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Fungal Diversity Analysis of Fresh Water Perungudi Lake, Tamil Nadu, India

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A B S T R A C T

Water is the most important substance required for life on the earth. But today good quality water has become a scarce commodity. The poor quality of water affects human health. Thus, it is an urgent need to monitor the water quality regularly. The variety of microbial populations and activities of these microbes are influenced by the chemical, physical and biological characteristics of particular water as well as the presence of growing aquatic plants. Microorganisms are always present in water which may be pathogenic or non – pathogenic. Pathogenic forms include various species of bacteria, viruses, fungi and protozoans. Further, the interaction of water and microorganisms help in the biochemical purification of wastewater in silt places and in biological ponds. Hence the present investigation was carried out to determine Fungal diversity in a freshwater lake located in Perungudi, Chennai, Tamil Nadu, India. Freshwater samples for the study were collected from two different sites of Perungudi Lake, Tamil Nadu, India for 1 year from January 2019 to November 2019 with an interval of 1 month. The water samples were examined for Total fungal count and Identification of fungal species. In Perungudi Site 1 the total count of fungi was high as 11.7×10^2 CFU/ml during January 2019 and less than 0.33×10^2 CFU/ml in July, September and November 2019 in site 2 during the period of study. This variation in fungal count in site 1 of the Perungudi lake is due to the environmental and climatic conditions of the lake site which may affect the zooplankton and Phytoplankton of the freshwater.

***Key words*:** Aquatic fungi, Biodiversity, Fungal count, Fungal isolation, Water pollution

Water is essential for human to survive and plants need water for growth. It typically makes up 80 – 95% of the mass of growing plant tissues. It is a key priority issue for economic growth, employment, social development and environmental sustainability. It is a resource that is vital to communities both for their own survival and their contribution to societal needs. Access to clean water is therefore the foundation of any sustainable community [1]. Pollution of water with Pathogenic microbes, such as bacteria, viruses, parasites as well as fungi has been increasing globally [2]. The impact of river and lake pollution on human health depends mainly on the usage of water as well as the concentration of pathogens in the water [3]. These have contributed to the observation that; infectious diseases continue to be one of the leading causes of mortality globally [4]. Waterborne pathogens present a great health risk to people using river or lake water for drinking, bathing, washing, construction, irrigation of crops eaten raw, fishing and recreational activities [1-4]. Microbial communities are the key members of many ecosystems on the earth [5]. Around one-third of global freshwater reserves are placed in subsurface streams and aquifers which represent a freshwater source for human consumption as well as irrigation [6]. The exploration of microbial communities through culture-dependent methods were used from ancient time. Although there is increasing evidence that freshwater fungal diversity is high, the study of the biodiversity of freshwater fungi is still in its infancy. In light of the rapid decline in freshwater biodiversity, it is timely and necessary to evaluate the diversity and potential ecological function of this fascinating and diverse group of freshwater organisms [7]. Hence based upon the above views and Literature cited, an attempt has been made to determine the fungal diversity in a freshwater lake, in Perungudi, Chennai, Tamil Nadu, India.

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MATERIALS AND METHODS

Collection and analysis of fungi and bacteria

Two different sites of the Perungudi lake were selected for the study. Water samples were collected for fungal diversity analysis from Site 1 and Site 2 for a period of 1 year from January 2019 to November 2019 with an interval of 1 month. The samples were collected in sterile plastic bottles from each site and brought to the laboratory for further study.

Total count

For isolation of fungi, 1ml of water sample was cultured on malt extract agar medium (MEA), following the pour plate method [8]. For each dilution, triplicates were prepared. Plates were incubated at room temperature and observed for 5 to 7 days for the appearance of colonies. Colonies were counted from all 3 plates and the average was recorded. The results obtained were presented as (Colony forming units) "Cfu/ml".

$$\text{Number of Cfu /ml} = \frac{\text{Average number of colonies / plate}}{\text{Volume of sample}} \times \text{Dilution factor of water sample}$$

Isolation of pure culture of fungi

Isolation of fungi was carried out by serial dilution method [9]. Cultures were maintained on Potato Dextrose Agar (PDA), Rose Bengal agar medium and Nutrient Agar medium.

Identification of fungal strains

The isolated fungal strains were identified by using Lacto phenol cotton blue stain and fungal slide culture technique. The standard plate count is a reliable method for enumerating fungi [9]. Identification of pure fungal culture was carried out using the aquatic fungi Manual [10-12].

RESULTS AND DISCUSSION

Total count of fungi

The results of the Total Colony forming units of fungi are presented in (Table 1). In site 1 maximum (7.33 CFU/ ml) colonies of Yeast were observed in march 2019 and a minimum number (0.33 CFU / ml) of colonies were found in November 2019, whereas in site 2 higher number (4.66 CFU / ml) Yeast colonies were found in March 2019 and less (0. 66 CFU / ml) number of colonies were recorded in September 2019. *Aspergillus flavus* showed a maximum (12 CFU / ml) number of colony-forming units in May 2019 and a minimum (3. 66 CFU / ml) in July 2019 in site 1. In site 1 *Aurebasidium pullans, Aspergillus nidulans* and *Aspergillus terreus* showed a maximum (12 CFU / ml) in May 2019 whereas in site 2 *Aspergillus fumigatus, Curvularia lunata, Chrysonilia sitophila, Cladosporium oxysporum, Aspergillus flavipes and Rhizopus stolonifera* were found (12 CFU / ml) in May 2019. *Aspergillus niger* showed fewer (3.66 CFU / ml) colony-forming units in July 2019 and more (5.3 CFU /ml) colonies were present in July 2019. In site 1 *Penicilium citrinum* showed (0.66 CFU / ml) number of colonies in September 2019. *Acremonium strictum* showed maximum (0.66 CFU / ml) number of colonies in September 2019 and minimum (0.33 CFU / ml) in the month of November 2019 in site 2. Thus, the total count of fungi was maximum (11.3×10^2 CFU /ml) in January 2019 and minimum (0.33×10^2 CFU /ml) in September and November 2019 in site 1 whereas in site 2 the total count was more (11.7×10^2 CFU /ml) in January 2019 and less (0.33×10^2 CFU /ml) in July, September and November 2019 during the period of study.

Isolation and Identification of fungal species

The results of fungi isolated from the water samples in site 1 and site 2 of the freshwater lake are depicted in (Table 2) and the results showed that 14 fungal species namely Yeast colonies, *Aurebasidium pullulans, Aspergillus nidulans, Aspergillus flavus, Aspergillus terreus, Aspergillus niger, Penicillium citrinum, Aspergillus fumigatus, Curvularia lunata, Chrysonilia oxysporum, Aspergillus flavipes, Rhizopus stonifer* and *Acremonium strictum* were found to be present in site 1 and site 2 of Perungudi lake. The total count of fungi ranged between 0.33×10^2 to 11.3×10^2 in site 1 and site 2 recorded 0.33×10^2 to 11.7×10^2. The fungi counts were higher in the dry season due to the presence of many air – borne spores in the environment.

Table 1 Total Fungal Count– Colony Forming Units (CFU)/ml from Site 1 and 2 of Perungudi Lake, Chennai

Months	Samples	Average CFU/ml
January	Site 1	11.3×10^2
	Site 2	11.7×10^2
March	Site 1	2.3×10^2
	Site 2	4.3×10^2
May	Site 1	0.66×10^2
	Site 2	0.66×10^2
July	Site 1	Nil
	Site 2	0.33×10^2
September	Site 1	0.33×10^2
	Site 2	0.33×10^2
November	Site 1	0.33×10^2
	Site 2	0.33×10^2

Freshwater fungi are a diverse and heterogeneous groups comprising many species from different orders in which Ascomycetes and Hyphomycetes are dominant. Research related to freshwater hyphomycetes were less considered as reported by [13]. who recognized them as 'Aquatic Hyphomycetes'. Later these fungi have also been described as ''Freshwater Hyphomycetes'' [14] and ''water-borne Hyphomycetes'' [15]. Hyphomycetes is one of the main orders which comprise 4 biological groups; viz., Ingoldian, aero aquatic, terrestrial aquatic hyphomycetes and submerged aquatic hyphomycetes [16]. The members of Chytridiomycetes and Oomycetes are mostly aquatic and commonly known as water moulds [17]. The main role of the freshwater Ascomycetes, Basidiomycetes and Mitosporic fungi in freshwater ecosystems involves the degradation of dead organic material [18]. The results of the above study revealed that out of 14 fungal species identified Yeast is dominant and the majority of fungi *Aspergillus niger, Aspergillus fumigatus, Aspergillus flavus* and *Pencillium citrinum* were likely to be originated from soil or entered the water with plant remains. The isolated fungi from the lake belong to the category of transient accidental microorganisms according to the ecological classification of heterotrophic microorganisms. Transient and accidental microorganisms can develop sporadic activity and soil fungi may participate in microbiological processes in water bodies [19]. Ingoldian fungi have conidial shapes that include tetraradiate, branched or filiform in which the dominant conidial shape is tetraradiate. The conidial shape in aquatic hyphomycetes is used to minimize downstream transport [15]. This is in accordance with the reports of [20] that *Aspergillus* is biologically one of the most successful of all fungi and is expected to occur on all sorts of organic debris [21]. Analysis of the taxonomic diversity revealed the dominance of phylum, Ascomycota in all the samples [22-23].

Fungi are ubiquitous achlorophyllous and heterotrophic organisms, which are directly influenced by environmental factors. They are cosmopolitan in occurrence. They are found in rivers, and oceans and occur commonly on decomposing organic matter. Excessive levels of nutrients and other chemicals lead to changes in aquatic life [24]. Heterotrophic organisms are usually present in natural water in direct proportion to the physicochemical nature of the aquatic environment [25]. Fungi play an important biological process

in an aquatic ecosystem. Aquatic fungi act as significant decomposers in the aquatic ecosystem of animal and plant remains. Aquatic fungi play a significant role in the energy flow, the productivity of the ecosystem and biodeterioration of organic materials [26]. These fungi also possess the ability to accommodate aquatic plants and animals including fish under certain conditions [10].

Table 2 List of Fungal colonies isolated from Sites 1 and 2 of Perungudi Lake, Chennai

Months	Samples	Species	Average CFU (10^2)	Total average CFU/ml
January	Site 1	Yeast Colonies	6.66	6.33
	Site 2	Yeast Colonies	2	2
March	Site 1	Yeast Colonies	7.33	7.33
	Site 2	Yeast Colonies	4.66	4.66
May	Site 1	*Aureobasidium Pullulans*	0.33	12
		Aspergillus nidulans	4.66	
		Aspergillus flavus	6	
		Aspergillus terreus	1	
	Site 2	*Aspergillus Fumigatus*	8.66	12
		Curvularia lunata	1.33	
		Chrysonilia Sitophila	0.33	
		Cladosporium oxysporum	1	
		Aspergillus flavipes	0.33	
		Rhizopus stolonifer	0.33	
July	Site 1	*Aspergillus niger*	1	3.66
		Aspergillus flavus	2.66	
	Site 2	*Aspergillus fumigatus*	2	5.3
		Aspergillus niger	3.33	
September	Site 1	*Penicillium citrinum*	0.33	0.66
		Yeast colony	0.33	
	Site 2	*Acremonium strictum*	0.33	0.66
		Yeast colony	0.33	
November	Site 1	Yeast colony	0.33	0.33
	Site 2	*Acremonium strictum*	0.33	0.33

CONCLUSION

Thus, to conclude that a total of 14 fungal species were isolated and identified from Perungudi Lake, Chennai district of Tamil Nadu, India. The study of biodiversity gives the relationships between organisms and the environment and unravels the mechanisms of adaptation to extreme environmental conditions. Hence the above global and Indian scenario on lakes provides unities for mycologists and bacteriologists to explore fungal diversity and exploits their ecological, medicinal and industrial potential.

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