

Comparative Study of Soil Fungi Between Mahemara Aniket and Chhatagarh of Shivanth River, Chhattisgarh

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ABSTRACT: Soil is major source of mycoflora that contain enormous kind of fungi and appropriate moisture required for the growth. The study area of shivnath river have a abundant source of fungal population. Comparative study of soil fungi of shivnath river represent mainly thermo-tolerant fungi in the month of April. The diverse form of fungal colonies obtained in both regions. Fungal growth occurs at different moisture level according to their growth habitat. The present study shows the five common fungi i.e. *Aspergillus flavus*, *Curvularia lunata*, *Cladosporium ladosporioides*, *Fusarium oxysporium*, *Cunninghamella echinulata*.

KEYWORDS: shivnath river, mycoflora, Thermo-tolerant, diverse, habitat.

I. INTRODUCTION

Water covers 70.9% of the Earth's surface, and is vital for all known forms of life. Less than 0.3% of all freshwater is in rivers, lakes, and the atmosphere, and an even smaller amount of the Earth's freshwater (0.003%) is contained within biological bodies and manufactured products. Water on earth moves continually through the hydrological pressure. Water plays an important role in the world economy, as it functions as a solvent for a wide variety of chemical substances and facilitates industrial cooling and transportation. Approximately 70% of the fresh water used by humans goes to agriculture. Shivnath river originates from the mountain at a height of 625 meters at Panabaras situated in the south western parts of Rajnandgaon and flows towards north east direction. 15 years ago, rights to the 23.5 km stretch of the Shivnath River in Durg district, Chattisgarh In 1998, it gave RWL the rights to supply water to the Borai industrial area for 22 years. Since then, villages on the banks of the river were not allowed to use its water for irrigation, fishing or any other purpose. More than 1000 families in the Durg district depended on the river for their livelihood. 80% used water for irrigation and 20%, for fishing. [17]

II. MATERIALS AND METHODS

(i) Study Area

The study area was selected from two distinct region of shivnath river in durg district region, the first one is mahamara and other chhatagarh. Soil sample collected from nearby bank of river. Soil and Sediment from the surface layer (top 5 cm) were collected aseptically, transferred into sterile polythene bags and were brought to the laboratory, stored in refrigerated conditions for further mycological analysis.

March marks the start of *summers* in Pollachi and it lasts till May. Summer is little bit hotter. Rise of temperature begins from the month of April, it is hot amongst. Temperature ranges between 27°C to 40°C during these *months*. Average temperature of Durg in summers remains around 38°C which is low as compared to summer temperatures in other parts of this state. The maximum rise of temperature one can experience is 40°C.

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(ii) Culture Media Preparation:

The following media were prepared and used in these studies.

I. Sabouraud's Dextrose Agar medium (SDA)

Dextrose (C ₆ H ₁₂ O ₆)	20gm
Peptone :	10gm
Agar-Agar :	20gm
Distilled Water :	1000ml

The specified quantity of the constituents were dissolved in one liter of distilled water and heated till agar was melted. The medium was poured in conical flasks (250ml) and were sterilized at 121°C at 15 psi pressure in an autoclave for about for 20 minutes. After autoclaving the flasks, containing media were allowed to cool until the flasks could be hold by hand. The medium was then poured into the already sterilized Petri dishes and culture tubes under sterilized conditions in laminar air flow and then allowed to solidify.

II. Potato Dextrose Agar medium (PDA) (Aneja, 1996):

Peeled potato:	200gm
Dextrose:	20gm
Agar agar :	20gm
Distilled water:	1000ml

Five hundred ml of water was taken in one liter capacity beaker and 200gm washed peeled and sliced potatoes were added to the beaker. Potatoes were boiled gently for 30 minutes or by the time till they are easily penetrated by a glass rod. Boiled potatoes were filtered through muslin cloth and squeezed out all the liquid.

In another beaker, 500ml of water was taken and heated, to which 20gm agar was added bit by bit to get it dissolved followed by addition of 20gm of dextrose. Potato extract was mixed with agar and dextrose and water was added to make volume up to 1000ml. The whole mixture was stirred gently to allow the proper dissolution of agar and dextrose. A pinch of streptomycin sulphate was added to the flasks containing medium to avoid bacterial contamination.

The PDA medium was poured into five conical flasks each of 200ml capacity. The flasks were plugged with cotton and wrapped with aluminum foil. Conical flasks with culture media were sterilized at 121°C at 15 psi pressure in an autoclave for about 20 minutes.

(iii) Identification.

Fungi were identified with the help of various monographs, reviews and other relevant research papers[1,2&6]. Help regarding the identification of these fungi were also taken from mycologist P.N. Chowdhary of National for Central Fungal Identification New Delhi.[3]

III. RESULTS AND DISCUSSION

40 fungal species are recorded during present investigation which are belonging to Zygomycetous (*Absidia cylindrospora*, *Mucor racemosus*, *Rhizopus stolonifer*), Duteromycetes (*Alternaria brassicola*, *Alternaria alternata*, *Aspergillus flavus*, *Aspergillus fumigaus*, *Aspergillus niger*, *Aspergillus oryzae*, *Cladosporium ladosporioides*, *Drechslera australiensis*, *Drechslera hawaiiensis*, *Drechslera specifier*, *Epicoccum purpurascence*, *Eurotium amestaldomi*, *Paecilomyces varioti*, *Penicillium chrysogenum*, *Penicillium citrinum*, *Penicillium oxalicum*, *Rhizoctonia bataticola*, *Trichoderma viride*, *Trichoderma harzianum*, *Trichothecium roseum*), Basidiomycetes (*Acremonium strictum*, *Fusarium oxysporium*, *Fusarium moniliforme*, *Fusarium solani*), and Ascomycetes (*Chaetomium globosum*, *Chaetomium indicum*, *Chaetomium funicola*).

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Table: Comparison of Soil Fungi According to Fungal Isolates
The table comprises presence and absence of various numbers of fungal populations in mehmara and chhatagarh region of Shivnath river

S.No.	Name Of The Fungi Isolated	Shivnath River	
		Mehmara	Chhatagarh
1.	<i>Absidia cylindrospora</i>	+ve	-ve
2.	<i>Acremonium strictum</i>	-ve	+ve
3.	<i>Alternaria alternata</i>	-ve	+ve
4.	<i>Alternaria brassicola</i>	-ve	+ve
5.	<i>Aspergillus flavus</i>	+ve	+ve
6.	<i>Aspergillus fumigaus</i>	-ve	+ve
7.	<i>Aspergillus niger</i>	+ve	-ve
8.	<i>Aspergillus oryzae.</i>	-ve	+ve
9.	<i>Chaetomium funicola</i>	-ve	+ve
10.	<i>Chaetomium globosum</i>	-ve	+ve
11.	<i>Chaetomium indicum</i>	-ve	+ve
12.	<i>Chaetomium spiralis</i>	-ve	+ve
13.	<i>Cladosporium cladosporioides</i>	+ve	+ve
14.	<i>Cladosporium sphaerospermum.</i>	+ve	-ve
15.	<i>Cunninghamella echinulata</i>	+ve	+ve
16.	<i>Curvularia lunata</i>	+ve	+ve
17.	<i>Drechslera australiensis</i>	-ve	+ve
18.	<i>Drechslera hawaiiensis</i>	-ve	+ve
19.	<i>Drechslera specifier</i>	-ve	+ve
20.	<i>Emericella nidulanse</i>	-ve	+ve
21.	<i>Epicoccum purpurascence</i>	-ve	+ve
22.	<i>Eurotium amestaldomi</i>	-ve	+ve
23.	<i>Fusarium moniliforme</i>	+ve	-ve
24.	<i>Fusarium oxysporium</i>	+ve	+ve
25.	<i>Fusarium solani,</i>	+ve	-ve
26.	<i>Mucor mucedo</i>	+ve	-ve
27.	<i>Mucor racemosus</i>	+ve	-ve
28.	<i>Paecilomyces varioti</i>	+ve	-ve
29.	<i>Penicillium chrysogenum</i>	+ve	-ve
30.	<i>Penicillium citrinum</i>	+ve	-ve
31.	<i>Penicillium oxalicum</i>	+ve	-ve
32.	<i>Phoma sorghina</i>	+ve	-ve
33.	<i>Rhizoctonia bataticola</i>	+ve	-ve
34.	<i>Rhizoctonia solani</i>	+ve	-ve
35.	<i>Rhizopus stolonifer</i>	+ve	-ve
36.	<i>Stachbotrys atra</i>	+ve	-ve
37.	<i>Theilavia terricola</i>	+ve	-ve
38.	<i>Trichoderma harzianum</i>	+ve	-ve
39.	<i>Trichoderma viride</i>	+ve	-ve
40.	<i>Trichothecium roseum</i>	+ve	-ve

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Table: Total Number of Common Fungal Isolates found among forty species of fungi in Mehmara and Chhatagarh region of shivnath river

S.No.	Name of Fungi	No. of Colonies	
		Mehmara	Chhatagarh
1.	<i>Aspergillus flavus</i>	14	16
2.	<i>Curvularia lunata</i>	09	12
3.	<i>Cladosporium cladosporioides</i>	17	14
4.	<i>Fusarium oxysporium</i>	14	17
5.	<i>Cunninghamella echinulata</i>	12	13

Aspergillus flavus

Colonies fast growing, mycelium mostly submerged, heavily sporulating, conidial heads yellow to green becoming brownish in age, radiate, splitting into several loose columns, reverse uncolored to pink. Conidiophores usually less than one mm in length, 10-20 μm diam., heavy walled, colorless, coarsely roughened. Vesicles were globose to subglobose, 25-45 μm in diam. Sterigmata were unit and biseriate, both conditions may occur even in the same head. Primaries were 6-10 x 4.0-5.5 μm . Secondary was 6.5-10 x 3-3 μm . In uniseriate condition was 6.5-14 x 3.0-5.5 μm . Conidia were globose to subglobose, minutely echinulate, 3.5- 4.5 μm . Sclerotia white at first becoming red brown or black. *Aspergillus flavus* has the capacity to grow on many nutrient sources. It is mainly a saprophyte and grows on dead plant and animal tissue in the soil, which is important for the recycling of nutrients. Because it grows well on decaying vegetation, one common place you might find *Aspergillus* is in compost or on fallen leaves. However, this fungus is also a parasite and can use living organisms as hosts. Host species include corn, peanuts, nut trees, cotton, and even humans. Typically this organism favors hot, dry conditions which makes it unique in comparison to other fungi. The best temperature for growth is 37°C (98.6°F). However, it can grow at any temperature between 12-48°C (54-118°F). *Aspergillus flavus* is omnipresent in nature but has a more limited growth indoors. *Aspergillus* molds thrive best in oxygen-rich environments and also on materials that are rich in carbon which they feed off of for nutrients. This organism favors hot, dry conditions which makes it unique in comparison to other fungi. The best temperature for growth is 37°C (98.6°F). However, it can grow at any temperature between 12-48°C (54-118°F). *Aspergillus flavus* is omnipresent in nature but has a more limited growth indoors. *Aspergillus* molds thrive best in oxygen-rich environments and also on materials that are rich in carbon which they feed off of for nutrients. *Aspergillus* can often be found growing inside dirty air conditioners inside of buildings or in your own home. This has even been known to be a problem in some hospitals. *Aspergillus* may also grow on or inside walls in homes, especially if the house is damp or has been damaged by flooding. [4 & 19]

Curvularia lunata

Colonies are fast growing, suede-like to downy, brown to blackish brown with a black reverse. Conidia are pale brown, with three or more transverse septa (phragmoconidia) and are formed apically through a pore (poroconidia) in a sympodially elongating geniculate conidiophore similar to *Drechslera*. Conidia are cylindrical or slightly curved, with one of the central cells being larger and darker. Germination is bipolar and some species may have a prominent hilum. *Curvularia* is a hyphomycete found in plants and soil of tropical and temperate countries. It is associated with open globe injuries and is a common cause of secondary fungal keratitis after diagnostic aspiration [11&15]. To date, however, only five known reports of endophthalmitis caused by *Curvularia spp.* have been documented [11 & 16].

Cladosporium cladosporioides

Colonies effuse, olive green or olivaceous brown, velvety; Conidiophores macronematous and micronematous, sometimes 150 μm long but generally much shorter, 2-6 μm thick, pale to mid olivaceous, smooth to verruculose. Ramoconidia were 0-1 septate, up to 30 μm long, smooth or occasionally minutely verruculose. Conidia formed in branched chains, mostly 0-septate, ellipsoidal or limoniform, 3-11 x 2-5 μm size, pale olivaceous brown. This fungus is xerophilic – growing well in low water activity environments (e.g., $a_w = 0.86-0.88$).[5] This species is also psychrophilic, it can grow at temperatures between -10 and -3 °C (14 and 27 °F).[9] *Cladosporium cladosporioides* occurs outdoor environments year-round with peak spore concentration in the air occurring in summer

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where levels can range from 2,000 spores up to 50,000 spores per cubic meter of air.[13] It is among the most common of all outdoor airborne fungi,[13] colonizing plant materials and soil.[9] Due to its tolerance of lower temperatures, *C. cladosporioides* can grow on refrigerated foods and colonize moist surfaces in refrigerators.[13]

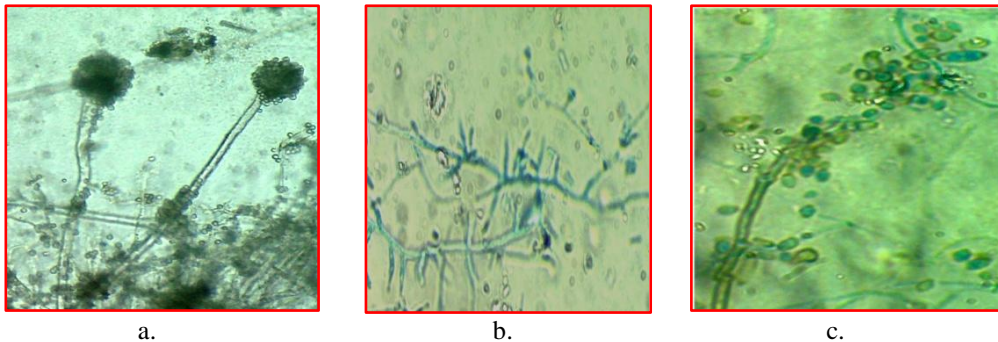
Fusarium oxysporum

Colony grew fast. Conidiophores consist of simple phialides, arising laterally on the hyphae or short branched conidiophores. Phialides developed as monophialidic from primary conidiophores, sub cylindrical while from secondary conidiophores, variable in shape, cylindrical, subcylindric to slightly obclavate and usually longer. Macro conidia were 3-5 septate, 27-60 x 3-5 μm size, thin walled, fusoid, pointed ends, occasionally falcate with terminal cell, hooked and pedicellate basal cell or pointed ends. The micro conidia observed abundant, oval-ellipsoidal, cylindrical, straight or curved, 5-12 x 2-3.5 μm in size, never forming chain. Chlamydo spores, terminal or intercalary, rough to smooth. *Fusarium oxysporum* is a highly ubiquitous, anamorphic fungi that infects a wide range of hosts causing various diseases such as vascular wilt, yellows, corm rot, root rot, and damping-off.[8 & 18] It is an abundant and active saprophyte in soil and organic matter, with some specific forms that are pathogenic to vegetables, flowers, and field crops.[7 & 8]

A few workers have reported growth studies in this genus in submerged culture.[12 & 18] Also, the changes induced by the fungus in the culture such as change in pH and the pattern of utilization of the substrate have also not been worked upon. Therefore, the present investigation was conducted to study the pattern of changes on the growth of the fungus for its utilization in diverse fields.

Cunninghamella echinulata

Cunninghamella is a filamentous fungus found in soil and plant material, particularly at Mediterranean and subtropical zones. It has also been recovered from animal material, cheese, and Brazil nuts. In addition to being a common contaminant, *Cunninghamella* is an opportunistic fungus. *Cunninghamella* colonies are rapidly growing (mature in 4 days), cottony, and white to tannish-gray in color.



- a. *Aspergillus flavus*- *Aspergillus flavus* is a saprotrophic fungi. It is best known for its colonisation of cereal grains, legumes, and tree nuts.
- b. *Fusarium oxysporium* – *Fusarium oxysporum* strains are ubiquitous soil inhabitants that have the ability to exist as saprophytes, and degrade lignin and complex carbohydrates associated with soil debris.
- c. *Cladosporium cladosporioides*-- *Cladosporium cladosporioides* is a darkly pigmented mold. Asexual spores in delicate, branched chains that break apart readily and drift in the air.

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IV. CONCLUSION

Shivnath river is the largest river of Chhattisgarh, contains various fungal population in distinct location. In the present study two distinct region of shivnath river was selected i.e. Mahamara Aniket and Chhatagarh. The isolation of fungal colonies in Sabouraud Dextrose Agar Medium (SDA) and Potato Dextrose Agar Medium (PDA). Total of 40 fungi were recorded from two sources through river soil, while the most common are 5 fungi i.e. *Aspergillus flavus*, *Curvularia lunata*, *Cladosporium ladosporioides*, *Fusarium oxysporium*, *Cunninghamella echinulata*. *Fusarium oxysporium* appears to be largely cosmopolitan. *Aspergillus flavus* is best known for its colonization of starchy cereal grains. They are thermo-tolerant and survive at high temperature and humid climate. *Cunninghamella* species are very fast growing fungi at first becoming rather dark grey and powdery. This is a common contaminant and recovered from animal materials. *Curvularia* are found in tropical region, in the month of April humidity and temperature became high that influences the occurrence of *Curvularia lunata*. *Cladosporium* is a parasitic fungus and are commonly found in dead animals and plants.

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