chemical parameters such as pH, Salinity (%), Electrical conductivity (dsm^{-1}), Organic Carbon (%),Organic Matter (%), Available Nitrogen (mg/kg), Available Phosphorus (mg/kg), Available Potassium(mg/kg), Available Zinc (ppm), Available Copper (ppm), Available Iron (ppm), Available Manganese (ppm), Cat ion Exchange Capacity (C. Mole $Proton^{+}$/kg), Calcium (mg/kg), Magnesium (mg/kg), Sodium (mg/kg) and Potassium (mg/kg) of the soil samples were done at Soil Testing Laboratory- Trichy, Department of Agriculture, Government of Tamil Nadu, India.

Statistical analysis

Pearson's correlation coefficient were analyzed and used to assess the relationship between the physico-chemical parameters and total number of fungal colonies. The data were computed and analyzed using Statistical Package for Social Sciences (SPSS) software.

Screening of enzymes

Amylase [15]

Screening and selection of potential isolates. The amylolytic fungal isolates were screened following the method of for their best enzymatic starch hydrolysis. The isolate with maximum clearance of zone was further studied and selected as the potential three fungal strains. Culture maintenance and preparation of pure isolates.

Cellulase [13]

The cellulase substrate used in the agar plate medium of clearing zone test was prepared according to the procedure as recommended. Cellulase activities of the highly active fungal filtrates were determined by using a carboxymethyl cellulase activity assay (CMC ase). Basal medium containing (g L^{-1}): CMC 10, $NaNO_3$ 6.5, K_2HPO_4 6.5, yeast extract 0.3, KCl 6.5, $MgSO_4 \cdot 7H_2O$ 3.0 and agar 17.5, was used for plate screening. In addition, conidia from one-week-old PDA plate's cultures were suspended in sterile water. A small well created in the middle of the screening agar plates and same number of conidia of each strain (~10^5) was inoculated into the wells. Plates were incubated at 28 °C for three to five days followed by 18h in the same conditions. Cellulolytic strains were selected based on the diameter of the cellulase hydrolysis and zone of surrounding the colonies were observed. For observations, plates were stained with 1% Congo red dye (0.5-1 h), followed by distaining with 1M NaCl solution for 15-20 min.

Protease

Production of proteolytic enzymes by fungal isolates was detected by using the Plate assay method. Which gelatin is the protein source of that growth medium. The fungal isolates were spot inoculated in Petri dishes and supplemented with 1% gelatin (Peptone, 5g; Beef extract, 3g; NaCl, 5g; Agar, 15g; Distilled water of 1 liter, pH 7). The Petri dishes were incubated at 28 ± 1°C for 3 days. After a week of incubation, gelatin degradation was observed as a clearing zone around fungal colony.

Molecular characterization of fungi

The 18S rDNA sequence for the *Aspergilius niger, A. flavus,* and *A. terreus,* have been deposited in gene bank http://www.ncbi.nim.gov./genebank.

The D_2 region of 18S rRNA gene sequence was used to carry out BLAST with the NR database of NCBI genbank database (URL http;//www.ncbi.n/m.nih.g). Based on maximum identity scores fist ten sequences were selected and Global pair wise sequence similarity between the sequence were performed using Needleman and Wunsuh algorithm available with the emboss sequence analysis suite. Multiple sequence analysis were performed using alignment program CLUSTAL W. The phylogenetic tree was constructed using MEGA 4. The evolutionary history was inferred using neighbor-joining method [18]. The boot strap consensus tree inferred from 500 replicated was taken to represent the evolutionary history of the taxa analyzed [19]. Branches corresponding to partitions reproduced

in less than 50% bootstrap replicates are collapsed. The percentage of replicated tree in which the associated taxa clustered together in the bootstrap test (500 replicates) are shown next to the branches, (next to the branches). The distance was computed using the kimura 2-parameter method [20] and are in the units of the number of base substation gaps substations per site Codon position included were 1^{st} +2^{nd} +3^{rd} +Noncoding. All positions gaps and missing data were eliminated from the dataset (complete deletion option). There was a total of 663 positions in the final dataset. Phylogenetic analyses were conducted in MEGA 4 [21]. Tree visualization was done with the tree view program.

RESULTS AND DISCUSSION

Study of marine fungal diversity plays a vital role to the understanding of the different process of the marine environment which will help to identify potential fungal organisms with novel bioactive compounds. In the present study, totally 44 species of fungi belong to 16 genera were isolated by plating techniques were identified and enumerated from costal area of Dhanushkodi. Among them Deuteromycetes were represented by 97.62% and Phycomycetes were 2.38% (Plate 1, Table 1). Our results are agreement with the finding of [22] who reported that 22 species belong to 10 genera are Deuteromycetes.

In the previous report *Aspergillus* sp. were seems to be the predominant genera with 21 species. The genus *Fusarium* sp was represented by four species followed by *Curvularia* sp and *Penicillium* were represented by five species and *Aspergillus niger* were represented from twenty-three species [23] recorded *Aspergilli* and *Penicillia* were predominant genera from south East coast of India [24] also reported that *Aspergillus* was dominant genera among the 23 colonies identified from Ramanathapuram District, Tamil Nadu, India.

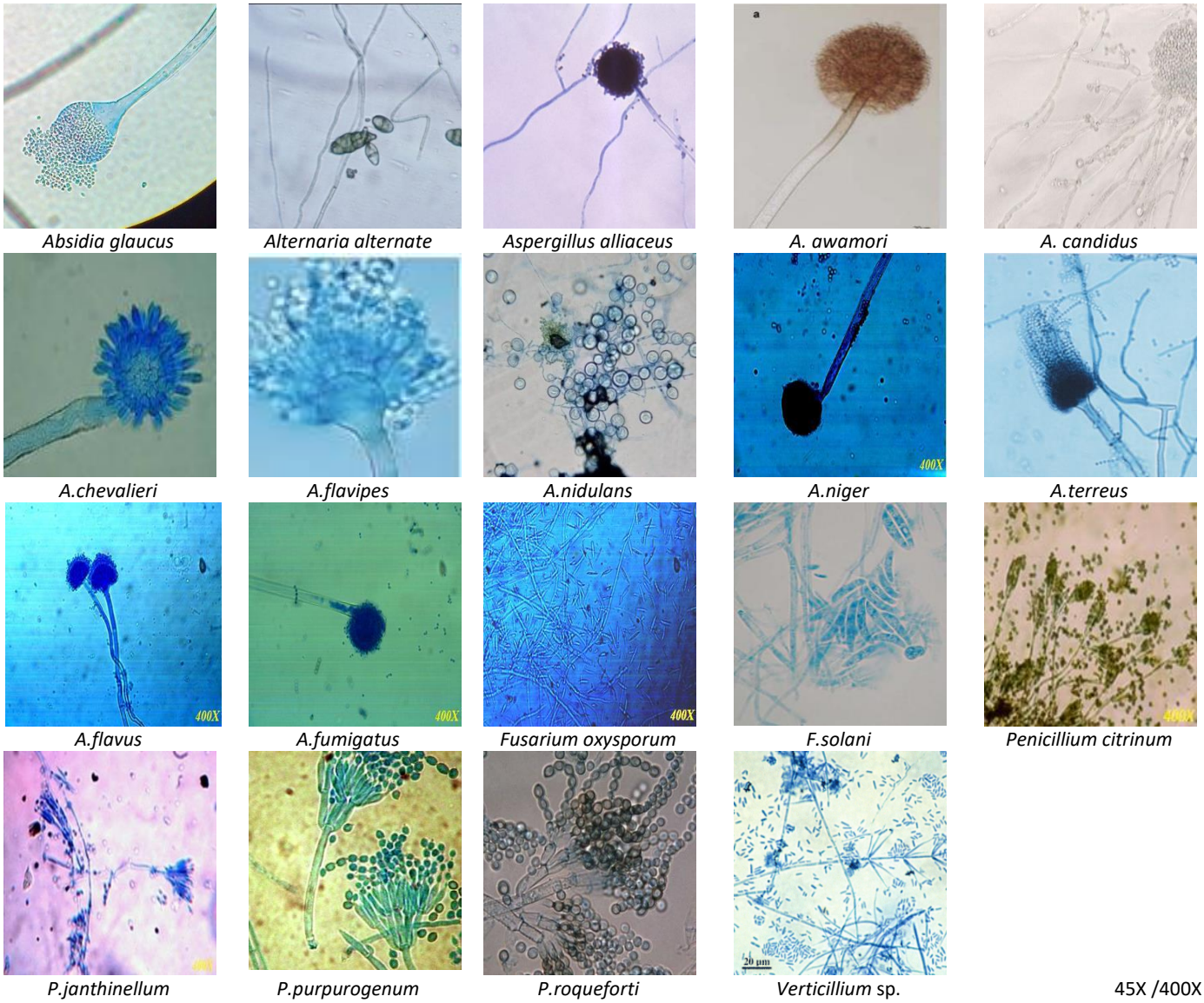

Plate 1 Identification of fungi from soil sample of Dhanuskodi east coast of Tamil Nadu

The maximum number of fungal species isolated from coastal area of Ramanathapuram District. The 44 fungal isolates species demonstrated that seasonal variation in the study stations. *Aspergillus niger*, *Aspergillus terreus* and *A. flavus* were commonly isolated from four season like post monsoon, summer, premonsoon and monsoon. The percentage contribution of fungal isolates from four seasons was analyzed. The maximum percentage of contribution was found with *Aspergillus niger* (7.8%), *A. terreus* (7.68%) and *Aspergillus flavus* (7.68%) from Dhanushkodi.

Aspergillus formed in the bulk which together contributed maximum percentage. The dominance of *Aspergillus* in any kind of coastal or marine soils was reported as unique feature but several investigations have identified different species of

Aspergillus as dominant one in different regions [22-13]. Evidently, the tolerance and adaptive mechanisms of dominance of the genus *Aspergillus* in coastal marine habitats may be due to their greater rate of spore production, dispersal partly due to growth in higher saline concentration.

In the present investigation from all the study stations were alkaline in nature. The pH of soil was ranged from 7.91 to 7.98. The major and minor elements such as nitrogen, phosphorus, potassium, zinc, copper, iron and manganese were showed variations in the study stations (Table 2, Fig 2). Alkaline condition has been explained as the characteristic feature of marine soils. Marine habitat such as coastal and brackish environs [27] sand dune and mangroves soils showed alkaline conditions as reported in the present study.

Table 1 List of Mycoflora isolated from coastal soil of Dhanushkodi during January to December 2019

Name of the fungi	Post monsoon		Summer		Premonsoon		Monsoon		Total number of colonies	Percentage of contribution
	TNC	MD	TNC	MD	TNC	MD	TNC	MD		
Aspergillus citrisporus	3	1	1	0.33	2	0.66	2	0.66	8	1.275
A. conicus	1	0.33	1	0.33	1	0.33	1	0.33	4	0.67
A. flavipes	3	1	8	2.6	9	2.9	4	1.3	24	3.827
A. flavus	9	2.9	12	3.9	14	4.6	13	4.29	48	7.655
A. fumigatus	3	4.3	10	3.3	11	3.6	15	4.95	38	6.060
A. fuscus	2	0.66	1	0.33	2	0.66	1	0.33	6	0.956
A. glaucus	1	0.33	1	0.33	1	0.33	1	0.33	4	1.637
A. nidulans	7	2.3	-	-	6	1.9	7	2.3	20	3.189
A. niger	12	3.9	11	3.6	14	4.6	13	-	50	7.974
A. spinulosum	-	-	1	0.33	1	0.33	1	0.33	3	0.478
A. sydowii	8	2.6	9	2.9	10	3.3		-	27	4.306
A. terreus	10	3.3	13	4.2	12	3.9	14	4.6	49	7.814
A. thomii	1	0.33	1	0.33	2	0.66	1	0.33	5	0.797
A. unguis	-	-	1	0.33	1	0.66	1	0.33	3	0.478
A. ustus	-	-	-	-	2	0.66	2	0.66	4	0.637
A. variegatus	1	0.33	1	0.33	1	0.33	1	0.33	4	0.637
A. versicolor	2	0.66	4	1.3	-	-		-	6	0.956
A. alliaceus	2	0.66	-	-	5	1.6	3	1	10	1.594
A. candidus	4	1.3	-	-	8	2.6	2	0.66	14	2.232
A. chevalieri	5	1.6	03	1	-	-	06	1.9	14	0.956
A. ochraceus	-	-	02	-	03	-	-	-	5	0.797
Absidia glauca	3	1	-	-	6	1.9	-	-	10	1.594
Acrocylindrium oryzae	1	0.33	1	0.33	1	0.33	1	0.33	4	0.637
Alternaria alternata	-	-	4	1.3	3	1	-	-	7	1.116
A. awamori	3	1	6	1.9	-	-	07	2.3	16	2.551
A. ruber	1	0.33	-	-	-	-	-	-	1	0.159
Chaetomium globosum	3	1	-	-	3	1	-	-	6	1.275
Curvularia geniculata	2	0.66	2	0.66	2	0.66	2	0.66	8	0.637
Fusarium oxysporum	10	3.3	9	2.9	12	3.9	14	4.6	45	5.263
F. solani	9	2.9	8	2.6	10	3.3	13	4.29	40	6.379
F.equiseti	-	-	2	0.66	2	0.66	-	-	4	7.177
F moniliforme	9	2.9	14	4.6	11	3.6	-	-	34	0.637
Gliocladium sp.	1	0.33	1	0.33	1	0.33	1	0.33	4	5.422
Helminthosporium sp.	2	0.66	1	0.33	-	-	1	0.33	4	0.637
Humicola sp.	1	0.33	1	0.33	-	-	1	0.33	3	0.478
Myrothecium verrucaria	1	0.33	1	0.33	1	0.33	1	0.33	4	0.637
Penicillium. Janthinellum	4	1.3	6	1.9	05	1.6	08	2.6	23	3.668
P. citrinum	11	3.6	9	2.9	07	2.3	06	1.9	33	5.263
P.purpurogenum	3	1	-	-	2	0.66	01	0.33	6	0.956
P. rouqueforti	-	-	5	1.6	1	0.33	-	-	6	0.956
Rhizopus stolonifer	2	0.66	2	0.66	2	0.66	2	0.66	8	0.275
Syncephalastrum sp.	2	0.66	1	0.33	2	0.66	2	0.66	7	0.116
Trichoderma polysporum	3	1	-	-	-	-	-	-	3	0.478
Verticillium sp	-	-	02	0.66	03	1	-	-	5	0.797
Total number of colonies	145		154		179		148		626	
Total number of genera	13									

*TNC-Total number of colonies, MD- Mean deviation

In the present investigation, relationship between various physico-chemical parameters of soil and total fungal colonies were statistically analyzed (Table 3). Available potassium and cat ion exchange capacity and manganese showed positive correlation. Similar results were represented in correlation analysis between physico-chemical parameters and fungal population revealed electrical conductivity showed positive correlation. [23] reported that the 39 isolates of mycoflora isolated from soil samples collected from coastal area of Tuticorin. The coastal soils contain a mycoflora reservoir composing of variety of a genera which contributes significantly to the ecological functioning of a marine ecosystem. The overall

investigation could be concluded that there is no uniformity in the diversity and distribution of marine fungi. Diversity of fungi are often influenced by the available nutrients and other physico-chemical parameters of the coastal ecosystem.

Table 2 Analysis of physico-chemical characteristics of the soil samples collected from Dhanushkodi

Physicochemical constituents	Collection Season			
	Post monsoon	Summer	Premonsoon	Monsoon
pH	7.91	7.98	7.91	7.92
Salinity (ppt)	0.36	0.45	0.784	24.1
Electrical conductivity (dsm^{-1})	0.26	0.49	0.51	0.42
Organic Carbon (%)	0.16	0.32	0.25	0.23
Organic Matter (%)	0.32	0.64	0.50	0.56
Nitrogen (mg/kg)	121.8	115.6	112.0	110.1
Phosphorus (mg/kg)	4.00	4.85	4.25	4.53
Potassium(mg/kg)	125.1	112.6	126.5	120.3
Zinc (ppm)	1.02	0.89	0.84	0.81
Copper (ppm)	0.52	0.49	0.42	0.41
Iron (ppm)	4.62	4.26	4.13	4.00
Manganese (ppm)	1.84	2.89	2.36	2.01
Cation exchange capacity (C. Mole $Proton^{+}$/kg)	28.6	24.8	23.0	22.0
Calcium (mg/kg)	11.3	13.2	12.6	11.7
Magnesium (mg/kg)	6.5	7.9	7.6	7.4
Sodium (mg/kg)	1.29	1.28	1.26	1.23

Table 3 Correlation of physicochemical parameters and total number of fungal colonies in soil samples of Dhanushkodi during Jan-Dec 2019

	pH	EC	OC	MA	N	P	K	ZN	C	FE	MN	CC	CA	MG	NA
pH	1														
EC	0.176991	1													
OC	0.940849	0.5	1												
OM	0.952683	-0.13055	0.793339	1											
N	0.887245	0.611051	0.991064	0.705045	1										
P	0.938453	-0.17386	0.765908	0.99904	0.673301	1									
K	-0.94639	0.150419	-0.78096	-0.9998	-0.69067	-0.99972	1								
ZN	0.87142	0.637026	0.986082	0.681082	0.999447	0.648352	-0.66625	1							
C	0.969549	0.412634	0.995177	0.849232	0.973199	0.825286	-0.83846	0.965016	1						
FE	0.792406	0.740613	0.952217	0.569495	0.984448	0.532939	-0.55288	0.989743	0.917663	1					
MN	0.858477	0.656698	0.981465	0.661968	0.998257	0.628497	-0.64679	0.999667	0.957932	0.993099	1				
CI	0.881121	0.621376	0.98923	0.695687	0.999914	0.663551	-0.68113	0.999797	0.9701	0.986666	0.998945	1			
C	0.717204	0.8128	0.910897	0.471446	0.957798	0.43236	-0.45365	0.966823	0.866025	0.993399	0.973091	0.961484	1		
MG	0.857146	0.658648	0.980966	0.660025	0.998101	0.626481	-0.64482	0.999597	0.957186	0.993399	0.999997	0.998823	0.973684	1	
NA	0.717204	0.8128	0.910897	0.471446	0.957798	0.43236	-0.45365	0.966823	0.866025	0.993399	0.973091	0.961484	1	0.973684	1

pH- Hydrogen ion concentration, EC- Electrical conductivity, OC- organic carbon (%), OM- organic matter (%), N - nitrogen(mg/kg), P - phosphorus (mg/g), K- pottassium (mg/g/kg), ZN- zinc (ppm), C - copper (ppm), FE - iron(ppm), MN - manganese (ppm), CI-cat ion exchange capacity (c.mole pro), CA- calcium (mg/kg), MG- magnesium (mg/kg), NA -sodium (mg/kg)

Screening of enzymes

In the recent years, the potential of microorganisms as biotechnological source of industrially relevant enzyme has stimulated interest in the exploration of extracellular enzymatic activity in several microorganisms (Table 4). Extracellular amylase, protease and cellulase enzymes were assayed by different *Phaeocremonium* sp and *Phaemoniella*, *Chlamydospora* isolates. In the present study, the dominant species such as *Aspergillus terreus* (21.19±0.28mm), *A. niger* (15.37±0.04mm) and *A. flavus* (19.26±1.89mm) showed maximum amylase activity followed by *Aspergillus flavus* (15.2± 3.27mm), *A. niger* (12.34±1.27mm) and *A. terreus* (19.45±1.28mm) the zone of clearance recorded showed best results for the protease activity. The cellulase enzyme was maximum observed in *Aspergillus flavus* (24.60±1.57mm), *Aspergillus niger* (13.12±1.9mm) and *A. terreus* (28.23±0.07mm), isolated from marine soil samples of Dhanushkodi.

Molecular analysis

In the present investigation 18s rDNA sequence of *A. niger*, *A. terreus* and *A. flavus* was performed to get accession number. *A. niger* (MW020327) *A. terreus* (MW019615) and *A. flavus* (MW020891). For the following organism. The 18S rDNA of *A. niger*, *A. terreus* and *A. flavus* has been deposited in *Genbank* http://www.ncbi.nim.gov./genebank. and phylogentic tree was also constructed by Neighbour –joining method and it concludes that the culture of other organisms based on nucleotide homology and phylogenetic analysis (Fig 4) were reported. Similar type of work was reported by [25] with regard to *T. harzianum.* The 18s rDNA of *T. harzianum* has been deposited in Genebank http://www.ncbi.nl.lh.gov/genebank. A phylogenetic tree was also constructed by Neighbour –joining method and it concluded that the culture of *T. harzianum* closely related to *Hypocera tixii* based on nucleotide homology and phylogenetic analysis.

Table 4 Screening of fungi with different enzyme activity by invitro method

Name of the fungi	Zone of clearance (mm)		
	Amylase	Protease	Cellulase
Aspergillus citrisporus	3.19±0.06	2.56±0.34	1.04±27.14
A. awamori	1.23±0.07	10.5±6.47	1.60±6.20
A. conicus	2.30±0.10	1.27±1.07	1.07±01.02
A. flavipes	1.45±0.10	2.30±4.12	2.30±7.17
A. flavus	19.26±1.9	15.2±3.27	24.60±1.57
A. fumigatus	3.15±0.71	1.34±6.24	4.30±14.12
A. fuscus	0.24±0.19	2.5±24.37	3.03±13.70
A. glaucus	2.15±1.35	1.27±1.28	0.27±0.17
A. nidulans	2.19±7.23	1.41±1.24	1.28±5.17
A. niger	15.37±0.04	12.34±1.27	13.12±1.9
A. spinulosum	1.27±0.21	0.25±0.27	0.51±2.01
A. sydowii	0.42±0.31	1.47±2.36	3.20±0.06
A. terreus	21.19±0.28	19.45±1.28	28.2±0.07
A. thomii	4.11±1.27	2.02±1.26	0.23±0.14
A. unguis	2.32±1.71	0.14±1.38	1.27±1.47
A. ustus	1.16±2.28	2.03±0.02	1.17±1.27
A. variegatus	2.36±2.24	1.22±0.16	2.43±0.28
A. versicolor	1.07±1.25	1.30±0.16	1.22±1.21
A.alliceus	1.23±0.40	2.24±0.14	1.13±0.23
A.candidus	3.24±1.23	0.31±0.25	0.41±0.13
A.chevalieri	1.18±1.42	1.25±0.31	1.52±0.11
A.ochraceus	0.54±0.18	1.12±0.25	1.38±0.14
A.ruber	0.73±0.24	0.37±0.11	0.39±0.01
Absidia glauca	0.13±1.26	2.25±1.07	2.24±0.01
Acrocylindrium oryzae	0.12±0.07	02.3±7.43	10.2±3.10
Alternaria fasiculata	0.29±0.09	1.24±6.53	09.0±7.07
Chaetomium globosum	4.17±1.12	2.34±0.49	2.17±1.08
Curvularia lunata	1.25±0.41	0.19±0.65	0.31±0.07
F. equiseti	0.24±0.23	0.17±0.34	0.37±0.24
F. solani	3.14±0.32	0.13±0.34	0.19±0.04
F.moniliforme	0.21±0.21	0.21±0.48	0.24±0.39
Fusarium oxysporum	2.42±0.25	3.59±0.28	0.22±0.48
Gliocladium sp.	2.31±0.02	0.12±0.14	0.67±0.19
Helminthosporium sp.	1.17±0.24	0.02±0.58	0.49±0.27
Humicola sp.	0.37±0.28	1.57±0.09	0.27±2.41
Myrothecium vearrucaria	0.36±0.40	0.67±0.24	0.49±0.20
Penicillium. Rougueforti	1.24±0.27	0.47±0.03	0.21±0.03
P.citrinum	0.26±0.34	0.17±0.59	0.16±0.28
P.purpurogenum	1.34±0.21	0.34±0.59	0.33±0.17
P. janthinellum	0.48±0.19	2.48±0.04	0.58±0.47
Rhizopus stolonifer	2.29±0.32	0.23±0.01	0.17±0.09
Syncephalastrum sp.	0.29±0.01	0.14±0.26	0.38±0.17
Trichoderma polysporum	0.19±0.11	0.18±0.26	0.27±0.10
Verticillium sp.	0.26±0.14	0.25±0.49	0.58±0.11

CONCLUSION

Even though marine fungi have been extensively studied in recent researches, our understanding and scientific knowledge of marine fungi is still very limited. Marine fungi produced unique and novel enzymes which have many industrial applications. So far, the major focus was on playing important roles in marine fungal taxa. Marine-derived fungi have revealed much promise in term of interesting enzymes amylase, protease, and cellulase with novel properties. Many biotechnological important enzymes have been produced by marine-derived fungal community isolated from a variety of marine habitats. There is still a very big scope to examine these fungi for other interesting and useful products, like extracellular polysaccharides and other secondary metabolites. Future studies should be focused on marine fungal biology to reveal interesting biochemical and physiological features useful to various new biotechnological processes by means of exploiting rare marine-derived fungi which will definitely pave the way for industrial mycology.

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Conflict of interest

We declare that we have no conflict of interest.

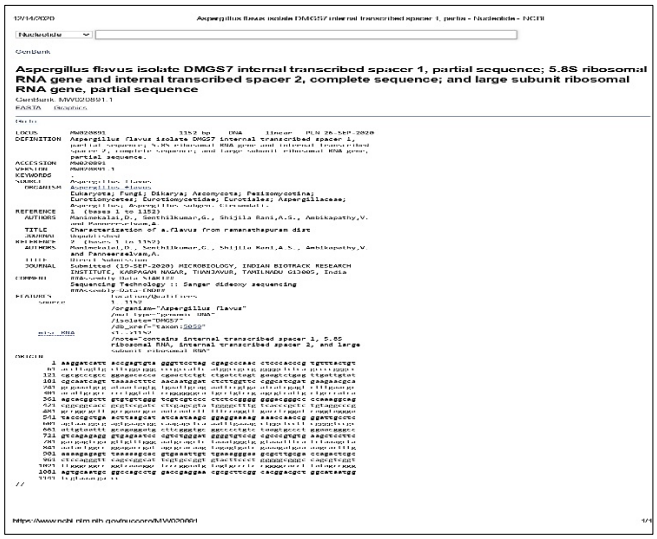

Fig 3 *Aspergillus flavus* genome NCBI submitted

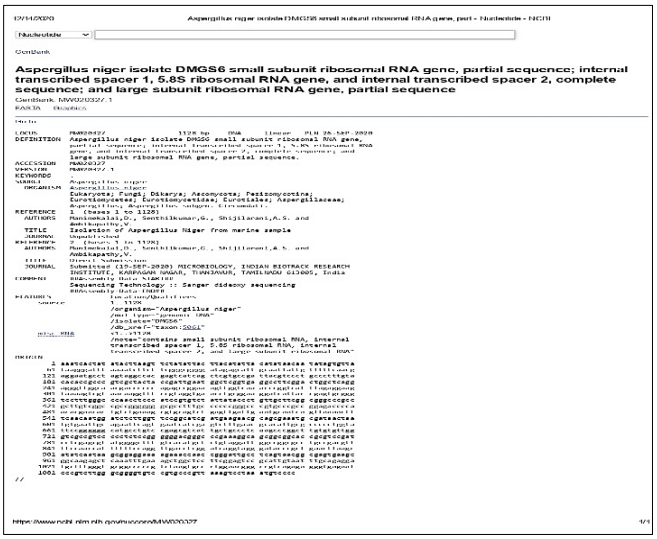

Fig 4 *Aspergillus niger* genome NCBI submitted

Fig 5 *Aspergillus terreus* genome NCBI submitted

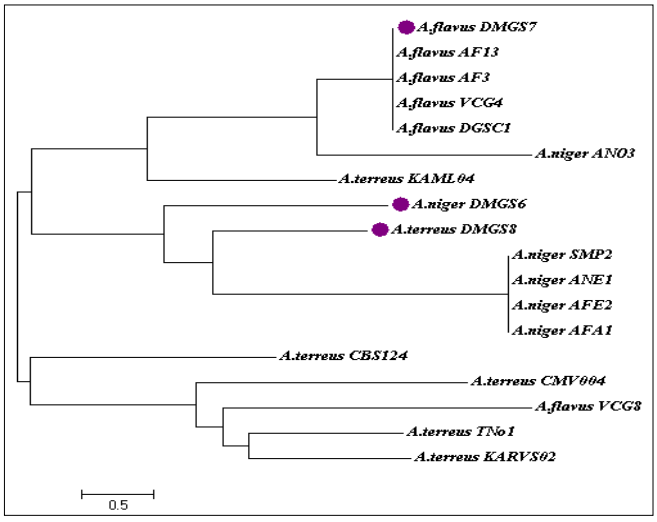

Fig 6 Analysis of phylogenetic tree for potential fungi

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